

TOWARDS A TOOL FOR RETROSPECTIVE ASSESSMENT OF EXPOSURE OF NON-WEANED CALVES TO THE BOVINE RESPIRATORY SYNCYTIAL VIRUS (BRSV)

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Introduction

Respiratory disorders are usually considered as a major health issue in cattle, particularly in young calves [1, 8, 13]. Among the great number of viruses and bacteria potentially involved, the Bovine Respiratory Syncytial Virus (BRSV) appears to be the most common and significant aetiological agent of respiratory disorders [8, 9, 13]. Outbreaks of BRSV infection mostly occur in non-weaned calves less than three months of age [14]. Currently, the most widespread method of indirect detection of BRSV infection relies on detection of total immunoglobulins G (IgG) specific of BRSV by Enzyme Linked Immunosorbent Assay (ELISA). Evidence of an active immune response to BRSV is a specific rise in titer of total IgG from at least 400% between two blood samples three weeks apart [5]. However, taking into account the very strong seroprevalence of specific BRSV antibodies in cows (between 70% and 95%) the frequency of calves with BRSV specific maternally-derived antibody is very high [4, 9]. Maternally-derived antibody suppresses antigen-specific serum antibody response during the first months of life. So, reliance on paired-serological testing to identify BRSV in calves less than 3 months of age is not recommended [15]. The passively transferred antibodies are mainly of the IgG1 isotype, whatever the etiologic agent concerned [4, 6]. Amount of IgG2 is very low in the colostrum [2, 7, 10]. Moreover, if IgG2 seem to appear only 3 weeks after the infection, they remain detectable during more than 80 days [7, 11, 15]. However, the only study dealing with IgG2 specific of BRSV in calves less than 3 months and in spontaneous BRSV natural infections was carried out on only five calves [11]. This study aimed at describing, under field conditions, the level of total IgG and IgG2 specific of BRSV obtained from a large sample of sera from young calves before and after colostrum intake and from their dams.

Materials and methods

Blood samples were collected from young calves of commercial herds located in the south of the area Pays de la Loire by the veterinarians of 3 clinics located near the National Veterinary School of Nantes. On the occasion of a calving or a caesarean, 5 ml of blood was collected into sterile vacutainers by jugular venipuncture of the newborn calf before colostrum intake. Five ml of blood was collected into sterile vacutainers by caudal venipuncture of their dams. Between 3 and 6 days after birth, 5 ml of blood was again collected into sterile vacutainers by jugular venipuncture on the same calves. These calves should have taken an adequate amount of colostrum under supervision of the farmer. They also should have been healthy from birth to the day of this second sampling. Lastly, these calves should not have been vaccinated. Sera were separated by centrifugation and stored at -20°C until required for testing. All the sera were analysed blindly the same day. Finally, 100 paired

sera of calves and the 100 sera of their dams were available for serological testing.

Detection of TG1g and G2Ig was made using commercial ELISA tests (LSI RSV Bovine Serum IgG and LSI RSV Bovine Serum IgG2). The cut-off value proposed by the manufacturer is S/P = 0.2 (for the two kits).

Results

The distribution of the S/P ratio for newborn calves before and after colostrum intake are shown in figure 1. Among the 100 newborn calves, before the intake of colostrum, 98 had a S/P (sample/probe) ratio of G2Ig < 0.04 and 99 had a S/P ratio < 0.08. After the intake of colostrum, the same 99 calves still had a G2Ig S/P ratio < 0.08, but only 6 of these 100 calves had a TG1g S/P ratio < 0.08. Among the 100 dams, 94 had a G2Ig S/P ratio > 0.08 and 97 had a TG1g S/P ratio > 0.08.

