Nippostrongylus
(helminth: nematode)

Overview

Nematodes are triploblastic pseudocoelomate unsegmented worms that undergo protostomial embryonic cleavage and grow by cuticular moulting (ecdysis). Two groups identified by the presence/absence of sensory phasmids have partly been ratified by molecular studies recognising three subclasses: Enoplia and Dorylaimia (both without phasmids) and Chromadoria (most with phasmids). Many phasmidian parasites of vertebrates are grouped in the chromadorian order Rhabditida; including spirurinids, tylenchinids and rhabditinids. The latter contains the infraorder Rhabditomorpha which includes strongyloid nematodes characterised by an expansion of the tail of the male known as the copulatory bursa (clasper with one dorsal and two lateral lobes with muscular rays). Many families are recognised: including the heligmosomatids which are hookworm-like parasites found in the intestines of small mammals and birds. They have direct life-cycles, usually involving eggs passed in faeces developing to L3 which are ingested by new hosts and develop to adults. *Nippostrongylus* spp., however, may also infect hosts percutaneously, when L3 penetrate the skin, migrate through the lungs and form adults in the gut. Infections in rodents are generally subclinical, but heavy infections may cause verminous pneumonia. *N. brasiliensis* has been used as a laboratory model, although it can become problematic in animal colonies.

Classification:

Domain:	Eukaryota (membrane-bound nucleus)
Supergroup:	Amorphea (unikonts with single flagellum, or nonflagellated amoebae)
Kingdom:	Metazoa (multicellular eukaryotes, heterotrophs, notably animals)
Group:	Protostomia (triploblastic, spiral cleavage)
Subgroup:	Ecdysozoa (cuticle moulted = ecdysis)
Phylum:	Nematoda (unsegmented, pseudocoelomate roundworms, tubular digestive tract, dioecious)
Class:	Chromadorea (spiral amphids, three oesophageal glands, usually annulated bodies, free-living and parasitic)
Order:	Rhabditida (Secernentea, Phasmidea) (secretors, with phasmids, bipartite oesophagus, single testis)
Suborder:	Rhabditina (free-living or parasitic in invertebrates/lower vertebrates)
Infraorder:	Rhabditomorpha (‘rod-shaped’ buccal cavity)
Superfamily:	Strongyloidea (bursate males, prominent buccal capsules, parasites of mammals, birds, reptiles)
Family:	Heligmosomatidae (adults filiform, reddish in colour, direct cycle)
Genus:	*Nippostrongylus* (parasitic in small intestines of rodents)
Species:	*N. brasiliensis* causes verminous pneumonia in rodents

Parasite biodiversity and host range: Most Metazoa are multicellular triploblastic animals with differentiated tissues, many being bilaterally symmetrical with a body cavity. Most invertebrate animals are protostomes as their embryonic development involves spiral determinate cleavage. Those that moult their external cuticles during their life-cycles (process known as ecdysis) are grouped together in the unique clade Ecdysozoa, including the nematodes (roundworms), onychophorans (velvet worms), tardigrades (water bears) and arthropods (myriapods, chelicerates, crustaceans and hexapods, all with jointed limbs). Nematodes (roundworms) are unsegmented tubular worms with a fluid-filled body cavity (pseudocoelom) that acts as a hydrostatic skeleton. They have longitudinal muscles and typically exhibit a sideways thrashing motion. They have well developed digestive tracts with various partitions: the foregut comprising the mouth (often with lips and papillae), buccal capsule (sometimes with ridges, rods, plates, spears, stylets or teeth) and oesophagus (glandular, muscular or both); the midgut (nonmuscular absorptive section); and hindgut (rectum) emptying through a subterminal anus (cloaca in males). Most nematodes are dioecious and form separate sexes. Male worms have a single testis (sometimes 2), an elongate vas deferens often equipped with a seminal vesicle and ejaculatory duct (glandular and/or muscular), 1-2 copulatory spicules (sometimes with an accessory gubernaculum), and bursate species with elaborate posterior claspers. Female worms are usually didelphic with 2 ovaries (some monodelphic or polydelphic), 2 oviducts usually with spermatheca, 2 uteri opening into a common vagina and a vulva often equipped with a muscular ovejector. Female worms are oviparous or viviparous and produce numerous eggs or larvae, respectively. Larval stages undergo several moults (L1-L4) before maturing into adult worms. Some nematodes have direct life-cycles where eggs or larvae infect definitive hosts (per os or per cutaneous), but many have indirect cycles where larvae first develop in invertebrate intermediate hosts before infecting definitive hosts (by ingestion, injection or deposition). Many nematode species are free-living in terrestrial and aquatic habitats, while some species from diverse groups have become plant or animal parasites. Two nematode groups identified by the presence/absence of sensory phasmids have partly been ratified by molecular studies recognising three subclasses: Enoplia and Dorylaimia (both without phasmids) and Chromadoria (most with phasmids). Most Enoplia are free-living marine organisms but some are found in freshwater, and on land as plant parasites. The Dorylaimia comprise numerous freshwater and terrestrial species,

