

# 1

## Biology and Life Cycles of Equine Parasites

Life cycles comprise the set of rules that parasites must obey as they interact with the environment and eventual hosts. A thorough knowledge of life cycles is not emphasized merely to torment veterinary students. Rather, life cycle details reveal opportunities to control parasites through chemical or management interventions, to exploit unfavorable environmental conditions, or to promote natural enemies that could be agents of biological control. Practical application of these approaches will be emphasized in individual chapters of this book.

At the root of all life cycles is a fundamental principle which distinguishes helminth parasites from other infectious agents such as viruses, bacteria, fungi, and protozoa. Through various types of clonal expansion, the latter can amplify their numbers within a host animal. Literally millions of individual organisms may arise from infective burdens that are orders of magnitude smaller. The reproductive products of helminths, however, are generally required to leave the host and undergo essential change in a different location. For most helminths, defecation is the most common means of exit, but exceptions include reproductive products that are ingested by bloodsucking arthropods (e.g., *Onchocerca*, *Setaria*). Some parasitic products become infective in the environment, others require intermediate hosts or vectors, but all, regardless, occur "outside the definitive host." Dramatic biological change is mandatory before the parasitic organism is capable of infecting a new host animal, or of reinfecting the original host.

Certainly more so than those organisms which amplify their numbers through clonal expansion, helminth disease is a numbers game. As the number of invading organisms increases, greater tissue damage or nutrient loss results, and the range and severity of clinical signs become more extensive.

In this chapter, we propose to describe the basic life cycles of the major helminth parasites of equids. Mention may be made of control opportunities revealed therein, but these will be discussed more fully elsewhere in the book.

## Nematodes

### Superfamily Strongyloidea

The members of the Strongyloidea (“strongyles”) are moderately sized, stout worms with substantial buccal capsules. The males have a copulatory bursa at the posterior end, and females of all species produce eggs that are similar in appearance. Eggs of small strongyles cannot be differentiated microscopically from those of large strongyles, and the only practical method of differentiation (other than molecular approaches) is through coproculture. The strongyloids of horses all have direct life cycles; intermediate or paratenic hosts are never used (Figure 1.1).

Strongyloid eggs pass in feces and hatch in favorable environmental conditions of moisture, temperature, and oxygenation. All species exhibit three sequential larval stages, first ( $L_1$ ), second ( $L_2$ ), and third ( $L_3$ ). The  $L_1$  and  $L_2$  stages feed on organic material in the environment, but the third stage develops within the sheath of the  $L_2$ . This protective covering helps  $L_3$ s to resist environmental conditions, but it has no oral opening, and third-stage larvae are unable to ingest nutrients. The  $L_3$  is the infective stage for all strongyloid nematodes of equids. Infection invariably occurs through inadvertent ingestion, whether while grazing or via oral contact with the environment.

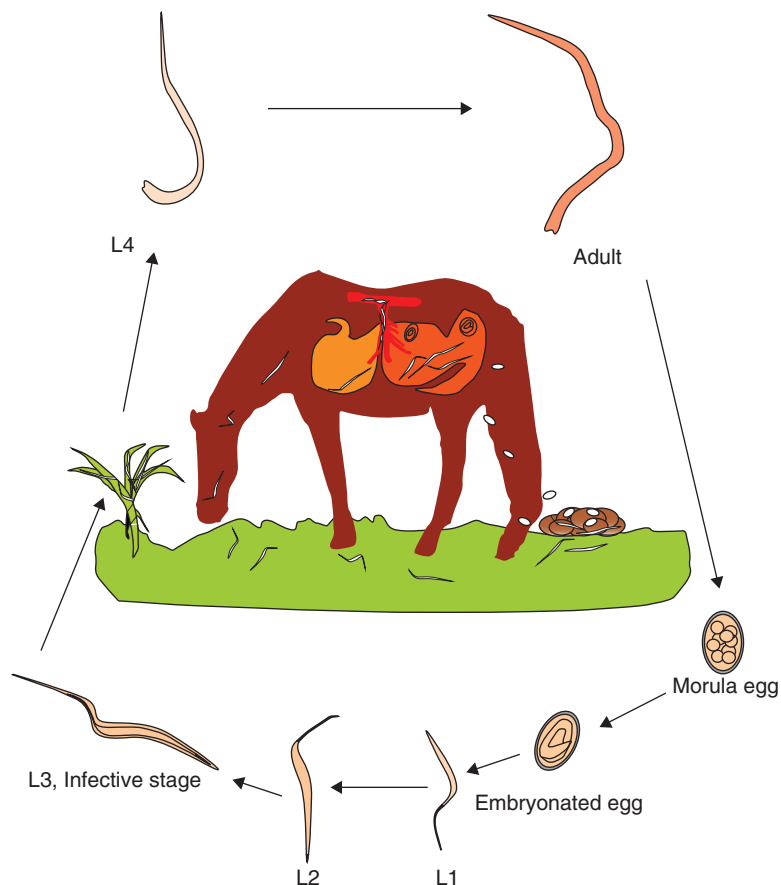
It appears that horses never develop complete immunity to strongyloids, and they are often the sole nematode parasites recovered from well-managed, mature equids. The Strongyloidea of horses comprise two distinct subfamilies: the Strongylinae and the Cyathostominae.

#### Strongylinae (large strongyles)

Members of the subclass Strongylinae tend to be larger, on average, than most genera which comprise the Cyathostominae. In addition, Strongylinae have large buccal capsules, adapted for attachment to, and even ingestion of, the intestinal mucosa. Strongyline larval stages often undergo extensive, albeit stereotypic, migration within the host prior to returning to the gut to mature and begin reproduction.

