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### Recommended Terminology and Advances in the Systematics of the Cyathostominae (Nematoda: Strongyloidea) of Horses

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## Recommended terminology and advances in the systematics of the Cyathostominea (Nematoda: Strongyloidea) of horses

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### Abstract

Terminology for common names for the Tribe Cyathostominea (cyathostomins), and disease caused by the nematodes (cyathostominosis), were recommended to replace the previously used names cyathostomes and cyathostomosis, which are ambiguous, inaccurate or synonymous, by the Third Internal Workshop on the Systematics of Cyathostominea of Horses, held in Stresa, Italy, 28 August 2001. The progress by this international working group at three workshops is reviewed briefly and a list of publications is provided. Included are an annotated checklist by genus and species of 93 species level names and the recognition of 52 species, redescriptions of seven species, and the description of one new species. Upon petition by workshop participants, the International Commission on Zoological Nomenclature placed *Cyathostomum tetracanthum* Mehlis, 1831 on the “Official List of Specific Names in Zoology”, ending more than a century of controversy over the names of cyathostomins. Some progress is described in molecular and morphological systematics and in the development of diagnostic molecular probes. A revised identification key is being prepared to the 52 species of the Tribe Cyathostominea.

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## 1. Introduction

This report summarizes progress and recommendations adopted at three, “International Workshops on the Systematics of the Cyathostominae of Horses”, convened at the last three meetings of the World Association for the Advancement of Veterinary Parasitology (WAAVP) in Sun City, South Africa (1997), Copenhagen, Denmark (1999) and Stresa, Italy (2001). These workshops were organized to facilitate international cooperation in solving problems in the terminology, identification and classification of nematodes of the tribe Cyathostominae (common name cyathostomins, not cyathostomes), parasites of horses.

These nematodes are the most common helminth parasites of horses and can cause considerable morbidity and mortality (Herd, 1990). Research activity on these nematodes is high because: (1) larval cyathostominosis (previously cyathostomosis or cyathostomiasis), a syndrome in which large numbers of larvae emerge from the walls of the large intestine and caecum and cause severe colitis that may result in death, is recognized increasingly (Mair, 1994; van Loon et al., 1995); (2) resistance to anthelmintics within the Cyathostominae has been reported widely (Fisher et al., 1992); and, (3) biological control prospects, using nematode-trapping fungi, appear to be promising (Larsen et al., 1996).

Infections with these nematodes typically include very large populations and numerous species. A total of 52 species of cyathostomins have been recorded in horses, donkeys, and zebras worldwide (Lichtenfels et al., 1998a; Matthee et al., 2002), but 10 of these species have been reported only from zebras or donkeys, and a few others have been reported only rarely (Lichtenfels et al., 1998a). Surveys worldwide have reported about 16–24 species of the Tribe Cyathostominae in most regions and from 4 to 14 species with a prevalence of 50% or higher (Reinemeyer et al., 1984; Carvalho et al., 1998; Lyons et al., 1999; Lichtenfels et al., 2001). However, the prevalence of the less common species is greatly underestimated. Chapman et al. (in press) reported that 9–15 species were found in a single animal when 200 worms were identified, but the number increased to 20–29 when all nematodes in a 5% aliquot were identified. Thus, most individual horses carry a burden of 5–10 common species (and several to many less common species), including many thousands (sometimes more than 100,000) of lumen-dwelling adult nematodes. In addition, populations of developing larval stages in the walls of the large intestine may be as large or larger than populations of adults in the lumen (Reinemeyer et al., 1984; Bucknell et al., 1995).

The major challenges to understanding and controlling these parasites are the species complexity of the nematode populations, our inability to identify eggs in feces and the difficulty in identifying larvae on pasture. Adult nematodes can be identified to species by only a few authorities using comparative anatomy. Larval stages are exceptionally difficult to identify and eggs are impossible to identify to even the subfamily level. Research worldwide on the development of diagnostic DNA markers and the testing of biological and biochemical control agents is hampered by the need to consult with an authority on the morphological identification of adults collected from sacrificed horses. However, recent studies in Scotland (Kaye et al., 1998; McDonnell et al., 2000; Hodgkinson et al., 2001) and in Australia (Hung et al., 2000), have examined molecular relationships of these species with a view to: (1) preparing a predictive classification; and (2) developing molecular markers for use in identification of both pre-parasitic and parasitic stages.

