A canine parasite dangerous to domestic cattle and bison

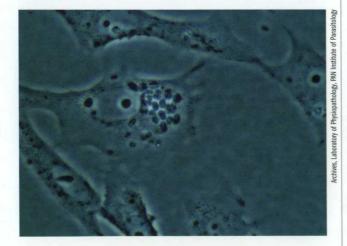
Invisible Danger

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Although this cattle-threatening parasite conceals itself within the cells of its hosts, the routes whereby it spreads can be detected using modern diagnostic techniques

Neospora caninum is a parasitic protozoan whose life cycle requires two hosts: a definitive host, i.e. the dog or coyote, perhaps also the wolf, and an intermediate host, which may be the dog, cow, sheep, horse, goat, deer, European and American bison, cat, monkey, fox, raccoon dog, rat, or lama. The parasite reproduces sexually in the small intestine of its definitive host, after which it enters the outside environment in invasive form (as oocysts), a source of infection for other animals. Once in an intermediate host, oocysts release tachyzoites (forms which penetrate cells), which can then change into bradyzoites (encysted forms). The parasites situate themselves within brain and spinal cord tissue.

In cattle, neosporosis (*Neospora caninum* infection) can take a significant economic toll, causing the loss of



Neospora caninum tachyzoites situate themselves inside the nervous system of host animals – in the brain and spinal cord tissue. However, they can also be kept in laboratory cell culture

calves, a need to renew calving, a longer lactation period, reduced meat quality, and lower milk yield.

Vertically or horizontally

Neosporosis manifests itself in adult cows in terms of reproductive disorders: early embryo death, miscarriages, and the bearing of dead calves. Calves born by infected mothers, if they survive, may continue to be asymptomatic carriers of the parasite for successive generations. Infected calves may also suffer from a lack of physical coordination, paralysis or muscular atrophy in their body section, or convulsions.

The spread of *N. caninum* within a farm herd proceeds in two ways, with 81-95% of cases involving the parasite being transferred "vertically," passed down from one generation to the next. Neosporosis also spreads during unsupervised calf births, via the licking and consumption of the placenta, which contains invasive forms of the parasite (the "horizontal" route). It is conjectured that healthy calves may also become infected when fed milk from infected cows. Fortunately, the parasite definitely does not pose a threat to people, although specific antibodies against *N. caninum* have been detected in individuals who come into contact with infected cattle.

Crucial findings

The Physiopathology Laboratory at the PAN Institute of Parasitology first launched research on *N. caninum* in 2000. We used the ELISA immunological method to study more than 2500 bovine sera. The presence of IgG class antibodies in the tested samples indicated the presence of the parasite. The percentage of infected cows in individual herds ranged from 1.7%-20.3%. Several of the 40 stud-bulls tested were also infected. We likewise showed that the parasite is passed down from one bovine generation to the next (from grandmother to mother to calf – both heifer and bull) with a frequency of more than 95%!

Further research has aimed to resolve a few riddles. First and foremost, we wanted to identify when fetal infection occurs during the course of pregnancy – whether egg cells are already infected with the parasite, or whether infection occurs at earlier or later stages of fetal development. To find out, we used the PCR (polymerase chain reaction) method to detect the presence of *N. caninum* DNA. The brain material of aborted fetuses and of calves born by infected mothers tested positive for

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Neosora caninum infection in adult cattle manifests itself in terms of reproductive disorders: miscarriages or the birth of weakened or dead calves

parasite DNA, yet oocytes and early fetal stages tested negative. These are extremely important findings, indicating that embryo transfer, a widely practiced means of bovine reproduction, does not involve a risk of neosporosis infection. Overall, our findings can be interpreted as supporting one of two conclusions: either the parasite infects embryos at a later stage of development, or the complex procedure of purification embryos before freezing eliminates the parasite from their surface.

We have also identified the presence of *N. caninum* DNA in semen samples taken from infected stud-bulls, indicating the possibility of neosporosis being transmitted sexually. This finding is also particularly important in light of research by Prof. L. Ortega-Mora et al., where cows inseminated with semen containing experimentally added tachyzoites became infected with the parasite.

We were the first research team in the world to identify the presence of parasite DNA in milk and colostrums (the first milk secreted postnatally), indicating that healthy calves may become infected when fed milk from infected cows. We also studied the impact of UV radiation, freezing, boiling, and sterilization on parasite survival, showing that only UV-treated parasites retained their vitality in the cell culture.

Field research has successfully developed a Polish isolate of *N. caninum*. Its tachyzoites are maintained in constant *in vitro* culture in Vero cells (monkey kidney epithelial cells), like those of the Swedish reference strain NC-1. We are using both strains in further immunological and molecular research.

Among bison

As members of the Bison Network, we are also working to study neosporosis among European bison. Of 320 serum samples from a sample bank, 7.4% tested positive, with the earliest positive serum deriving from a bison shot in 1988 – the same year the species *N. caninum* was first time described by Prof. Dubey in the United States. Moreover, as many as 15-17% of the animals shot at present are infected with the protozoan, a fact that poses a threat to the heath status and conservation Poland's bison population.

Our laboratory is Poland's only research center studying *N. caninum* so comprehensively. Research findings have so far been published in 36 original papers, review publications, and conference reports, and have been repeatedly presented at conferences organized within the framework of Cost Action 854, an EU research consortium for studying "Protozoal Reproduction Losses in Farm Ruminants." In 2002 we became members of the Management Committee for Cost Action 854, representing Poland. We have likewise presented our findings at domestic conferences. Unfortunately, there continues to be scarce interest among cattle farmers and veterinary services in the possibility of testing for the parasite within herds and working to eliminate the risk it poses.

Further reading:

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