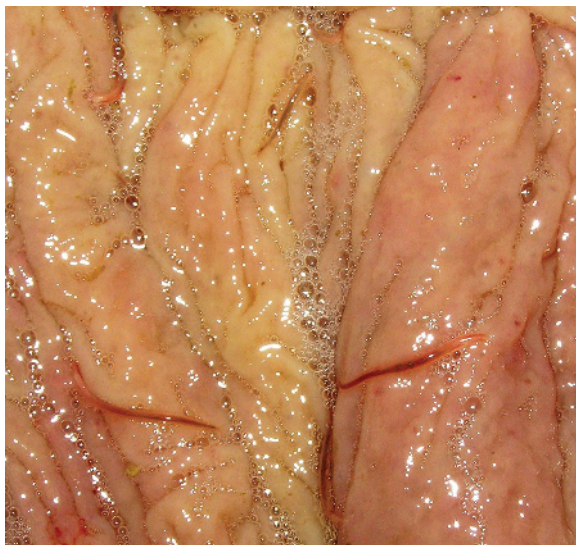
#### *Strongylus vulgaris*

*Strongylus vulgaris* is widely acknowledged as the single most pathogenic nematode parasite of horses. The adult worms measure about 1.5–2.5 cm in length, and the females are larger than the males. Adults are usually found attached to the mucosa of the cecum and the ventral colon (Figure 1.2). After ingestion from the environment, third-stage larvae invade the mucosa of the distal small intestine, cecum, and colon. Here, they molt to the fourth stage ( $L_4$ ), before penetrating local arterioles and migrating proximally beneath the intimal layer. Migrating *S. vulgaris*  $L_4$ s



**Figure 1.1** Strongyle life cycle. The life cycle of strongyle parasites. Parasitic stages can be seen above the horse and preparasitic stages below it. Fertilized eggs are shed by adult females in the cecum, and excreted to the environment in the feces. Here, the eggs hatch and a first-stage larva ( $L_1$ ) emerges. The  $L_1$  then molts to  $L_2$  in the feces. Another molt gives rise to the  $L_3$ , which retains its  $L_2$  cuticle and thus has a double-layered sheath. The  $L_3$  leaves the fecal pat and migrates onto forage, where it is ingested by a horse. Inside the horse, the  $L_3$  exsheathes and invades the mucosa of the large intestine. Large strongyles undergo extensive migration in various organs of the horse, while cyathostomins encyst in the mucosal lining of the large intestine. After returning to the large intestinal lumen, the worms reach sexual maturity and start shedding eggs

leave subintimal tracts in their wake and congregate near the root of the cranial mesenteric artery. A portion of the infecting larvae may continue to migrate, even to the root of the aorta near the left ventricle. Migrating  $L_4$ s have been found in numerous vessels arising from the aorta, including the renal arteries, and external and internal iliac arteries. The pathologic characteristics and consequences of these arterial lesions will be discussed in Chapter 2.



**Figure 1.2** Adult *Strongylus vulgaris* attached to the cecal mucosa. (Source: Photograph courtesy of Tetiana Kuzmina)

Larvae reach the cranial mesenteric artery about 2 weeks postinfection. Here, they reside for about 4 months before returning to the large intestine. The final molt to the L<sub>5</sub> stage occurs about 90 days after infection, while larvae are still present in the artery. Young adults are transported by the bloodstream to the large intestine, where pea-sized nodules form around them in the submucosa of the ventral colon and cecum. Adult worms eventually emerge from these nodules and mature in the intestinal lumen for an additional 6 weeks. Females begin to lay eggs from 5.5 to 7 months after infection (Ogbourne & Duncan 1985).

Third-stage larvae of *S. vulgaris* are very distinctive in coprocultures and are easily differentiated from those of all other strongyloid nematodes occurring in horses (Figure 8.6). The diagnosis of *S. vulgaris* will be described further in Chapter 8.

### *Strongylus edentatus*

*Strongylus edentatus* is a larger worm than *S. vulgaris*, measuring about 2.5–4.5 cm in length, and apparently is more prevalent. Adults are usually attached to the mucosa of the base of the cecum and the proximal ventral colon. The larvae undergo one of the most complex and fascinating migratory routes described in equine parasites. Following ingestion of infective L<sub>3</sub> stages from the environment, larvae migrate via the portal system to the liver, where they molt to the fourth stage within the parenchyma. Following migration within the liver, larvae migrate beneath the peritoneum via the hepatorenal ligament to various locations in the flanks and ventral abdominal wall (hence, the common term, “flank worm”). At necropsy, the large

L<sub>4</sub>s are found beneath the peritoneum covering various abdominal organs, and can also be located in isolated, hemorrhagic, and edematous lesions beneath the parietal peritoneum (see Chapter 2).

The final molt to the fifth stage occurs after about 4 months postinfection. Young adults migrate back to the large intestinal walls, where purulent nodules form and eventually rupture to release adult worms into the lumen. Altogether, this extensive migration results in a prepatent period of up to 1 year (McCraw & Slocombe 1978). The eggs of *S. edentatus* cannot be distinguished from those of other equine strongyles, large or small. Third-stage larvae of *S. edentatus* can be differentiated readily from those of *S. vulgaris* or the cyathostomins, but are fairly similar to the L<sub>3</sub> stage of the genus *Triodontophorus*.

