

#### SHORT NOTE [NOTA CORTA]

# PREVALENCE AND INCIDENCE OF Cryptosporidium spp. IN CALVES FROM THE CENTRAL REGION OF VERACRUZ, MEXICO

[PREVALENCIA E INCIDENCIA DE Cryptosporidium spp. EN BECERROS DE LA ZONA CENTRO DE VERACRUZ, MEXICO]

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## **SUMMARY**

In order to determine the prevalence and incidence of Cryptosporidium spp. in bovine production units (BPU) located in the central area of Veracruz, Mexico, 272 grazing dual-purpose calves from 13 BPU were studied. Fecal samples were collected from each of all the calves, and were examined following the Kinyoun technique. Of the 13 BPU included, in "El Clarin" and "La Soledad" samples were collected every two weeks to determine the incidence of calves positive to Cryptosporidium spp. The Cryptosporidium spp. oocysts were microscopically identified. Overall prevalence was 35.7 % with a confidence interval at 95 % of 30.0 to 41.4 %. No significant differences were found between male and female calves. The prevalence was higher in calves younger than 4 months old (42.9 %) than in calves older than 4 months old (28 %) (P = 0.01). The incidence was 170 and 100 new cases for each 1000 calf-day at risk in "El Clarin" and "La Soledad", respectively. In conclusion, Cryptosporidium is a prevalent parasite in BPU of the central region of Veracruz and it has a high incidence.

**Key words:** *Cryptosporidium*; calves; tropic; prevalence; incidence.

## INTRODUCTION

The members of the genus *Cryptosporidium* are protozoan parasites that colonize the epithelial cells, particularly those found along the digestive tract, in a

#### RESUMEN

Con el fin de determinar la prevalencia e incidencia de Cryptosporidium spp. en unidades de producción bovina (UPB) ubicadas en la zona centro de Veracruz, México, se examinaron 272 becerros de doble propósito mantenidos en pastoreo, pertenecientes a 13 UPB. De todos los becerros se colectaron muestras de heces que se analizaron según la técnica de Kinyoun. De las 13 UPB seleccionadas, en "El Clarín" y "La Soledad" se colectaron muestras cada dos semanas para determinar la incidencia de becerros positivos a Cryptosporidium spp. Los ooquistes del parásito se identificaron por microscopía. La prevalencia general fue 35.7 % con intervalo de confianza al 95 % de 30.0 a 41.4 %. No hubo diferencias significativas entre machos y hembras. La prevalencia fue mayor en becerros menores de cuatro meses de edad (42.9 %) que en mayores de cuatro meses de edad (28 %) (P = 0.01). La incidencia fue 170 y 100 casos nuevos por cada 1000 becerro-día a riesgo en "El Clarin" y "La Soledad", respectivamente. Se concluyó que Cryptosporidium es un parásito prevalente en las UPB del centro de Veracruz y que tiene elevada incidencia.

**Palabras clave:** *Cryptosporidium*; becerros; trópico; prevalencia; incidencia.

wide number of vertebrates, including mammals, birds, fish and reptiles (Fayer *et al.*, 1997).

Taxonomy of *Cryptosporidium* in mammals has been in dispute since 1980, and for some time only two species were considered based on their morphological

and genetic differences, hosts, and sites of infection. Nowadays, 13 species are known in mammals, of which C. parvum, C. andersoni, C. hominis and C. ryanae have been reported in cattle (Xiao et al., 2004; Fayer, 2010). Of these latter species, C. parvum is the most important, since it represents one of the main etiologic agents for neonatal diarrhea in calves and has a great zoonotic potential (Garber et al., 1994; Olson et al., 2004). Infections with Cryptosporidium spp. can have a great impact in immunocompromised mammals (Hunter and Nichols, 2002), and in public health because the oocysts are resistant to the environment and can contaminate different food sources (Karanis et al., 2007). Currently, there are no effective treatments against cryptosporidiosis, so the basic epidemiological information can contribute to the development of strategies of prevention and control of the disease (Romero et al., 2001).

In Mexico, the studies on the prevalence of *Cryptosporidium* spp. in calves are incipient; most of them have been conducted in dairy cattle (Maldonado-Camargo *et al.*, 1998; Castillo *et al.*, 2009), and no literature is available on studies carried out in beef or dual-purpose cattle in tropical regions.