including major groups of plant and animal parasites. The Chromadoria is represented by many marine groups as well as a terrestrial group of plant and animal parasites. The taxonomic ranks of many nematode assemblages vary considerably depending on which classification system has been followed. Molecular phylogenetic studies, however, have supported the separate classification of most groups, particularly at the level of superfamily. Collectively, species from at least 16 superfamilies are considered to pose serious threats to human and animal health as infectious diseases.

CLASSIFICATION* OF SUPERFAMILIES OF PARASITIC NEMATODES
Class: Enoplea (Aphasmidea, Adenophorea) (gland-bearers, cylindrical oesophagus, no phasmids, setae, two testes)
Subclass: Dorylaimia (five or more oesophageal glands, buccal stylet (odontostyle), free-living or parasitic)[clade I(2)]
Order: Trichinellida (Trichocephalida, Trichurida) (single spicule, stichosome oesophagus, L1 with buccal stylet)
Superfamily: Trichinelloidea (oesophagus with short anterior muscular and long posterior glandular portions)
Class: Chromadorea (spiral amphids, 3 oesophageal glands, usually annulated bodies, free-living and parasitic)
Order: Rhabditida (Secernentea, Phasmidea) (secretors, phasmids present, amphids anterior, bulbous oesophagus)
Suborder: Rhabditina (free-living or parasitic in invertebrates/lower vertebrates)[clade V(9)]
Infraorder: Rhabditomorpha (‘rod-shaped’ buccal cavity)
Superfamily: Rhabditoidea (open tube stoma, excretory system with lateral canals)
Superfamily: Strongyloidea (bursate males, prominent buccal capsules, parasites of mammals, birds, reptiles)
Suborder: Spirurina (animal parasites, many use invertebrate intermediate hosts (IH))[clade III(8)]
Incertae sedis Superfamily: Dracunculoidea (elongate parasites of vertebrate tissues, freshwater crustacean IH)
Infraorder: Ascaridomorpha (large roundworms, three large lips, numerous caudal papillae)
Superfamily: Ascaridoidea (ascarids, eggs thick-shelled, larvae may undertake hepato-pulmonary migration)
Superfamily: Heterakoidea (preanal sucker anterior to cloaca in males, direct cycle, infection by egg ingestion)
Infraorder: Gnathostomatomorpha (‘jaw-mouthed’ due to unique bulbous armed heads)
Superfamily: Gnathostomatoidea (first IH copepod, often use paratenic hosts)
Infraorder: Oxyuridomorpha (pinworms, pointed tails, oesophagus with terminal bulb, males with single spicule)
Superfamily: Oxyuroidea (common in mammals, birds, reptiles, amphibians)
Infraorder: Spiruromorpha (enigmatic clade linked by molecular characters, indirect cycles with IHs)
Superfamily: Acuarioidea (small parasites mostly of birds, with cephalic cordons, ptilina or serrated shields)
Superfamily: Camallanoidea (conspicuous phasmids, L1 with dorsal tooth, ovoviviparous, L1-L3 in copepod)
Superfamily: Filarioidea (tissue-dwelling filarial parasites, lack lips, infect tissues/vessels, arthropod IH)
Superfamily: Habronematoidea (unique head structures with small pseudolabia and median lips)
Superfamily: Physalopteroidea (stomach worms in mammals, insect IH)
Superfamily: Spiruroidea (pseudolabia, bipartite oesophagus, infect birds (crop/gizzard), arthropod His
Superfamily: Thelazioidea (eye-worms of birds and mammals, transmitted by insects)
Suborder: Tylenchina (fungal, plant and animal parasites)[clade IV(10,11,12)]
Infraorder: Panagrolaimomorpha (free-living or parasitic (insects, reptiles, amphibians, mammals))
Superfamily: Strongyloidoidea (dauer stages, lip region without processes, striated cuticle)

