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DAIRY RESEARCH BULLETIN 4

In vitro inhibition of *Cryptosporidium parvum* sporozoite binding to a susceptible bovine kidney cell line

INTRODUCTION

Cryptosporidium parvum is a protozoa that causes the disease Cryptosporidiosis, or Crypto, in calves. The disease affects calves from birth to six weeks of age and causes watery diarrhea, which can lead to dehydration and even death. Since there are not any treatments or vaccines for Crypto, disease prevention methods are important. In addition to sanitation and reducing calf-to-calf contact to reduce oocysts ingestion by calves, feed additives that can prevent *C. parvum* infection are of interest.

Also, with the increase in interest of natural compounds that can be utilized to prevent and/or treat infections, utilizing yeast cell wall complex carbohydrates from *Saccharomyces cerevisiae*, like those in Naverde, may be an option for producers. This research evaluated the efficacy of yeast cell wall complex carbohydrates as a means of inhibiting *C. parvum* sporozoite binding to susceptible bovine kidney cells.

METHODS

► Two commercially available feed additives were used in this trial. Naverde (hydrolyzed yeast cell wall + hydrolyzed yeast) and a Positive Control (saponin-based product) were used at concentrations of 20, 40, and 60 mg/mL. Their supernatants were incubated with *C. parvum* sporozoites for one hour and then added to susceptible Madin-Darby bovine kidney (MDBK) cells. Next, the plates were incubated for two hours at 37° C. Positive and negative controls were included in the experiment. Following incubation, indirect immune-

fluorescence assay (IFA), including fixation with methanol and staining with specific primary and secondary antibodies, was conducted. A fluorescent microscope was used to count bound sporozoites, and results were reported as percent binding inhibition. There were six replicates per treatment. Percent binding inhibition was calculated by $100 - [(treatment\ average\ count / +\ control\ average\ count) * 100]$.



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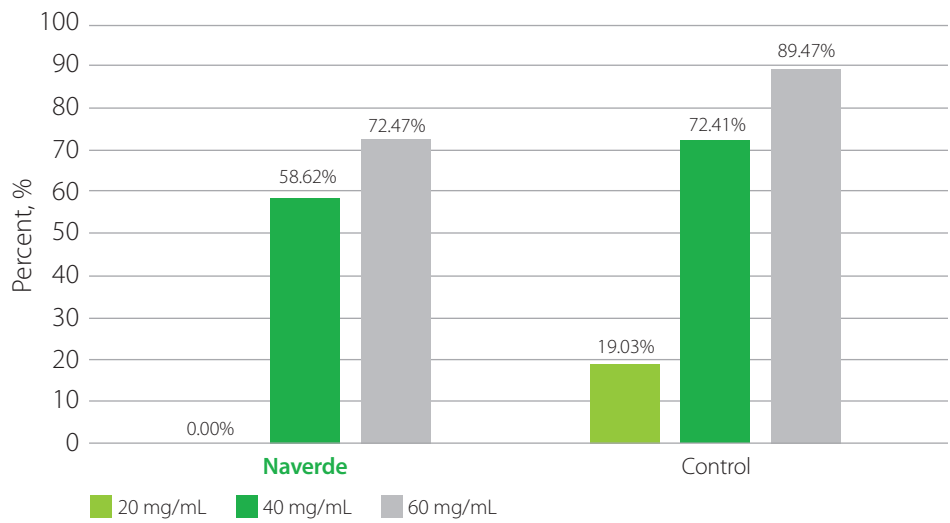
RESULTS

► Naverde and the Control demonstrated binding inhibition of *C. parvum* (Figure 1). The dose-dependent inhibition of binding by Naverde was 58.62% and 72.47% at 40 and 60 mg/mL, respectively. At 20, 40, and 60 mg/mL, the Control demonstrated 19.03%, 72.41%, and 89.47% binding inhibition rate, respectively.

IMPLICATIONS

Since pathogenic *C. parvum* can cause disease and even death in young calves, the industry is interested in natural alternatives that can be utilized to prevent infections. The yeast cell wall complex carbohydrates in Naverde were effective at preventing *C. parvum* sporozoite attachment to susceptible cells in vitro. Thus, evaluating Naverde in feed for calves to prevent Crypto merits consideration.

Figure 1. Percentage binding inhibition of *Cryptosporidium parvum* sporozoites to susceptible cell line in vitro.



P.O. Box 221
Newfield, NY 14867
844.NAT.BIOL (628.2465)
naturalbiologics.com