### *Strongylus equinus*

*Strongylus equinus* is another large strongyle with a prolonged life cycle, and a prepatent period of 8–9 months from infection to egg production. The adult worms are of about the same size as *S. edentatus*. The larvae molt to L<sub>4</sub> stage upon invading the mucosal layers of the cecum and colon. They are then described as migrating across the abdominal cavity and through the pancreas to finally reach the liver, where they migrate for several weeks. On the way back to the large intestine, larvae again migrate through the pancreas and large L<sub>4</sub>s and L<sub>5</sub>s can be found free in the peritoneal cavity (McCraw & Slocombe 1984). The third-stage larvae are very distinctive in coproculture. This nematode species has become exceedingly rare in domestic herds. One author (CRR) has not recovered a single specimen of *S. equinus* from necropsies of hundreds of parasitized control horses over the past 25 years, and it typically is reported only from regions or equine populations with no or very sparse anthelmintic usage.

### *Strongylus asini*

*Strongylus asini* is a common internal parasite of zebras and donkeys in Africa. It resembles *S. vulgaris* in many ways but genetically is more closely related to *S. edentatus* and *S. equinus* (Hung *et al.* 1996). Adults occur in the cecum and colon, but larvae are found attached to the lining of hepatic and portal veins (Malan *et al.* 1982). Fourth-stage larvae migrate within the liver and hepatic cysts are reportedly found in zebras. Peritonitis has been reported in association with migrating *S. asini* larvae (Jaskoski & Colglazier 1956).

### *Triodontophorus spp.*

Although they are technically “large strongyles,” the several species of *Triodontophorus* are nonmigratory. Their larvae encyst within the lining of the large intestine and emerge to become adults. The prepatent period is thought to be approximately 2–3 months (Round 1969). *Triodontophorus brevicauda* and *T. serratus* are probably the most prevalent species of large



**Figure 1.3** Most strongyle eggs are very uniform in size. The only exceptions are the eggs of *Triodontophorus* spp. (arrow), which are about twice the size of a typical strongyle egg. (Source: Photograph courtesy of Tina Roust and Maria Rhod)

strongyles in managed horses, presumably because of a shorter life cycle than *Strongylus* species. Perhaps their privileged location in the mucosa protects them from the action of anthelmintics (e.g., ivermectin) that are effective against migrating strongylids.

*Triodontophorus* females apparently produce eggs that are significantly larger than those of the other strongyline and cyathostomin genera (Figure 1.3). *Triodontophorus* larvae can be differentiated from other strongylid nematodes by coproculture (see Chapter 8). One species, *T. tenuicollis*, causes distinct pathology that will be described elsewhere.

### Other Strongylinae

*Craterostomum acuticaudatum*, *Oesophagodontus robustus*, and *Bidentostomum ivaschkini*

These species have nonmigratory life cycles, and are only classified as Strongylinae on the basis of their large buccal capsules. The larvae derived by coproculture can be differentiated, but as the species prevalences are so low, larvae are more likely to be confused with similar, but more common, genera. None of these species has been associated with any distinct pathology.

### Cyathostominae

The Cyathostomins (also known as small strongyles, cyathostomes, or trichonemes) comprise numerous genera, including *Cylicocyclus*, *Cyathostomum*, *Cylicostephanus*, *Coronocyclus*, *Cylicodontophorus*, *Gyalocephalus*, *Poteriostomum*, *Petrovinema*, and *Parapoteriostomum* in North America and

worldwide. Lesser-known genera, such as *Hsiungia*, *Tridentoinfundibulum*, *Skrjabinodentus*, *Caballonema*, and *Cylindropharynx*, have been recovered from indigenous equids in Africa and Asia (Lichtenfels *et al.*, 2008).

The basic life cycle of all cyathostomins is virtually identical, with development to the infective third stage in the environment. Once ingested by a horse, however, L<sub>3</sub> cyathostomins do not migrate systemically. (In this handbook, migration is used consistently to indicate leaving one organ and entering another.) Rather, incoming larvae invade the mucosa or submucosa of the cecum and ventral colon, or, to a lesser extent, the dorsal colon. Cyathostomins never encyst in the lining of the descending colon or rectum. Some species apparently invade no deeper than the mucosa, whereas others encyst within the submucosa. In addition, species may prefer certain alimentary organs or even sites within an organ for encystment.

Cyathostomins first invade the large intestinal lining as early third-stage larvae (EL<sub>3</sub>). These are synonymous with infective larvae which have shed their protective integument. Early L<sub>3</sub>s are small (<1 mm) and most genera contain only eight intestinal epithelial cells. Soon after they enter the mucosa, a fibrous capsule, of host origin, forms around the EL<sub>3</sub>, and from this stage forward, these tissue larvae are referred to as “encysted” (see Chapter 2). The EL<sub>3</sub> may be a transient stage, with immediate progression through other larval stages, or individual worms may persist as EL<sub>3</sub>s for more than a year or two. The EL<sub>3</sub> is the one in which arrested development occurs.

With progressive development, the EL<sub>3</sub> molts into a late L<sub>3</sub> stage (LL<sub>3</sub>), which is significantly larger, features a tubular buccal cavity, and has more than eight intestinal cells. The LL<sub>3</sub> remains within the cyst and ultimately molts into an L<sub>4</sub> stage, which has a distinct, goblet-shaped buccal capsule. The L<sub>4</sub> grows within the cyst, but ultimately the cyst wall ruptures and the L<sub>4</sub> enters the lumen of the large intestine. The stage of emergence is also termed “excystment,” which is the chief pathologic event during the cyathostomin life cycle (Chapter 2).

The late L<sub>4</sub> (LL<sub>4</sub>) within the large intestinal lumen grows in size and eventually molts into the adult, or L<sub>5</sub> stage. The adult develops within the sheath of the L<sub>4</sub> stage, and individual worms that are in the penultimate stage of development exhibit the buccal capsule and other cephalic features of the adult, positioned just inside the remnants of the L<sub>4</sub> stage, which are about to be shed and discarded.