*Contemporary genotypic classification schemes recognize strong monophyletic clades at the level of superfamily and infraorder, while previous phenotypic classification schemes had ranked many as separate orders.

The superfamily Strongyloidea comprises a range of worms often with prominent buccal capsules and specialised oral structures well-suited to their feeding habits on host tissues and/or fluids. Adults of most species are parasitic in the gastrointestinal tracts of mammals and some birds, while larval stages feed on bacteria in the external environment, although some larvae may infect invertebrates as intermediate or paratenic hosts. The adult worms are sexually dimorphic, the smaller males characterised by an expansion of the tail (bursa) which is used as a copulatory clasping organ. Many classification schemes group these ‘bursate’ nematodes into one or more superfamilies in the order Strongylida (with suborders containing the strongyles, trichostrongyles, hookworms and lungworms), although the families essentially remain the same. Many families are recognised on the basis of parasite morphology, biology, life-cycle, host specificity and tissue tropism; including the following which contain many notorious parasites of vertebrates.

Representative Strongyloidea (cf. Strongylida) [with bursate males]				
Family	**Characters**	**Definitive Hosts**	**Transmission***	**No. genera**
Trichostrongylina (trichostrongyles)				
Heligmonellidae (hookworm-like)	body coiled, cephalic vesicle, ridged synlophe, bursa asymmetrical	mammals, birds	transdermal penetration of L3	56
Trichostrongylidae (trichostrongyles)	reduced buccal capsule, ridged synlophe, oesophagus lacking bulb, thin-shelled eggs	artiodactyls, birds	ingestion of L3	50
Molineidae (stomach/intestinal worms)	reduced buccal capsule, cephalic vesicle, female tail with spine or cusps, oviparous/viviparous	mammals, birds, reptiles	ingestion of L3	61
Strongylina (strongyles)				
Strongylidae (strongyles)	large buccal capsule often armed with teeth, leaf crown around mouth	mammals, reptiles, birds	ingestion of L3	32
Chabertiidae (nodule worms)	large buccal capsules, leaf crown of labial collar, L3 sheathed	artiodactyls, primates	ingestion of L3	22
Syngamidae (gapeworm)	cup-shaped buccal capsule, armed with teeth, male attached to female	birds, mammals	ingestion of L3 or invertebrate PH	7
Stephanurinae (kidneyworm)	buccal capsule armed with teeth, leaf crowns and external epaulettes	suids	transdermal penetration or ingestion of L3 or PH	1
Ancylostomatina (hookworms)				
Ancylostomatidae (hookworms)	large buccal capsule bent dorsally, armed with teeth/cutting plates	primates, carnivores, artiodactyls	transdermal penetration of L3 (sometimes *per os*)	20
Metastrongylina (lungworms)				
Metastrongylidae (lungworms)	small buccal capsule, 2 trilobed lips, bursa with reduced dorsal lobe	suids	ingestion of IH carrying L3	1
Protostrongylidae (lungworms)	small buccal capsule, bursa with large lobes, gubernaculum	artiodactyls	ingestion of IH carrying L3	17
Angiostrongylidae (lungworms)	no or reduced buccal cavity, short club-shaped oesophagus	carnivores, rodents	ingestion of IH or PH carrying L3	28
Dictylocaulidae (lungworms)	small buccal capsule, bursa with large lobes, short stout spicules	ungulates, reptiles	ingestion of L3	5
Filaroididae (lungworms)	small buccal capsule, reduced male bursa, infective L1	carnivores	ingestion of L1	4

