Cryptosporidiosis
Summary

Introduction

1. This note provides a summary of an analysis prepared by a DISCONTOOLS group of experts on cryptosporidiosis. The group: reviewed current knowledge on the disease, considered the existing disease control tools, identified current gaps in the availability and quality of control tools, and finally made suggestions for the research necessary to develop new or improved tools. Full details are available on the web site at http://www.discontools.eu/.

Disease profile

2. Cryptosporidiosis is caused by infection with protozoan parasites in the genus *Cryptosporidium*. Currently, close to 50 species of *Cryptosporidium* have been described, and over 120 genotypes, the taxonomic status of which requires further clarification. As further genotyping and biological information becomes available, some of these are likely to be re-categorised as species. The most common species causing disease in mammalian livestock is *C. parvum*; both *C. parvum* and *C. hominis* are important human pathogens, with other species, particularly *C. mortiferum* and *C. meleagridis,* being notable human pathogens in specific geographical regions. A wide range of domestic and wild animals are affected by cryptosporidiosis. Those *Cryptosporidium* species infecting non-mammalian host groups are mostly non-zoonotic, with the notable exception of *C. meleagridis*. We have primarily focussed on *Cryptosporidium parvum*.

3. *C. parvum* has a very wide host range and is predominantly a parasite of young hosts, with some degree of immunity rapidly developing. It is responsible for a substantial proportion of both sporadic and outbreak-related cases of human cryptosporidiosis, although occurs less commonly in some low- and middle-income countries, where *C. parvum* identified in people is often of a non-zoonotic subtype.

4. *C. hominis* is morphologically identical to, but genetically distinct from, *C. parvum.*

5. There are good animal disease models for *C. parvum* (neonatal calves and lambs) and infection models (neonatal or SCID mice).

6. The environmental transmission stage of the parasite, the oocysts, are protected by an outer shell that allows them to survive outside the host in moist, cool environments for prolonged periods (>6 months). The oocysts are also very resistant to chlorine-based disinfectants; those based on phenols or hydrogen peroxide seem to be the most effective.

Risk

7. Cryptosporidiosis remains a significant public health threat and an important disease of young livestock. The main symptom is diarrhoea, usually self-limiting, but not infrequently prolonged. Older animals are often infected but are usually asymptomatic; they serve as a source of infective oocysts.

8. Cryptosporidiosis is an important cause of moderate-to-severe diarrhoea. People who have not previously been infected are at higher risk of disease. Although children are most commonly affected, any age group can develop cryptosporidiosis. The risk of disease following infection is highest in the very young, the elderly, and the immunocompromised.

9. Infection is initiated through ingestion of infective oocysts shed by infected animals or people or present in contaminated food/feed or water. Animal husbandry practices in relation to housing, feeding, cleaning and disinfection, and birthing patterns, livestock management, and facilities can all have an impact on the spread of cryptosporidiosis. Inappropriate disposal of waste, manure and faeces can lead to contamination of water courses that may be used by animals or humans. Infection by aspiration and inhalation has been reported to occur.

Diagnostics

10. Infection results in both humoral and cell mediated immunity. Local antibody production in the gastrointestinal tract also occurs. Parasite-specific antibodies are produced. As the disease

generally occurs in the neonate, serum antibodies are not present and as a consequence serological assays are not helpful in this age group.

11. Diagnosis is based on detection of *Cryptosporidium* oocysts, specific antigens, or nucleic acids present in faeces. In medical microbiology labs in high-income countries, *Cryptosporidium* is included in many of the commercially available multiplex qPCR kits, that have become a routine part of diarrhoea diagnostics over the last decade. These pathogen-panel qPCR kits are not widely available for animal diagnostics.
12. Several antibody-based commercial detection kits are available, all of which rely on the capture of oocyst wall antigens from concentrated or un-concentrated faecal samples, depending on assay format. These include immunofluorescent microscopy tests, ELISA, and immunochromatography-based kits. In recent years, the use of point-of-care (PoC) pen-side tests, also based on antigen detection (immunochromatography), and detecting multiple pathogens, have become more common in veterinary diagnostics, but are not available globally.
13. Other fluorescent staining methods (such as auramine phenol), with or without faecal concentration, are often used in clinical laboratories, and have been shown to be a low-cost reliable option.
14. High-throughput qPCR assays that can be used to differentiate between species or genotypes of interest would be of value and are available, but are currently usually restricted to specialist or reference laboratories.

Vaccines

15. Vaccines against human cryptosporidiosis are currently unavailable, but a vaccine for cattle is now available. Calves receive passive immunity via colostrum from vaccinated dams, reducing the severity and duration of neonatal diarrhoea caused by cryptosporidiosis.
16. The vaccine has been demonstrated to reduce disease in calves and also decrease the need for antimicrobial use. However the vaccine has not been available long enough for a long-term evaluation of their effect at the farm level.
17. There is a lack of information about whether the currently available vaccine is effective in hosts other than cattle, and for species that are not *C. parvum*; vaccines for livestock for other hosts (sheep, poultry, pigs) could be of value and for species other than *C. parvum*.

Pharmaceuticals

18. Halofuginone lactate is approved for use in newborn calves and can be used to prevent or reduce diarrhoea due to infection with *C. parvum*. Paromomycin is approved for use in newborn calves, lambs, and goat kids, reducing diarrhoea and oocyst shedding. It is known to be effective in high doses for the treatment of cryptosporidiosis in animal models. As paromomycin is not absorbed it is excreted into the farm environment, which may have implications for environmental effects and resistance development.
19. Evaluation of different treatment regimes with existing compounds to reduce potential side effects in terms of toxicity to the environment, the user and the animal, would be of value.
20. There may be some potential for development of new compounds for treatment of animals.
21. More robust, reliable, and informative *in vitro* models for improving and developing treatments would be an important advance. In addition, cost-benefit data (e.g., evaluation of the impact of preventive animal treatment on outbreak-associated costs) could provide impetus.

Knowledge

22. Animals, including wildlife, can be infected with *Cryptosporidium* and act as potential reservoirs for infection.
23. There are still significant areas of uncertainty in our understanding and knowledge about *Cryptosporidium* and cryptosporidiosis, especially in relation to pathogenesis, immunology, vaccinology, genetics (host and parasite, and their interactions), physiology, influence of the gut microbiome, and epidemiology.

Research is needed to fill these knowledge gaps as many are closely linked to the research requirements to develop more effective tools for cryptosporidiosis control.

Conclusions

24. Cryptosporidiosis is a widespread intestinal disease in animals and humans. *Cryptosporidium parvum* is a major pathogen in ruminant livestock and of major medical importance in people. It is a ubiquitous organism, with robust transmission stages, and cannot be eliminated. However, its transmission could be controlled and reduced. The recent adaptation of molecular methodologies for *Cryptosporidium* research will help to develop novel therapies and/or vaccines for cryptosporidiosis.

Further knowledge on the impact of the recently available vaccine that can provide passive immunity to neonatal calves will be valuable in directing further research and development, particularly regarding other species of livestock hosts and other species of *Cryptosporidium*. Effective biocides and a greater range of treatment possibilities would contribute to animal health and decrease the level of environmental contamination with infective oocysts.

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