Discussion

This study allowed to confirm (i) the absence of detection of Ig G total and Ig G2 specific of BRSV on newborn calves before colostrum intake, (ii) the transmission of total IgG specific of BRSV by colostrum intake of their dams which have high sera level of total IgG and IgG2 specific of BRSV and (iii) lastly, under field conditions on a large sample of newborn calves, the quasi-absence of IgG2 specific of BRSV after colostrum intake. Based on the cut-off values provided for adult cows by the manufacturer the seroprevalence obtained on the dams, 90% of IgG2 and 95% of total IgG specific to BRSV, in Western France, seems to be similar to the high seroprevalences found in adult cattle in other European countries like the United Kingdom [9] and Denmark [11]. According to the syndesmochorial placentation of cattle, the calves were born almost agammaglobulinemic [10]. This step of checking allowed us to estimate the ability of the two ELISA tests used (total IgG and IgG2) to classify all the newborn calves sampled before colostrum intake as non infected. The results provided by the tests fit well with the agammaglobulinemia of newborn calves before colostrum intake. After colostrum intake, the S/P ratio of total IgG and IgG2 specific of BRSV increase (fig 2). Nevertheless when S/P ratio of IgG2 or total IgG specific of BRSV of the dams are high, S/P ratio of IgG2 remain low for their calves. As discussed by Uttenthal et al.[11], the presence of cattle with IgG1 but no IgG2 probably represent newly infected animals or animals with residues of maternally derived antibodies whereas cattle with IgG2 but no IgG1 probably represent convalescent animals. So detection of IgG2 could be used on non-weaned calves in endemic areas of BRSV infection. Concerning the newborn calf having a S/P ratio of IgG2 specific of BRSV of 0.1245, this value remains much higher than the values obtained on other calves. This calf should be considered as an outlier. This high value can be easily explained by a lack of specificity of ELISA tests [15]. Moreover, this calf had the highest S/P ratio of total IgG

specific of BRSV, so a non-lethal infection during the gestation, sometimes suspected but never evidenced, cannot be thrown out [7].

Conclusion

The obtained antibodies profiles are consistent with previous experimental studies [2, 10, 14]. Then, the potential interest of this technique based on the detection of IgG2 specific of BRSV, as a method for the retrospective detection of the circulation of the BRSV for non weaned calves, comes from the quasi-absence of this isotype in the colostrum which would make it possible to detect only a post-infectious immune response of the calves, with only one blood sample contrary to the usual methods. From our results it is possible to propose that a cut-off value adapted to non-weaned calves can be lower than the cut-off value of 0.2 provided (for adult cows) by the manufacturer. Indeed, values around 0.08 comply with two classical methods of cut-off value determination. The first one consists in taking the highest value of a negative population. So, in this situation to propose cut-off values around 0.08. And the second method, consists in taking the average value increase of 2 standard-deviations. So, in this second situation to propose the cut-off value of 0.0732. Then, cut-off values around 0.08 seem to be relevant [3].

References

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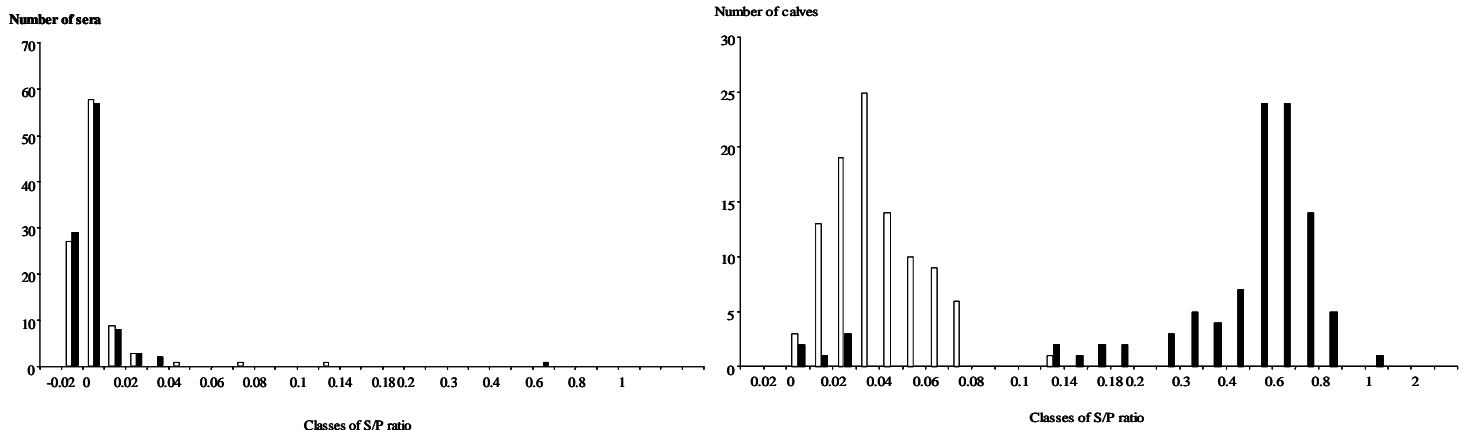


Fig. 1: Number of sera of the newborn calves before (left hand) and after (right hand) colostrum intake according to classes of S/P ratios obtained with the IgG2 test (□) and the total IgG test (■) specific of BRSV.