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Diet Aloe Supplementation in Pregnant Buffalo Cows Improves Colostrum Immunoglobulin Content

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ABSTRACT

Colostrum ingestion in ruminants is the one way to get maternal antibodies. Calves start suckling soon after birth, and the absorption of immunoglobulin (Ig) lasts up to 24 hours. Many diseases of neonatal calves are related to a poor colostrum quality, which is determined by the content of IgG. The polysaccharide fractions of Aloe has been reported as potent B cell stimulators either in vitro or in vivo studies. Aim of the present research was to evaluate the influence of Aloe arborescence fed to pregnant dry buffalo cows on IgG concentration of colostrum. Twenty-four pluriparae buffalo cows were divided into two homogeneous groups during the last two months of pregnancy. Group A received a supplementation of 50g/day/head of a commercial product containing Aloe arborescence, while group B was the control. Within one hour from calving, samples of colostrum were collected from each subject and underwent IgG assay. A significant ($P<0.05$) increase of colostrum IgG concentration was detected in colostrum from dams supplemented with Aloe. Results showed that Aloe supplementation can increase the immunological properties of colostrum thus resulting in improving passive transfer in newborn calves.

Keywords: aloe, buffalo, colostrum

INTRODUCTION

Buffalo calves depend on the passive transfer of colostral IgG to provide humoral immunity during the neonatal period[1] and adequate passive transfer of immunity, determined by measurement of serum IgG concentration, is a critical determinant of short-term health and survival for neonatal calves[2]. To ensure adequate passive transfer of immunity, neonatal buffalos should receive good quality colostrum in an amount equivalent to 5% to 7% of their body weight divided into 4 to 6 feedings of equal proportions, preferably within 3 to 12 hours after birth. Failure to ingest and/or absorb sufficient colostral IgG, termed FPT, is a secondary immunodeficiency condition that has been linked to increased risk of illness and death from bacterial septicemia and common neonatal infectious diseases and is well recognized among ruminant species. Calves with FPT have an increased risk of illness and death until at least 6 to 7 weeks of age. The influence of management practices on passive transfer of immunity is well established for several ruminants. Administration of colostrum substitutes has had limited success in terms of calf survival rate. Increases in neonatal morbidity and mortality rates are well accepted consequences of FPT among juvenile ruminants, both before and after weaning[3]. Nevertheless, relatively little has been done in the recent years to identify possible feeding supplies that may be able to improve passive immunity in calves and, additional, very little has been done concerning buffalo, despite newborn mortality has been reported to be very high in such species. In such contest, the genus Aloe plant, whose four species

namely, *Aloe barbadensis Miller* (syn. *Aloe vera*; Liliaceae), *Aloe ferox* (syn. *Cape Aloe*; Liliaceae), *Aloe arborescens* (syn. *Candelabra Aloe*; Liliaceae) and *Aloe perryi baker* (syn. *Perry's Alo*; Liliaceae) have been traditionally applied for the medicinal practice over thousands of years in many cultures of the world, could play an interesting rule. Indeed, the polysaccharide fractions of Aloe have been reported as potent B cell stimulators either in vitro[4] and in vivo[5] studies. This research aimed to explore the influence of supplying the diet of pregnant buffalo cows with *Aloe arborescens* on the colostrum immunoglobulin content.

MATERIALS AND METHODS

Twenty-four Italian Mediterranean pluriparae buffalo cows were divided into two groups (homogeneous for parity, body condition score and milk yield in the previous lactations) during the last two months of pregnancy. Both groups were fed a diet (12% crude proteins/dry matter; 6.0 MJ/kg dry matter) constituted by oat straw, corn silage and concentrate. Group A received a supplementation of 50g/day/head of a commercial product (*VigoorsanÒ - NUTRIZOO sas, Italy*) containing *Aloe arborescence*, while group B was the control. Within one hour from calving, samples of colostrum were collected from each subject and divided into two aliquots. The first was analyzed for fat, protein and lactose (Milko Scan 133B, Foss Matic, Hillerod, Denmark, calibrated with an appropriate standard for buffalo milk). The second aliquot of colostrum was centrifuged at 4000 rpm to remove fat and, then, ultracentrifuged at 30000 rpm, the intermediate layer was used from IgG assay. Serum IgG concentration was determined by use of a commercially available radial immunodiffusion according to the manufacturer's specifications (Bethyl Laboratories, Montgomery, USA). Briefly, 5 iL of serum was added to 1 well of a 48-well plate containing anti-bovine IgG antiserum dissolved in 1.5% agarose in tris-buffered saline solution and 0.1% sodium azide. Three reference standards (1.25, 5, and 10 mg/mL) included in the kit were tested concurrently with each sample, therefore, all samples were diluted 1:10. The plate was incubated at room temperature (23°C) for 24 hours, and the precipiting ring diameter was measured. The IgG concentration of test samples was determined by comparing precipiting ring diameter for test samples to a semilog plot generated from results for the reference standards. One-way ANOVA was used to detect statistical differences between groups.

RESULTS AND DISCUSSION

Chemical composition and immunoglobulin concentration of colostrum collected from groups A and B is reported in table 1. Buffalo colostrum was rich in fat and proteins (especially IgG) in agreement with our previous results[6], however, no significant differences were registered between groups, even if proteins were higher for group A, fed diet supplemented with *Aloe arborescens*.

Table 1. Chemical composition (%) and immunoglobulin concentration (IgG, mg/ml) of colostrum.