## 2. Recommendations adopted in Sun City (1997) and progress that followed

1. Publish a checklist of species and genera of the Cyathostominae. A checklist was published (Lichtenfels et al., 1998a) recognizing 51 valid species in 13 genera. The 42 other species were listed as synonyms of valid species, 10 as *species inquirenda*, and one as *nomen nudum*. In 1999 at Copenhagen it was agreed that no changes should be made to the checklist until a planned phylogeny was completed. A 52nd species, *Cylicocyclus asini*, was described recently (Matthee et al., 2002).
2. Ask the International Commission on Zoological Nomenclature (ICZN) to validate the names *Cyathostomum tetracanthum* and *Cyathostomum catinatum* of Looss, 1900. This nomenclatural adjudication was required to resolve an ambiguity over which species should bear the names *C. tetracanthum* and *C. catinatum* following the discovery by Hartwich (1986) of long-lost types of the former species. The application was listed 31 March 1998 in *Bulletin of Zoological Nomenclature* 55, 1 and the case published (Gibbons and Lichtenfels, 1999) in December 1999. Opinion 1972 on Case 3075 was published in *Bulletin of Zoological Nomenclature*, June 2001 and the following ruling made:
  - (1) All previous type specimens of the nominal species *Strongylus tetracanthus* Mehlis, 1831 are set aside and specimen no. 087757.00 in the US National Parasite Collection, Beltsville, MD collected by Looss in 1899 is designated as the neotype.
  - (2) The name *Cyathostomum* Molin, 1861 is placed on the “Official List of Generic Names in Zoology”.
  - (3) “*tetracanthus* Mehlis, 1831” type species of *Cyathostomum* Molin, 1861 and “*catinatum* Looss 1900” published in the binomen *C. catinatum* are placed on the “Official List of Specific Names in Zoology”.
  - (4) *Cylichnostomum* Looss, 1901 and *Cylicostomum* Railliet, 1901 are placed on the “Official Index of Rejected and Invalid Generic Names in Zoology”.
  - (5) “*hexacanthum* Wedl, 1856” and “*aegyptiacum* Railliet, 1923” are placed on the “Official Index of Rejected and Invalid Specific Names in Zoology”.
3. Several poorly described species should be redescribed. The following species have been redescribed: *Cylicocyclus triramosus* (by Kharchenko et al., 1997); *Cylicocyclus nassatus* and *C. ashworthi* (by Lichtenfels et al., 1997); *C. radiatus* (by Lichtenfels et al., 1998b); *Coronocyclus sagittatus* and *C. coronatus* (by Lichtenfels et al., 1999); *Cyathostomum montgomeryi* (by Kharchenko et al., 2001).
4. An updated identification key to reflect the species and genera recommended by the workshops should be developed. Projects that include the development of an updated identification key have been approved and funded by the Agricultural Research Service, US Department of Agriculture (J.R. Lichtenfels) and by the United States Civilian Research and Development Foundation (V.A. Kharchenko and J.R. Lichtenfels).
5. Specimens should be provided to researchers willing to determine molecular characteristics for systematic analysis and the development of diagnostic probes for species of the Cyathostominae. Papers published on the molecular systematics of the Cyathostominae and related nematodes include Hung et al. (1997) on *C. nassatus* and *C. ashworthi*; Kaye et al. (1998) on an intergenic spacer region between the 18S and 26S rRNA genes

of cyathostomin species; Hung et al. (1999) on molecular evidence for cryptic species within *Cylicostephanus minutus*; Hung et al. (2000) on a molecular systematic framework for equine strongyles based on ribosomal DNA sequences; and McDonnell et al. (2000) on phylogenetic analysis of mitochondrial and large ribosomal RNA gene (rDNA) sequences and the first internal transcribed spacer of the rDNA array (ITS-1) sequences of Cyathostominae and Strongylinae. Jane Hodgkinson (Hodgkinson et al., 2001) presented information on the development of five oligoprobes for cyathostomins at the 2001 Workshop in Stresa.

### 3. The second workshop in 1999 made additional recommendations

1. A phylogeny of the Strongylidae is needed to determine whether it is a monophyletic group and to determine the status of some members of the Strongylinae. Both the preliminary morphological phylogeny of Lichtenfels and Hoberg (unpublished) and the DNA phylogeny of Hung et al. (2000) supported: (a) removing *Cylicocyclus ultrajectinus* from *Cylicocyclus*; (b) subdividing *Cylicostephanus*; (c) moving some of the Strongylinae to the Cyathostominae; (d) recognizing a clade, including the species of *Cyathostomum*, *Coronocyclus*, most species of *Cylicocyclus*, *Cylicostephanus*, *Cylicodontophorus* (sensu stricto) and *Tridentoinfundibulum*; and (e) placing *Strongylus* species basal to other strongylids of horses.
2. Surveys of small strongyles can be improved with more thorough sampling. Melanie R. Chapman in collaboration with Tom Klei assessed data to see why some surveys report more species of small strongyles than others. The number of species recovered increased with the number of specimens examined. A count of 1300 worms from a single animal rather than the standard 100–200 worms gives a 95% confidence level of finding all the species present.
3. Vitaliy Kharchenko outlined additional species which should be redescribed including: *Cylicocyclus gyaloecephaloides*; *C. insigne*; *C. ultrajectinus*; *C. elongatus elongatus*; *C. elongatus kotlani*; *Cylindropharynx* spp.; *Cylicocyclus adersi*; *Trichonema aethiopicus*; *T. aequatorialis*; *T. maestri*; *T. symmetrum* and *T. zavattarii* (see Lichtenfels et al., 1998a). These are yet to appear.
4. Louis Carvalho presented morphological data for larval stages; part of thesis research which he updated at the 2001 Workshop in Stresa.

### 4. 2001 Workshop recommendation

Reports at the 2001 Workshop included several surveys of cyathostomins, species assemblies, probe development, larval morphology and morphological phylogeny.

A single recommendation was adopted at the third workshop in Stresa in 2001: whereas, several common names, including cyathostomes and small strongyles, have been used for nematodes of the Subfamily Cyathostominae, Tribe Cyathostominae; and whereas, these names are either ambiguous, inaccurate, or synonymous; The Third WAAVP International Workshop on the Systematics of the Cyathostominae of Horses recommends the use of the

common name *cyathostomin*(s) for the nematode(s), and *cyathostominosis* for the disease they cause. This term was formed in accordance with the WAAVP Standardized Nomenclature of Animal Parasitic Diseases (Kassai et al., 1988).

Progress stimulated, in part, by the three workshops has been considerable. The first workshop in Sun City was supported by funding from Hoechst-Roussel Vet, Merck Company (AgVet Division, now Merial), and Pfizer Corporation, Central Research Division. Support for the Sun City workshop was also contributed by the World Association for the Advancement of Veterinary Parasitology, the Foundation for Research Development, Republic of South Africa, the Brayton H. Ransom Memorial Trust Fund and the Agricultural Research Service, US Department of Agriculture.

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