Effect of Maternal Immunomodulation on Colostral Specific Antibodies and Their Transfer in Buffaloes Neonates Against Foot and Mouth and Rinderpest Vaccines

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Abstract: The colostral and seral antibody titre was significantly higher in levamisole treated group of buffaloes. Other immunopotentiator group differed non significantly. Levamisole hydrochloride can be used effectively along with vaccine in pregnant buffaloes to produce colostral specific antibodies.

Key words: Immunomodulation, buffaloes, antibody titres, levamisole, vitamin E-selenium, Bacille Calmette Guérine

Introduction
During first month of life neonatal mortalities has been reported 39.8% (Afzal et al., 1983) reducing the availability of replacement heifers. Buffalo young calves manifest high susceptibility and subsequent mortality to foot and mouth (Afzal et al., 1983) and rinderpest disease (Chaudhry et al., 1993). 47% incidence of rinderpest in calves has been reported (Rostler et al., 1998). For protection against pathogens, newborn bovine calves depend upon passively acquired immunity obtained through colostrum ingestion. Under certain conditions the immunoglobulin do not attain the required level in the colostrum which exposes the neonate to mortality.

Immunoprophylaxis in the neonates by the use of maternal vaccination procedure alone or in combination with immunopotention to produce lactogenic immunity or colostrum specific immunity. No previous report indicating the use of immunopotentiators along with vaccination during the late gestation on production of colostrum specific antibodies and their transfer to neonates in buffaloes especially under environmental conditions of Pakistan could be available to author.

The evaluation of efficacy of levamisole hydrochloride, vitamin E-selenium and bacille calmette guérine as an immunomodulator in combination with foot and mouth and rinderpest vaccines on the production of colostrum specific antibodies and their transfer to buffalo neonates is underconcentration here.

Materials and Methods
Experimental animals and treatment schedule: A total of 40 pregnant, clinically healthy buffaloes at Livestock Production Research Institute, Bahadarmagar, Okara, Pakistan were studied. They were kept under the similar feeding, housing and managemental conditions. Buffaloes were divided into five groups of eight animals each. Animals from group I were neither vaccinated nor treated with any immunopotentiator and served as un-vaccinated control. Animals in group II were vaccinated against foot and mouth disease vaccine, 6 ml/animal, subcutaneously and tissue culture rinderpest vaccine (1 ml/animal), subcutaneously. Vaccines were procured from Veterinary Research Institute, Lahore, Pakistan and were given 10 wks prior to the expected date of parturition. The sensitizing and booster doses of vaccines were given 7 days apart. Animals in group II, IV and V were vaccinated as group II, additionally, levamisole hydrochloride @ 0.6 mg/kg b.w. orally, vit E-selenium (Etoxol-SE, Boxtel, Holland), 10 ml and bacillus calmette guérine (BCG, Pasteur Merieux, Lyon, France), 0.6 ml/animal subcutaneously were used, respectively, as an immuno-modulators. The immunomodulation treatment was given 7 days before and along with sensitizing dose of experimental vaccines.

Sampling protocol: For colostral antibody titres about 50 ml of colostrum (first milking) from each experimental buffalo was collected immediately after parturition, later the colostrum samples were collected at 12h interval upto 36th following parturition and were stored at 20°C. The first serum sample from calves born to buffaloes of all experimental groups was collected prior to colostrum feeding (0 hr) and then at an interval of 6hr after colostrum feeding upto 36th. Later the serum samples were collected fortnightly until day 84 of age. All the serum samples were stored at -20°C.

Measurement of antibody titres: Antibody titres in colostrum and serum samples against FMD serotypes A and O and rinderpest were measured by indirect hemagglutination test. For preparation of antigen, the cell culture fluid of FMD serotype A and O grown on BHK cell line was procured from veterinary research Institute, Lahore, Pakistan. After harvesting the suspension was centrifuged at 10000 g for 15 minutes to remove cell debris. The supernatant was ultracentrifuged at 48000 g for 30 minutes preparative ultracentrifuge. The Pellets suspended in phosphate buffered saline and sonicated served as antigen. For preparation of Rinderpest antigen the tissue culture fluid containing the vacval strain of rinderpest virus harvested on bovine kidney cell culture was procured from the Veterinary Research Institute, Lahore, Pakistan. Other procedure is the same as described for FMD antigen. For preparation of antigen sensitized erythrocytes method described by Tokuda and Warrington (1970) was used. The final suspension of sensitized erythrocytes was 1.5 and 2%, respectively for FMD and Rinderpest antigens.