Worldwide, there are no reports available on the incidence of *Cryptosporidium* spp. in calves. Therefore, the objectives of the present study were: 1) to determine the prevalence of *Cryptosporidium* spp. in bovine production units (BPU) located in the central region of the State of Veracruz, Mexico, and 2) to calculate the incidence of *Cryptosporidium* spp. in calves from two of the BPU included in the study.

#### MATERIALS AND METHODS

### Area of study

The study was conducted from May to November 2010, in 13 dual-purpose BPU belonging to the municipalities of Tlapacoyan, Martinez de la Torre, San Rafael and Nautla, all located in the central northern region of the State of Veracruz, Mexico. A total of 272 calves from all the BPU were included in the study. The selection and sampling of the BPU were carried out by convenience (number of calves and facility of the sampling).

## Animals, sampling and laboratory diagnosis

To determine the prevalence within each BPU, from each calf a fecal sample was taken directly from the rectum with a plastic bag previously identified with the number of the animal. After that, the samples were refrigerated and transported to the laboratory.

In the BPU "El Clarin" and "La Soledad" the incidence of *Cryptosporidium* spp. was also calculated (Thrusfield, 2007), since in these BPU the appropriate conditions for the fortnightly handling of the animals and sampling were given. Thus, to calculate the incidence of *Cryptosporidium* spp., fecal samples were collected from the calves every two weeks during four months (June to September) to 18 calves at 1 to 32 days of age at the first sampling in "El Clarin", and during five months (May to September) to 17 calves at 69 to 156 days of age at the first sampling in "La Soledad".

To detect *Cryptosporidium* spp. oocysts, fecal smears were stained according to Kinyoun's technique (Romero *et al.*, 2001) and were directly observed in the microscope at 100X and with an ocular micrometer scale. A calf was considered as positive when at least one oocyst was observed in the three microscope fields of view that were checked.

### Statistical analysis

In each BPU evaluated, the prevalence of calves positive to Cryptosporidium spp. was calculated by dividing the number of positive cases by the total of calves evaluated and multiplying by 100 (Thrusfield, 2007). The incidence was calculated according to the formula by Thrusfield (2007) to calculate the true incidence, and it was done by dividing the number of new cases of calves positive to *Cryptosporidium* spp. by the number of calf-day at risk. The results of incidence were extrapolated to a hypothetical population of 1000 calf-day at risk (Thrusfield, 2007). The Chi-square test was used to evaluate the effect of sex and age of the calves on the infection by Cryptosporidium spp. (Epi Info v. 6). A P < 0.05 value was used to determine the existence of statistically significant differences. The descriptive statistics, such as frequencies and proportions, was calculated using the Excel program.

#### RESULTS

Calves infected with *Cryptosporidium* spp. were detected in all the 13 BPU studied. Overall prevalence was 35.7 % with a confidence interval (CI) at 95% of 30 % to 41.4 %, ranging from 18.8 % in the BPU La Playa to 63 % in the BPU San Valentin (Table 1). Data of prevalence for "El Clarin" and "La Soledad" correspond to the first sampling carried out in these BPU.

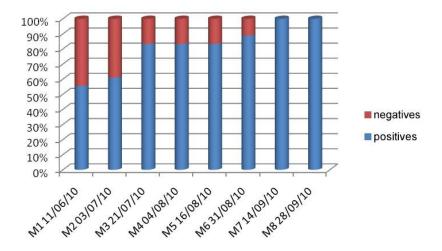
**Table 1.** Prevalence of *Cryptosporidium* spp. in calves from bovine production units (BPU) of the central region of Veracruz, Mexico.

BPU	Calves sampled	Calves positive to oocysts	Prevalence (%)	IC 95%*
Aramburo	30	12	40.0	22.5 - 57.5
Cabellal	30	6	20.0	5.7 - 34.3
El Clarín	18	10	55.6	32.6 - 78.5
Embarcadero	20	8	40.0	18.5 - 61.5
Esperanza	24	7	29.2	11.0 - 47.4
Нс	19	4	21.1	2.7 - 39.4
Manterola	17	4	23.5	3.4 - 43.7
Piedra pinta	20	6	30.0	9.9 - 50.1
La Playa	16	3	18.8	0.0 -37.9
Rancho viejo	17	5	29.4	7.8 - 51.1
San Rafael	17	8	47.1	23.3 - 70.8
San Valentín	27	17	63.0	44.7 - 81.2
La Soledad	17	7	41.2	17.8 - 64.6
Total	272	97	35.7	30 - 41.4

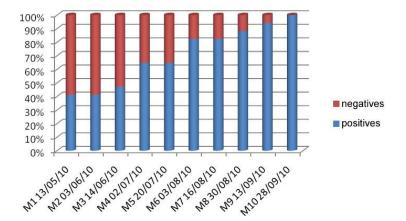
<sup>\*</sup>CI = Confidence interval at 95 %.