In addition to the larval stages, adult cyathostomins also exhibit distinct site preferences (Table 1.1). Although it is not unusual for each organ of the large intestine to harbor at least some specimens of any species, the majority of individuals of any species are usually recovered either from the cecum, ventral colon, or dorsal colon. No species occupies the descending colon or rectum as a preferred niche, so specimens recovered from those locations are considered to be exiting the host.

**Table 1.1** Examples of predilection sites of common cyathostomin species.

<p><b>Cecum</b>  <i>Coronocyclus coronatus</i>  <i>Cyathostomum alveatum</i>  <i>Cylicocyclus elongatus</i>  <i>Cylicostephanus calicatus</i>  <i>Petrovinema poculatus</i></p> <p><b>Ventral colon</b>  <i>Coronocyclus labiatus</i>, <i>Cor. labratus</i>  <i>Cyathostomum catinatum</i>, <i>Cya. pateratum</i> (also dorsal colon), <i>Cya. tetracanthum</i>  <i>Cylicocyclus auriculatus</i>, <i>Cyc. brevicapsulatus</i>, <i>Cyc. radiatus</i>, <i>Cyc. leptostomum</i>,  <i>Cyc. nassatus</i>, <i>Cyc. ashworthi</i>, <i>Cyc. ultrajectinus</i> (also dorsal colon)  <i>Cylicodontophorus bicoronatus</i>  <i>Cylicostephanus asymmetricus</i>, <i>Cys. minutus</i></p> <p><b>Dorsal colon</b>  <i>Cyathostomum pateratum</i> (also ventral colon)  <i>Cylicocyclus insigne</i>, <i>Cyc. ultrajectinus</i> (also ventral colon)  <i>Cylicostephanus goldi</i>, <i>Cys. longibursatus</i>  <i>Parapoteriostomum euproctus</i>, <i>Par. mettami</i>  <i>Poteriostomum imparidentum</i>, <i>Pot. ratzii</i></p>
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Source: Information from Tolliver (2000).

Female cyathostomins can begin to lay eggs as soon as 5 weeks after infection (Round 1969), but due to arrested development, some may not complete maturation until more than 2 years after initial infection of the host (Gibson 1953). Cyathostomins can remain in arrested development longer than any other nematode species that spends its entire parasitic life cycle in the alimentary tract. The reasons for this strategy are unclear, but the evolutionary advantages are fascinating. If climatic conditions did not permit prolonged environmental survival, it would be very beneficial for the parasite if the host transported sources of new contamination and infection. Similarly, the same strategy would suffice if nomadic horses returned to the same grazing areas, but only after intervals longer than 1 year.

Encysted cyathostomin larvae are not 100% susceptible to any known anthelmintic regimen. For this reason, it is impossible to clear a horse of all its cyathostomins. If a horse were dewormed heroically and then transferred to a sterile environment with no hope of fecal/oral reinfection, that animal would eventually begin to pass strongyle eggs again at some point in the future. And, as demonstrated by Smith (1976a, 1976b), if the horse were held in such an environment for a prolonged period and dewormed repeatedly, it may require more than 2 years before the episodic contamination would cease permanently.

The duration of survival of individual cyathostomins has not been determined with certainty, but is thought to be on the order of 3–4 months.

### **Ascaridoidea**

The superfamily Ascaridoidea comprises very large, stout nematodes with three prominent lips surrounding the oral opening. Ascarids have some of



**Figure 1.4** Adult *Parascaris equorum* in the small intestine of a weanling. (Source: Photograph courtesy of Tetiana Kuzmina)

the most complicated life cycles of any nematode of veterinary importance, but the ascarid of horses has the simplest of all.

#### *Parascaris equorum*

*Parascaris equorum* is the largest nematode parasite of horses, and mature females can reach 50 cm × 1–2 cm in size (Figure 1.4) and produce approximately 200 000 eggs per day. As adults, equine ascarids reside in the small intestine, with small numbers occasionally recovered from the stomach or cecum. Females lay distinctive eggs that are passed in the feces. Under favorable environmental conditions, eggs can become infective within 2 weeks. The infective stage is a larvated egg containing a coiled, second-stage larva. Once larvated, ascarid eggs can remain infective in the environment for up to 10 years.

Horses are infected by ingesting infective ascarid eggs from the environment. The eggs are covered by a sticky, protein coating which enables them to adhere to vertical surfaces, and even to the haircoat or udder of a mare. Foals and weanlings are most commonly infected by ascarids; and transmission is greatly assisted by the tendency of juvenile horses to investigate their environments orally.

When a larvated ascarid egg is ingested from the environment, the egg loses its protective coating after passing sequentially through the acidic and basic conditions in the stomach and small intestine, respectively. A larva emerges from the egg shell in the small intestine and penetrates the gut lining. Migrating larvae enter the lymphatics or venules draining the small intestine, and are carried passively to the liver. After infection, most larvae are found in the liver within 2–7 days postinfection. Larvae migrate within the hepatic parenchyma, which may result in inflammatory lesions and fibrous migratory tracts. Focal, white, fibrotic lesions are often seen just below the capsule of the liver (milk spots), similar to the condition caused in swine by migrating *Ascaris suum* (see Chapter 2).

Migrating third-stage larvae are found in the lungs beginning about 2 weeks after infection. Here, they exit the pulmonary venules and capillaries, and rupture alveolar membranes to enter the airways. Migrating ascarid larvae usually reside within the lungs for about 2 weeks. Eventually, the larvae migrate proximally in the pulmonary tree or are coughed up into the pharynx. Regardless of the mechanisms, they are swallowed and return to the stomach and small intestine within 4 weeks postinfection. Once in the small intestine, the worms grow progressively, and eggs appear in the feces from 75 to 90 days postinfection.