Titre micro titration plates (Flow Lab. U.K.), containing 8 rows (A to H) and wells were used to measure the antibody titres. Six samples were titrated row wise at a time, leaving the last 2 rows of wells for positive and negative controls. All samples were serially diluted as 1:2 through 1:2048. The geometric mean titre were also calculated (Thrusfield, 1988). The statistical differences between treatments were calculated by the analysis of variance using Completely Randomized Design (Steel and Torrie, 1980). The geometric mean titre (GMT) against FMD serotypes A and O and Rinderpest in colostrum of all experimental buffaloes are depicted in Fig. 1. The GMT against FMD type A and O (Fig. 1 a,b) was significantly higher in the colostrum of levamisole treated buffaloes at 0 and 12 hrs as compared
Fig. 1: Colostral geometric mean titres in experimental buffaloes (a) against FMD serotype A (n = 40) (b) against FMD serotype O (n = 40) (c) against Rinderpest vaccine (n = 40)

Fig. 2: Serum antibody titres in calves born to all experimental buffaloes (a) against FMD serotype A (n = 40) (b) against FMD serotype O (n = 40) (c) against Rinderpest vaccine (n = 40)

to rest of four groups. Although the vaccinated only and vaccinated + vit E-Se and vaccinated + ECG treated buffaloes showed numerically higher GMT at this stage but statistically the difference was non significant. While no statistical difference was observed at 24 and 36 hr in the colostrum of all experimental buffaloes. Colostral geometric mean titre against Rinderpest was significantly higher at 0, 12 and 24 hrs in levamisole treated group of buffaloes compared to rest of all groups. At this stage all other treated groups differed non significantly (Fig. 1 c). The calf serum antibody titres against FMD type A, O and Rinderpest vaccines are depicted in Fig. 2. Before colostrum feeding all calves had non significant level of antibodies against experimental antigens. After colostrum feeding the antibody titre in all experimental calves rose. From 0hrs following colostrum feeding to 14 days of age the serum antibody titre against FMD serotype A and O (Fig. 2a, b) and Rinderpest (Fig. 2c) vaccine was significantly higher in calves born to levamisole treated buffaloes as compared to rest of all groups. All other treatment groups differed non significantly. From 14 days to culmination of experiment (day 84) no significant difference was observed among all the treated groups. But all differed significantly with that of calves born to un-vaccinated control group buffaloes.

Discussion
Calves of most farm animal species including those of buffaloes are born essentially agammaglobulinemic since transplacental transfer of immunity from the dam does not occur. Hence the protection against pathogens is provided by transfer of immunoglobulins from the dam via colostrum (Lablanct, 1988). The two prime determinants of adequacy of transfer are the level of immunoglobulins and amount of colostrum fed (Noeck et al., 1984). Quality of colostrum is dependent upon an immunological experience of the dam. The gut-mesenteric-gut link of immunological experience from mother to suckling offspring is an important factor for postnatal health and appropriate preparation for weaning (Peña, 1984). During immediate postnatal period there is a critical relationship between the immunoglobulins acquired from colostrum in the plasma of calves and their susceptibility to infections (Singhvi and Anand, 1986). Specific resistance of neonatal calves can be enhanced by vaccination of the dams.
before parturition to stimulate the production of specific antibodies in the colostrum (Spire, 1982). But high level of progesterone during gestation and high level of estrogens and corticosteroids around calving suppress the immune status of pregnant mothers (Staples, 1983 and Tizard, 1987). A reliable immunity may be attained by potentiating the immune response to vaccines using immunopotentiating agents (Babik et al., 1982).

Vaccination of pregnant mothers against all experimental antigens 10 days prior to the expected date of parturition in combination with levamisole hydrochloride as an immunomodulation resulted in significantly higher colostral antibody titres as compared to other treated groups. These results are in agreement with that of Flesch (1982), who reported higher calf survival rate by immunopotentiating the pregnant cows with levamisole hydrochloride in late gestation.

In cattle experiments involving levamisole as an immunopotentiator with infectious bovine rhinotracheitis and herpes virus also respond favourably (Kaneena, 1981). The most favourable results have been obtained in the prevention of neonatal diseases, morbidity and mortality in new born calves after treatment of pregnant cows with levamisole (Flesch, 1977). The exact mechanism by which levamisole hydrochloride potentiate the immune function is not known but it has been shown to increase the chemotactic responsiveness of polymorphonuclear cells and monocytes. Migration inhibition and lymphokine production in response to antigen stimulation could also be restored (Olson, 1976). The gradual decline in colostral antibody titre in all experimental buffaloes is also corroborated with the findings of Afaq (1982).

Unsuckled neonates possess extremely low level of immunoglobulins in their sera, as observed in this study, because of syndesmochorial placentation (Tizard, 1982). These results corroborate the earlier studies of the total immunoglobulins in buffaloes (Awall, 1984). Vit E-selenium and BCG treated buffaloes did not show higher colostral (pregnant mothers) and serum (calves born to treated buffaloes) antibody titres. Vit E and selenium increase hormonal and cellular immunity (Mulhern, 1986). Most of the workers have agreed that both vit E and selenium stimulate immune response in vit E-selenium deficient animals (Stabal, 1991).

BCG treated group showed very irregular pattern of antibody production throughout the experimental period. BCG treatment suppressed the antibody production instead of enhancing it in buffaloes and against the vaccines used in this study. BCG has been reported to enhance host resistance and immune response (Weiss, 1980) but most of previous work was concluded on laboratory animals. Species difference and presence of more than one antigen in conjunction with BCG resulted in immunosuppression. Moreover, the dose of BCG used (0.5ml) was arbitrarily selected which might have reached immunosuppressant levels. Levamisole hydrochloride is an effective immunopotentiator in buffaloes and can be used along with vaccines in the last trimester of pregnancy to produce specific colostrum. But whether it provides the protective level, awaits further study.

References