*IH = intermediate host, PH = paratenic (transport) host, L1 = first-stage larva, L3 = third-stage larva

Several families in the superfamily Strongyloidea have been grouped together with the trichostrongyles, while others have placed them in a new superfamily Heligmosomoidea; namely: Heligmonellidae (56 genera in mammals); Heligmosomidae (8 genera in small mammals); Herpetostrongylidae (13 genera in reptiles and marsupials); Nicollinade (2 genera in monotremes and marsupials); Ornithostrongylidae (8 genera in mammals); and Viannaliidae (7 genera in mammals). These worms are generally coiled and their synlophe is not bilaterally symmetrical. The family Heligmonellidae contains small worms with cephalic vesicles, rudimentary buccal capsules (reduced to an annulus), bursate males, and monodelphic oviparous females without spined tails. Worms have a synlophe with conspicuous longitudinal elevations (ridges), some of which are fused (comaretes) and some supported by underlying thickenings (paired thickenings termed carenes). Four subfamilies are recognised: Nippostrongylinae (carene present or absent, lateral ridges poorly developed, synlophe with 4-46 circumferential ridges continuous along length, axis of orientation of ridges 45-67° from sagittal axis); Brevistriatinae (carene present or absent, lateral ridges poorly developed, absent on ventral quadrant, axis of orientation of ridges 67-90° from sagittal axis); Heligmonellinae (carene absent, lateral ridges strongly developed, axis of orientation of ridges < 45° from sagittal axis); and Pudicinae (carene present, cuticular ridges absent on ventral quadrant, axis of orientation of ridges 67-90° from sagittal axis, dorsal ray divided in proximal half or with ventral comaretes).

Genus	**No. spp.**	**Definitive Hosts**	**Location**	**Adult worms**	**Worm eggs**
Subfamily Nippostrongylinae					
Nippostrongylus	10	rodents	small intestines	2-6 mm long, reddish in colour, reduced buccal capsule, oblique cuticular ridges, bursa asymmetrical (2-2-1), larvae with longitudinal alae	50-70 x 28-35 µm, ellipsoidal, thin-shelled

The subfamily Nippostrongylinae contains 29 genera (*Bunomystrongylus, Carolinensis, Euzetoda, Guerrerostrongylus, Hasanuddinia, Hassalstrongylus, Heligmonina, Heligmonoides, Hypocristata, Malaistrongylus, Malvinmea, Mammanidula, Mawsonema, Maxomystrongylus, Melomstrongylus, Montistrongylus, Neoheligmonella, Nippostrongylus* (syn. *Austroheligmonema*)*, Odilia, Orientostrongylus, Paraheligmonelloides, Parasabanema, Rattustrongylus, Sabanema, Spalacina, Stilestrongylus, Suttonema, Trichofreitasia* and *Yatinema*) parasitic in rodents, dermopterans, lagomorphs, primates and insectivores. The genus *Nippostrongylus* is characterised by small red worms with cephalic vesicles, small buccal cavities, synlophes with oblique cuticular ridges and bursate males with asymmetrical dorsal rays. Ten species are recognized in a wide range of rodents and some in lemurs, lagomorphs and talpid insectivores. Infections are more common in temperate and tropical regions where warm moist conditions are more favourable for the survival of free-living stages. The host specificity of individual parasite species varies from narrow (oioxenous) to broad (euryxenous). The species *N. brasiliensis* is frequently found worldwide in rodents and has been used extensively as a laboratory model.