	Fat	Proteins	Lactose	IgG
Group A	15.4 ± 0.7	14.2 ± 0.9	3.1 ± 0.2	78.54a ± 8.3
Group B	15.4 ± 0.5	13.7 ± 0.8	3.0 ± 0.8	71.28b ±9.0

a,b: P<0.05

Concerning immunoglobulin concentration, animals of both groups included in the trial produced a good quality colostrums; indeed, value of 60 mg/ml registered within one hour from calving is considered a threshold for discriminate scarce or sufficient concentration[7]. Despite that, colostrum of group A showed a significant (P<0.05) IgG increase (78.54 ± 8.3 vs 71.28 ± 9.0 mg/ml); such results suggest that *Aloe arborescens* supplementation may increase the immunological properties of colostrum thus resulting in improving passive transfer in newborn calves. If true, it is supposable that, in case of dams producing a medium or low quality colostrum, the Aloe supplementation may increase

its immunological properties thus reaching a quality still acceptable to ensure passive transfer in calves. As a whole, present results were in agreement with those of our previous study[7]. Other authors[8] reported lower IgG concentration (54.0 mg/ml) in buffalo colostrum; however, the study was carried out on a different buffalo breed (Murrah). The mechanism by which Aloe may lead to such a result needs further studies, however, it has to be underlined that cell surface polysaccharides[9] when recognized by pattern recognition receptors (PRRs) are effective stimuli for the activation of quiescence macrophages and other immune cells[10]. These immunostimulatory substances are generally named polysaccharide biological response modifiers (BRMs). Polysaccharide BRMs are not limited to microbial origin but also to botanical origin. According to the sugar compositions, there are three major groups of polysaccharide BRMs, which are â-1,3-Dglucans, á/â-1,4-mannan and highly branched polysaccharide of very heterogenous monosaccharide compositions. â-1,3-D-Glucans are mainly derived from cell wall or cytoplasmic reserve of fungus[11,12]. á/â-1,4-mannan is mainly derived from yeast cell wall and freshly layer of Aloe leaves[13]. In particular, some authors[4] described the isolation and characterization of the polysaccharide BRM, PAC-I, which was purified from Aloe vera L. var. chinensis (Haw.) Berg., which is a variant of Aloe vera barbadensis Miller and has wide occurrence in China. PAC-I was determined to have mannose as the major monomeric unit. â-1,4-D-Linked mannose contributes to the polysaccharide main skeleton. The molecular weight of PAC-I was 10,000 kDa. PAC-I was demonstrated to exhibit potent stimulatory effects on B and T lymphocytes.

Successively, the same authors[5] reorted that the administration of PAC-I into allogeneic mice stimulated systemic TNF-á production in a dose-dependent manner and prolonged the survival of tumor-bearing mice. PAC-I is thus a potent stimulator of murine macrophage.

In conclusion, our results suggest that Aloe supplementation may improve buffalo colostrum immunological properties thus leading to a higher passive immunization of calves.

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REFERENCES

1. **Barrington GM, Parish SM.** 2002. Ruminant immunodeficiency diseases. In: Smith BP, *Large animal internal medicine.* 3rd ed. St Louis: CV Mosby Co, p.1600-1602.

2. **Constant SB, LeBlanc MM, Klapstein EF.** 2004. Serum immunoglobulin G concentration in goat kids fed colostrum or a colostrum substitute. *J Am Vet Med Assoc*, 205:1759-1762 .

3. **Argüello A, Castro N, Zamorano MJ.** 2004. Passive transfer of immunity in kid goats fed refrigerated and frozen goat colostrum and commercial sheep colostrum. *Small Ruminant Research* 54:237-241.

4. **Leung MY, Liu C, Zhu LF, Hui YZ, Yu B, Fung KP.** 2004. Chemical and biological characterization of a polysaccharide biological response modifier from Aloe vera L. var. chinensis (Haw.) Berg. *Glycobiology* 14:501–10.

5. **Liu C, Leung MYK, Koon JCM, Zhu LF, Hui YZ, Yu B, Fung KP.** 2006. Macrophage activation by polysaccharide biological response modifier isolated from Aloe vera L. var. chinensis (Haw.) Berg. *International Immunopharmacology* 6:1634–1641

6. **Lombardi P, Avallone L, d'Angelo A, Bogin E.** 1996. Glutamyltransferase and serum proteins in buffalo calves following colostral ingestion. *Eur. J. Clin. Chem Clin Biochem.* 34:965-968.

7. **Lombardi P, Avallone L, Pagnini U, d'Angelo D, Bogin E.** 2001. Evaluation of buffalo colostrum quality by estimation of enzyme activity levels. *Journal of Food Protection*, 64:1265-1267.

8. **Dang AK, Kapila S, Purohit M, Singh C.** 2009. Changes in colostrums of Murrah buffaloes after calving. *Trop Anim Health Prod* 41:1213-1217.

9. **Sironi M, Sica A, Riganti F, Licciardello L, Colotta F, Mantovani A.** 1990 Interleukin-6 gene expression and production induced in human monocytes by membrane proteoglycans from Klebsiella pneumoniae. *Int J Immunopharmacol* 12:397–402.

10. **Gordon S.** 2002 Pattern recognition receptors: doubling up for the innate immune response. *Cell*; 111:927–30.

11. **Garner RE, Hudson JA.** 1996 Intravenous injection of Candidaderived mannan results in elevated tumor necrosis factor alpha levels in serum. *Infect Immun*; 64:4561–6.

12. **Ni Y, Turner D, Yates KM, Tizard I.** 2004 Isolation and characterization of structural components of Aloe vera L. leaf pulp. *Int Immunopharmacol*; 4:1745–55.

13. **Nose M, Terawaki K, Oguri K, Ogihara Y, Yoshimatsu K, Shimomura K.** 1998 Activation of macrophages by crude polysaccharide fractions obtained from shoots of Glycyrrhiza glabra and hairy roots of Glycyrrhiza uralensis in vitro. *Biol Pharm Bull*; 21:1110–2.