Adult ascarids continue to grow and may persist within the gut for several months. Ultimately, the majority of horses develop very strong acquired immunity to *Parascaris*, and egg shedding eventually ceases, even without benefit of anthelmintic treatment. Because of this effective acquired immunity, ascarid infections are commonly observed in sucklings, weanlings, and yearlings, but are seen only occasionally in horses after approximately 18 months of age. Immunity is acquired, however, and not just attributable to the age of the host.

In recent years, many practitioners have observed patent ascarid infections in mature horses, and some individual adult horses resume egg shedding repeatedly after apparently effective anthelmintic treatments. At present, it is unknown whether this phenomenon occurs only in horses with unique immune deficiencies, or whether some isolates of *Parascaris* do not elicit a typical immune response.

### *Parascaris univalens*

Few veterinarians are aware that a second species of ascarid exists in equids. *Parascaris univalens* differs from *P. equorum* primarily in that the genome of the former consists of only one chromosome pair, whereas the latter has two. This parasite was studied extensively in Europe during the late 1800s, and it is an interesting item of trivia that the biologic phenomenon of mitosis was first observed in the eggs of *P. univalens*. More recently, the parasite has been used to study the phenomenon of chromatin diminution (Muller & Tobler 2000). Virtually nothing is known about the applied aspects of *P. univalens* parasitism. It is considered a cryptic (sibling)

species, which cannot be distinguished morphologically from *P. equorum*, and some observations suggest that hybrids exist. It is assumed that many details of its life cycle are identical to *P. equorum*. However, it has not been established whether there are important differences between these parasites in their prevalence, pathogenicity, or the host's immune response to them. Also, it has not been determined whether these species differ in their ability to develop resistance to various anthelmintic classes.

### **Oxyuroidea**

The Oxyuroidea, or pinworms, comprise a superfamily of nematodes which reside in the posterior alimentary tract. In addition to equids, other host species include humans, rodents, primates, and sheep. The oxyuroids have a unique biological adaptation in that the females do not shed eggs into the feces. Rather, they protrude from the anus and deposit eggs in a sticky film in the perineal area. The warm, moist conditions present there likely assist in larval development. Ultimately, the dried, proteinaceous film flakes off, and eggs are dropped randomly into the environment, where they may persist for several months.

#### *Oxyuris equi*

This is the common pinworm of horses. Adult females are white and moderate in size (5–8 cm × 5 mm, and have a sharply pointed tail—thus, the common name for this group). Males are fewer in number and only approximately one-third the size of the adult females. Adult pinworms reside in the descending colon and rectum, presumably so the females will have a shorter commute to the maternity ward.

Female pinworms may be seen protruding from the anus, but are also found in fresh feces or are observed adhering to a palpation sleeve following a rectal examination.

The larvated eggs are deposited in sheets of a sticky film, which is similar in composition to dried egg albumin. Eggs are ingested from the environment, in much the same fashion as those of *P. equorum*. Third-stage larvae emerge from eggs in the small intestine and reportedly develop within the mucosa of the cecum and colon. As pinworms approach adulthood, they relocate distally in the alimentary tract. Adults do not attach to the gut wall, and have negligible pathogenicity.

Diagnosis of pinworms is accomplished by demonstrating eggs in the perineal region, either by use of the scotch tape technique or by perianal scraping (Chapter 8). Pinworm infections are not likely to be diagnosed by routine fecal flotation because of their unique reproductive behavior. However, eggs are more likely to appear in a fecal sample that was collected directly from the rectum. We attribute this to mechanical transfer of eggs to the surface of a lubricated glove (and thus onto the fecal samples) when the hand is inserted into the anus.

### *Probstmayria vivipara*

*Probstmayria* is a lesser-known and extremely small pinworm that is occasionally recovered from horses. These worms are nearly invisible to the naked eye, but might be observed during microscopic examination of fresh colonic contents. This pinworm is not known to cause any distinct clinical signs. *Probstmayria*'s reproductive behavior is rare among parasitic nematodes; it is viviparous and can complete its entire life cycle without leaving the host. For this reason, infections usually comprise massive numbers of worms, but again, they have no clinical impact.

## **Rhabditoidea**

The rhabditoid nematodes are all fairly primitive, and exhibit unique life cycle adaptations such as free-living generations and an apparent absence of parasitic males.

### *Strongyloides westeri*

*Strongyloides westeri* is a small nematode (6–9 mm) that parasitizes the small intestine of suckling foals. Females (parasitic males are unknown) are embedded within the mucosa at the base of the villi and produce small (50  $\mu\text{m}$   $\times$  40  $\mu\text{m}$ ), thin-shelled, round to slightly elliptical eggs already containing a larva. Patent infections are seen only in foals because absolute immunity is developed prior to around 5 months of age. Larvated eggs appearing in the feces of a yearling or older horse invariably are those of strongyles. Eggs pass in the feces, and L<sub>1</sub> larvae which emerge in the environment can follow various patterns of development. Some larvae become free-living males or females. The ones we are concerned about halt their free-living development as third-stage larvae, and are restricted to a parasitic existence.

Foals are infected with *Strongyloides* by one of three possible routes: skin penetration by third-stage larvae, ingestion of third-stage larvae from a contaminated environment, or lactogenic transmission from the mare. The latter route is possible because in immune adult horses, *Strongyloides* larvae do not become established in the alimentary tract. Rather, they are distributed to various somatic tissues, where they may reside for years. In mares, the hormones of pregnancy and lactation presumably stimulate the somatic larvae to resume migration and travel to the mammary glands. From this location, they are present in the mare's milk from the fourth day postpartum, and are ingested by her suckling foal (Lyons *et al.* 1973).

Most *S. westeri* infections in foals are asymptomatic, but symptomatic infections are described in Chapter 2.