***Nippostrongylus* species**	**Definitive hosts**	**Location [Clinical signs]**	**Distribution**
N. brasiliensis (syn. *N. muris, Heligmosomum muris*) (rat intestinal worm, rat 'hookworm')	Rodentia: murid (black rat, Philippine black rat, brown rat, African grass rat, soft-furred rat, ricefield rat, lesser ricefield rat, Himalayan field rat, dusky field rat, Polynesian rat, Tanezumi rat, chestnut white-bellied rat, bush rat, house mouse, wood mouse, Eurasian harvest mouse, Algerian mouse, typical striped grass mouse, striped field mouse, Natal multimammate mouse, African pygmy mouse, gerbil), cricetid (golden hamster, reed vole, deer mouse), nesomyid (Gambian pouched rat), chinchillid (chinchilla); Eulipotyphla: soricid (greater white-toothed shrew); Lagomorpha: leporid (rabbit)	small intestines [pneumonia]	worldwide
N. djumachani (syn. *Longistriata*)	Rodentia: murid (bandicoot rat)		Asia, North Africa
N. magnus (syn. *Austroheligmonema*)	Rodentia: murid (fawn-footed mosaic-tailed rat, bush rat, brown rat, black rat, dusky field rat)		Australia, New Guinea, Indonesia
N. marhaeniae	Rodentia: murid (Moluccan prehensile-tailed rat)		Indonesia
N. rauschi	Dermoptera: cyanocephalid (Philippine flying lemur)		Philippines
N. rysavyi (syn. *Oswaldonema*)	Rodentia: cricetid (Chinese striped hamster, black rat)		Asia
N. sembeli	Rodentia: murid (yellow-tailed rat)		Indonesia
N. smalesae	Rodentia: murid (Whitehead's spiny rat)	small intestines	Indonesia
N. typicus (syn. *Austroheligmonema*)	Rodentia: murid (bush rat, dusky field rat)		Australia, New Guinea
N. witenbergi	Rodentia: murid (short-tailed bandicoot rat)	small intestines	Asia, North Africa

Parasite morphology: *Nippostrongylus* spp. form 3 different morphological stages in their developmental cycles: eggs, larvae (4 consecutive stages designated L1-L4); and adult worms. Freshly-laid eggs are thin-walled, ellipsoidal in shape measuring 50-70 x 28-35 µm, and contain a morula usually comprising 16-20 cells (sometimes 4-16 cells). First-stage larvae (L1) released from hatched eggs are elongate (280-300 µm long), bound by a thin transparent cuticle with lateral alae and have a rounded head, a rhabditiform (bulbed) oesophagus, and a tapering tail terminating in a sharp point. L2 are larger (470-750 µm long), bound by a thicker cuticle with obvious striations and longitudinal alae, and have rounded heads with an elongate buccal cavity, a rhabditiform oesophagus, darkened intestinal cells, and an attenuated tail. L3 are initially ensheathed in that they retain the L2 cuticle as a protective covering, but over time (days-weeks) the sheath contracts and develops an anterior split through which the L3 slowly and tentatively emerges. Ensheathed larvae measure up to 770-820 µm in length, while freed L3 measure from 600-750 µm. Infective L3 are bound by a thick cuticle with longitudinal alae, and have a rounded head with a small compressed buccal cavity, a thin strongyliform (non-bulbed) oesophagus, and a short tail. L4 are parasitic stages in which the genital primordia have begun to develop into specific male or female organs. Adult worms are small in size measuring from 2-6 mm in length and have a reddish colour due to endogenous haemoglobin present in the cuticle and tissues (and not from feeding on host blood). Their heads have an anterior expansion of the cuticle (cephalic vesicle) up to 50-70µm wide and they have a small buccal capsule without teeth or lips. Worms are bound by a distinctive synlophe with 14 or more oblique cuticular ridges (axis of orientation 45-67° from sagittal)