Of the 272 calves sampled, 119 (43.7 %) were females and 153 (56.2 %) were males, resulting positive to the presence of *Cryptosporidium* spp. oocysts 30.2 % (IC<sub>95%</sub>: 22.7 - 39) of the females and 39.8 % (IC<sub>95%</sub>: 32.4 - 47.7) of the males (P > 0.05). With respect to the age of the calves, 140 calves younger than 4 months old and 132 calves 4 to 9 month-old were sampled, and the resulting prevalence of *Cryptosporidium* spp. was 42.9 % and 28 %, respectively (P = 0.01).

The number of new cases of calves infected with *Cryptosporidium* spp. increased until reaching 100 % in "El Clarin" and "La Soledad", where the incidence was 170 and 100 new cases for each 1000 calf-day at risk, respectively (Figures 1 and 2).



**Figure 1.** Fortnightly dynamics of new cases of calves infected with *Cryptosporidium* spp. in "El Clarin", Veracruz, Mexico.



**Figure 2.** Fortnightly dynamics of new cases of calves infected with *Cryptosporidium* spp. in "La Soledad", Veracruz, Mexico.

#### **DISCUSSION**

The first objective of the present study was to determine the prevalence of *Cryptosporidium* spp. in BPU located in the central region of Veracruz, Mexico. The overall prevalence found was slightly higher than that reported by Maldonado-Camargo *et al.* (1998) in calves from dairy herds in the central region of Mexico (Hidalgo, Jalisco and State of Mexico), but it was lower than the prevalence observed by Castillo *et al.* (2009) in dairy herds in Aguascalientes, Mexico. The differences in the prevalence among BPU can be due to factors such as the production system, management conditions during calf rearing (individual or group management), breed, age, overcrowding, as well as the hygienic measures (Maldonado-Camargo *et al.*, 1998; Díaz *et al.*, 2004).

As for the relationship between sex and age of the calves with the elimination of Cryptosporidium spp. oocysts, the results of this study were consistent with those reported by other authors (Garber et al., 1994; Atwill et al., 1999; Díaz et. al., 2004). No statistical differences were found among sex of the calves and elimination of oocysts, but there was a difference concerning the age of the calves, being the prevalence higher in calves younger than 4 months old than in older calves. This is particularly important in conditions where lactating calves graze together with weaned calves (older than 4 months old), since calves younger than 2 months old are more susceptible to get the infection and to develop the disease, whereas the older calves or the adults tend to be less receptive and to eliminate fewer oocysts; however, their role as asymptomatic carriers of the parasite must not be

underestimated (Garber et al., 1994; Atwill et al., 1999; Fayer et al., 2000).

The second objective of this study was to calculate the incidence of *Cryptosporidium* spp. in calves from two of the BPU that were selected. The spread velocity of elimination of oocysts in the BPU "El Clarin" and "La Soledad" was 170 and 100 new cases for each 1000 calf-day at risk, respectively. No reports were found on the incidence of *Cryptosporidium* spp. in dual-purpose calves or in calves from temperate climates to compare the results obtained in the present study. However, these results showed that the velocity with which the calves from the two BPU eliminated oocysts was high, and that there was also a major difference in the number of new cases between the two BPU evaluated. It is likely that some management factors of the calves rearing in the two BPU (restricted suckling vs. artificial rearing), breed (3/4 Holstein 1/4 Zebu vs. 1/2 Holstein ½ Zebu), climatic conditions, and grazing system might have influenced on the dynamics of oocysts elimination.

#### **CONCLUSION**

*Cryptosporidium* is a parasite that is highly prevalent in the BPU from the central region of Veracruz, Mexico, and has a high incidence.

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