### *Halicephalobus deletrix*

*Halicephalobus* (syn: *Micronema*) is a free-living rhabditoid nematode that occasionally takes up residence within living tissues. It usually gains entry

to the mammalian body through grossly contaminated lacerations or possibly through mucous membranes. *Halicephalobus* causes granulomatous lesions and is locally or systemically invasive. Spontaneous infections are seen occasionally in horses, and generally involve cephalic tissues (gingival and underlying bone, sinuses, brain) or well-vascularized organs such as the kidney (Ferguson *et al.* 2008). Human infections have been reported, but generally are subsequent to severe tissue damage and gross contamination with manure or soil.

Atypically for most parasitic nematodes, the adult worms reproduce within the host, resulting in superinfections with larvae in all stages of development. Anthelmintics are ineffective, and infections are invariably fatal.

### **Spiruroidea**

All spiruroid nematodes require an arthropod intermediate host for transmission to a vertebrate vector. The spiruroids affecting horses either occur as adults in stereotypic locations, or as larvae in a variety of aberrant tissues.

#### *Habronema muscae*

*Habronema muscae* are approximately 1–2.5 cm in length, and occur in the stomach of equids. They produce very tiny (16  $\mu\text{m}$   $\times$  45  $\mu\text{m}$ ), thin-shelled, larvated eggs that are passed in the feces. In the environment, larvae emerge and are ingested by adult dipterans (e.g., *Musca domestica*), or are swallowed by feeding maggots. Infection is completed via ingestion of dead flies in feed stuffs or water. Alternatively, infective *Habronema* larvae may travel to the mouthparts of living flies, and be deposited in wounds or at mucocutaneous junctions during feeding activities.

Within the stomach, the parasites become adults in about 8 weeks. Adult *Habronema* are found in close contact with the gastric mucosa, but they cause no clinical problems. The larvae deposited in wounds or at mucocutaneous junctions, however, can result in proliferative lesions that grow and ulcerate throughout the fly season. This condition is known as cutaneous habronemiasis, or summer sores. The presence of spiruroid larvae can be verified through histopathologic examination of biopsied tissues.

#### *Habronema microstoma*

*Habronema microstoma* is a less common species within this superfamily that uses stable flies (*Stomoxys calcitrans*) as intermediate hosts. There are no major differences in the biology or pathogenicity of the two *Habronema* species.

#### *Draschia megastoma*

The life cycle of *Draschia megastoma* is virtually identical to that of the *Habronema* spp., and the house fly (*M. domestica*) is the preferred

intermediate host. The major biological difference is that adult specimens of *Draschia* are found in large (5 cm × 5 cm), tumor-like, fibrous masses that are usually located near the *margo plicatus* of the stomach, which is the junction of the glandular and nonglandular gastric epithelium of equids. The historical prevalence of *Draschia* adults and associated lesions was 40% of 55 horses in 1984 (Reinemeyer *et al.* 1984). However, *D. megastoma* apparently has become quite rare because one of the authors (CRR) has not seen a single gastric lesion in hundreds of horses necropsied since 1985.

#### *Thelazia lacrymalis*

Horses are the definitive hosts of one species of *Thelazia*, or eye worms. As adults, *Thelazia* are found within the conjunctival *cul-de-sac* or beneath the nictitating membrane. Adult females produce larvae, which are present in the tear film of an infected eye. The usual intermediate host is the house fly, *M. domestica*, or face fly, *M. autumnalis*. Apparently, flies feeding on ocular discharges ingest larvae, which then develop to the infective stage within the body of the fly. Another horse is infected when the vector fly returns to feed on its lachrymal secretions. Infective stages leave the mouthparts of the fly, enter the conjunctival sac of the horse, and initiate a new infection.

Eye worms are thought to be relatively innocuous. They do not cause any direct damage, but might transmit pathogenic bacteria from one horse to another.

### **Filarioidea**

The filarioidea comprise a superfamily of long, thin nematodes that often reside in organs with no direct connection to the external environment. So, these worms are challenged with distributing their reproductive products into the external environment so they can undergo the essential development necessary to infect a new generation of hosts. Filarioidea accomplish this goal by producing small, motile reproductive stages known as microfilariae. Microfilariae circulate in the blood or lymph, or migrate to the skin. From these locations, they are ingested by arthropod intermediate hosts which feed on the tissues or secretions of live horses.

#### *Onchocerca*

*Onchocerca cervicalis* and *O. gutturosa* adults are found deep in the connective tissues of the nuchal ligament, and those of *O. reticulata* occupy connective tissues in the distal limbs. Microfilariae are produced by female worms, and they enter the circulatory system and travel to the dermis and epidermis. Here, they are ingested inadvertently by *Culicoides* (midges) or *Simulium* (black flies) during their feeding activities. Microfilariae develop within the tissues of the fly, migrate to the dipteran mouthparts as infective L<sub>3</sub>s, and reinfect another equid during subsequent

feeding episodes. In the new host, infective stages migrate to the target connective tissues and begin reproducing approximately 6 months after inoculation. Infection can be diagnosed by demonstrating microfilariae in skin biopsy specimens incubated in saline.

The presence of adult *Onchocerca* is usually asymptomatic, but can be a nidus of persistent bacterial infections. The microfilarial stage in skin causes cutaneous onchocerciasis, which is characterized by localized itching, hair loss, and self-trauma, and may be exacerbated by treatment with effective drugs of the macrocyclic lactone class. Although macrocyclic lactone therapy kills microfilariae and temporarily sterilizes adults, mature worms eventually resume reproduction and the clinical signs of cutaneous onchocerciasis may ultimately recur (Sellon 2007).

### *Setaria equina*

*Setaria equina* is a filarioid nematode that resides free within the abdominal cavity of equids. Although not pathogenic, it is a very prominent finding at necropsy which is hard to disavow in the presence of lay witnesses. Microfilariae are produced within the peritoneal cavity, but enter the circulation and can be found in peripheral blood. From here, they are ingested by feeding mosquitoes, and transmission is similar to that described earlier for the genus *Onchocerca*.

### *Parafilaria multipapillosa*

Adult *Parafilaria* occur in subcutaneous and intermuscular connective tissue of horses. Nodules form in the overlying skin and may rupture and bleed or leak tissue fluids. First-stage larvae are present in the exudate from bleeding lesions, and are ingested by feeding horn flies (*Haematobia irritans*). Larvae develop to the infective third stage within the fly, and are transferred to horses when flies feed on lachrymal secretions or skin wounds. The larvae then migrate in the subcutaneous tissues and develop to the adult stage within a year. Eggs and microfilariae can readily be identified in smears taken from lesion exudates.

## **Trichostrongyloidea**

Trichostrongyloids are fairly small nematodes which reside within the stomach or abomasum and small intestine of grazing animals. The free-living portions of the life cycle are virtually identical to those of the strongyloid nematodes discussed earlier. Most trichostrongyloids are parasites of ruminants.

### *Trichostrongylus axei*

*Trichostrongylus axei* is the only nematode that horses share with other domestic animals. This parasite occurs in the abomasum of sheep, cattle, and goats, and there is some possibility of cross-infection among the various host species.

*T. axei* females reside in the stomach, and produce eggs which are deposited in feces. They are fairly similar to those of the strongyloid group, but tend to be slightly smaller, more delicate, and one end of the egg is somewhat pointed. *Trichostrongylus* infection can be diagnosed readily by differential coproculture (see Chapter 8). Horses are infected by accidental ingestion of larvae during grazing. Incoming larvae invade gastric glands and develop to the adult stage, whereupon they emerge into the lumen, and begin to lay eggs 3–4 weeks after infection. Certain horses develop massive infections of *T. axei*, involving thousands of individual worms. These horses exhibit hypertrophy of the glandular mucosa, but it is unknown if this condition results in any digestive disturbances.

In ruminants, *T. axei* infection is susceptible to anthelmintics of the benzimidazole and macrocyclic lactone classes; similar efficacy is likely in horses. However, a specific label claim does not exist for the equine products, due to the difficulty of demonstrating efficacy against infections of such low prevalence.

#### *Dictyocaulus arnfieldi*

*Dictyocaulus arnfieldi* is the lungworm of equids. Adults live in the terminal bronchioles and can be found in the major airways. Subsequent to reproduction, larvae deposited in the bronchial secretions are carried proximally to the pharynx by the ciliary apparatus or spontaneous coughing. The larvae are then swallowed and passed in the feces. Diagnosis involves using the Baermann technique to demonstrate larvae in the feces.

*D. arnfieldi* is considered a normal parasite of donkeys, because it reproduces readily and induces little pathogenicity. Horses, however, will rarely support an infection to the adult stage because they are not suitable definitive hosts. Thus, attempting to diagnose lungworm infection in a horse by demonstrating reproductive stages in the feces is a fruitless endeavor. Infection of a horse can be confirmed by a transtracheal wash to demonstrate eosinophilic bronchitis, or treatment with a macrocyclic lactone anthelmintic may be curative. Infected horses invariably have a history of sharing common pasture with donkeys.

## Cestodes

### Anoplocephalidae

Equids harbor only three species of cestodes, and only one of those can be considered common. All are members of a closely related family, and like nearly all other cestodes require an intermediate host for transmission. Unlike nematodes, equine cestodes apparently do not release individual eggs on a regular basis. Rather, terminal (gravid) proglottids probably detach and disintegrate during transit to the external environment. This results in a rather patchy distribution of cestode eggs within the fecal output of infected horses, with obvious diagnostic implications.



**Figure 1.5** Anterior end of an *Anoplocephala perfoliata* specimen obtained from the cecum of a horse at necropsy. *A. perfoliata* is characterized by four suckers (arrows) with corresponding lappets (arrowheads) just beneath them. (Source: Photograph courtesy of Tina Roust and Maria Rhod)

In the environment, cestode eggs within feces are ingested by free-living soil mites of the family Oribatidae, which are endemic in soils worldwide. After ingestion, an oncosphere (essentially the scolex of a future adult worm) is digested from the egg within the alimentary tract of the mite. The oncosphere migrates into the hemocoel (body cavity) of the mite, and develops into an infective stage known as a cysticercoid. Cysticercoids probably remain infective for the life span of the mite host. It is likely that infected mites can persist in the environment for longer than a single climatic season.

Horses are infected via inadvertent ingestion of vector mites while grazing. The cysticercoids are digested free of the mite's tissue in the horse's gastrointestinal tract, and primitive scolexes attach to the lining of the preferred region of gut. Adult cestodes are able to regenerate an entire organism (known as a strobila) from the attached scolex.

#### *Anoplocephala perfoliata*

*Anoplocephala perfoliata* is the most common cestode of equids worldwide, and has been reported from every continent except Antarctica. It is a moderately sized worm, ranging from 4 to 8 cm in length and 1 to 2 cm in width (Figure 1.5). Unlike the cestodes of other mammalian species, it is rare to observe proglottids in the feces of horses, at least of those infected with *A. perfoliata*. *Anoplocephala* infection can be diagnosed by fecal examination, but this technique has fairly low sensitivity in horses, as discussed elsewhere (see Chapter 8).

*A. perfoliata* is a rare exception to the general rule that all adult cestodes reside within the small intestine of the respective host. Adult and

developing *A. perfoliata* are mostly found attached to the lining of the cecum, and the majority tend to cluster on the cecal side of the ileocecal valve. It is not uncommon for additional masses of cestodes to be distributed in two or three locations within the cecum, and individual specimens can also be found attached to the mucosa of the ventral colon. The longevity of individual specimens of *A. perfoliata* is unknown, but is more likely to be months rather than weeks or years.

Several studies have demonstrated a clear, seasonal pattern in the prevalence and abundance of *A. perfoliata*. In temperate climates, most patent infections are observed in the second half of the year, reflecting infections that were acquired and established over the preceding grazing season (Meana *et al.* 2005).

### *Anoplocephala magna*

True to its name, *Anoplocephala magna* is the largest cestode occurring in equids, and may achieve 80 cm in length. *A. magna* normally attaches to the mucosa in the distal small intestine, and can be differentiated from *A. perfoliata* by its relative size and preferred location in the host. For definitive identification of individual specimens, *A. perfoliata* exhibits small structures, termed “lappets,” beneath each sucker, whereas *A. magna* lacks lappets (Figure 1.2).

Nearly a century ago, *A. magna* was reportedly far more prevalent than *A. perfoliata*, but the relative ranking of these species has reversed over time. At the present time, *A. magna* is encountered infrequently in North America.

### *Anoplocephaloides mamillana* (formerly *Paranoplocephala mamillana*)

This is a very uncommon parasite of equids, which normally attaches to the mucosa of the proximal small intestine. It is a very tiny worm, only 6–50 mm long and 4–6 mm wide. The eggs differ in appearance and size from those of the *Anoplocephala* species. *Anoplocephaloides mamillana* is little more than a biological oddity and diagnostic differential; infections are not known to have any clinical impact.

## Arthropods

Only one arthropod will be discussed herein, namely, members of the genus *Gasterophilus*, known commonly as bot flies.

Horse bot flies are members of a larger family, known as Oestrid flies. Although the details and the host distributions differ markedly, all oestrid flies employ the same general strategy, which is for their offspring to avoid unfavorable environmental conditions by passing their larval stages within the body of an intended host. The Oestrids of large domestic animals deposit eggs or larvae directly onto the intended host. Once the larval stage becomes active (after egg-hatching in some cases), they enter



**Figure 1.6** *Gasterophilus* spp. eggs attached to the haircoat of the leg. (Source: Photograph courtesy of Tetiana Kuzmina)

the host by specific routes. Thus, some Oestrids (e.g., *Hypoderma* of cattle) hatch from eggs attached to the haircoat, and the larvae penetrate intact skin and undergo sustained systemic migrations. Others (e.g., *Oestrus* of sheep) are deposited as larvae within the nares, and migrate only locally and develop within the sinuses. In most cases, the larvae overwinter within the host, emigrate from the host in spring, pupate in the soil, and emerge as adults to complete another generation. Most Oestrids are univoltine, meaning they propagate only a single generation per year.

Female flies of the genus *Gasterophilus* attach eggs to individual hairs of equid hosts (Figure 1.6), and larvae gain access to the oral cavity via routes that vary by species. Bot larvae generally overwinter within the equine alimentary tract, pass from the host in the feces during spring or early summer, and pupate in loose soil. Adult flies emerge from the soil 1–2 months later and emerge to mate and reproduce. Adult Oestrids have very brief life spans, due in part to their absence of mouthparts, which renders them incapable of ingesting nutrients.

#### *Gasterophilus intestinalis*

*Gasterophilus intestinalis* is the most prevalent and numerous of the bot species in domesticated horses. Female flies hover and glue individual eggs to hair shafts on the distal forelimbs, and occasionally along the neck and mane. Eggs hatch in response to contact with the horse's lips (Bello 1967), hatch immediately, and attach to the lips and tongue. Eggs which are laid on the mane are probably ingested by herd mates during mutual grooming. First-instar larvae are embedded within folds in the tongue.

They subsequently molt to the second instar, and move to gingival pockets around the molars and premolars. The second instars eventually emerge, to be swallowed and passed into the stomach. Here, they attach to the mucosa in the nonglandular portion of the stomach and develop to the third instar. *G. intestinalis* larvae are dark red and spiny, and about 2 cm long and 5–8 cm wide. Burdens of several hundred bots are common in horses, and can be visualized easily and even enumerated gastroscopically. Features of bot pathogenicity are described in Chapter 2, and control is discussed in Chapter 7.

#### *Gasterophilus nasalis*

Female *Gasterophilus nasalis* flies deposit their eggs in the intermandibular area. The eggs hatch spontaneously and larvae crawl independently to the lips, enter the oral cavity, and develop in pockets in the tongue and around the cheek teeth. Ultimately, second instars are swallowed and develop further in the alimentary tract. Second and third instar *G. nasalis* prefer to attach in the ampulla of the duodenum, just a few centimeters past the pylorus. This area of the equine alimentary tract can be accessed and visualized endoscopically, so it is possible to detect this parasite antemortem.

#### *Other Gasterophilus species*

Other bot species which apparently do not occur in North America include *G. inermis* and *G. hemorrhoidalis*. The latter species attaches in masses in the distal small colon and rectum of donkeys in Africa, and has been documented as a cause of rectal prolapse. Other minor species are distributed around the globe, but none has distinctive pathogenicity.

## Trematodes

Trematodes are uncommon parasites of horses in most developed countries. The liver fluke, *Fasciola hepatica*, occasionally infects horses, but is seen only in areas where fascioliasis is endemic in traditional, ruminant hosts. Horses with liver fluke infections inevitably have been pastured where microclimates favor the development of molluscan intermediate hosts. Readers are referred to Nansen *et al.* (1975) for a detailed description of the life cycle and clinical features of equine *Fasciola* infection.

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