sometimes supported by poorly-developed underlying thickenings (carenes). Adult worms are sexually dimorphic, with females being larger than males (2.5-6.0 x 0.09-0.13 mm cf. 2.1-4.5 x 0.08-0.10 mm). Mature males have an asymmetrical copulatory bursa consisting of two large lateral lobes supported by unequal rays (comprising muscular elements following nerve channels to terminal papillae) arranged in a 2-2-1 configuration, and a small dorsal lobe (supported by a dorsal ray split posteriorly into unequal branches). They also have 2 brown filiform spicules measuring from 0.44-0.55 mm in length. Mature females have a bell-shaped posterior end with a short conical tail, usually curved ventrally and lacking a terminal spine. They are monodelphic with a single ovary and uterus terminating in a posterior vulva overlaid by a small circular cuticular projection.

Site of infection: Adult worms are found in the small intestinal mucosa of their rodent hosts, predominantly in the duodenum and jejunum, but sometimes extending into the ileum. Developing larval stages may be found in various host organs and tissues as infective L3 undertake somatic migration via the circulation and/or lymphatics, with most forming L4 in the lungs and migrating up the trachea to reach the gut. Worm eggs are voided into the external environment and give rise to free-living larval stages before becoming infective to rodents.

Pathogenesis: Light infections often remain asymptomatic, but heavier infections may cause acute and/or chronic clinical disease, the severity of which depends on parasite burdens (number of infective L3), host susceptibility (young and malnourished (iron and protein deficient) hosts most at risk) and immune status (prior exposure stimulates protective immune responses). There are 3 stages of infection which cause different types of disease: first, skin penetration by larvae causing dermatitis; then pulmonary migration of larvae causing lobar pneumonia and compensating emphysema; and finally, enteric feeding by adults causing malabsorption and anaemia. Infective L3 penetrate the skin and feed on host tissues while migrating through loose connective tissue to subcutaneous veins. Moderate to heavy infections may cause small haemorrhages and mild inflammatory responses in dermal tissues, which usually subside spontaneously within 2-3 days. Many larvae then migrate to the lungs where they feed and develop to L4, causing pulmonary inflammation, petechial haemorrhages, oedema, increased mucus production, alveolar consolidation and airway blockages leading to verminous lobar pneumonia (similar to chronic obstructive pulmonary disease (COPD)) with focal areas of compensating emphysema. L4 then ascend the bronchial muco-ciliary escalator to the trachea where they are swallowed reaching the small intestines after several days. Immature and mature worms burrow in the intestinal villi and crypts, often forming small pockets of worms coiling spirally around villi. Worms have small mouthparts and feed by releasing excretory-secretory (ES) products (including enzymes) and then ingesting the resultant slurry of host tissues and gut content. Feeding lesions may cause small haemorrhages with blood lost in the faeces, sometimes resulting in anaemia, particularly in iron-deficient individuals. However, local T-cell dependent immune responses cause most of the damage to the intestinal mucosa, which becomes grossly thickened and oedematous with red thickened pockets sometimes double normal size. The architecture and integrity of the mucosa changes markedly, with epithelial degeneration, autolysis, crypt hyperplasia, villous atrophy and sprue-like lesions producing a flat mucosa with reduced surface area leading to maldigestion, malabsorption, diarrhoea, anorexia, lethargy, malnutrition, weight loss and reduced growth rate (failure to thrive). Nevertheless, infected animals acquire a strong protective immunity resulting in the expulsion of adult worms, although sometimes a small population may survive for a prolonged period (known as concomitant or premunitive immunity, rather than sterile immunity).

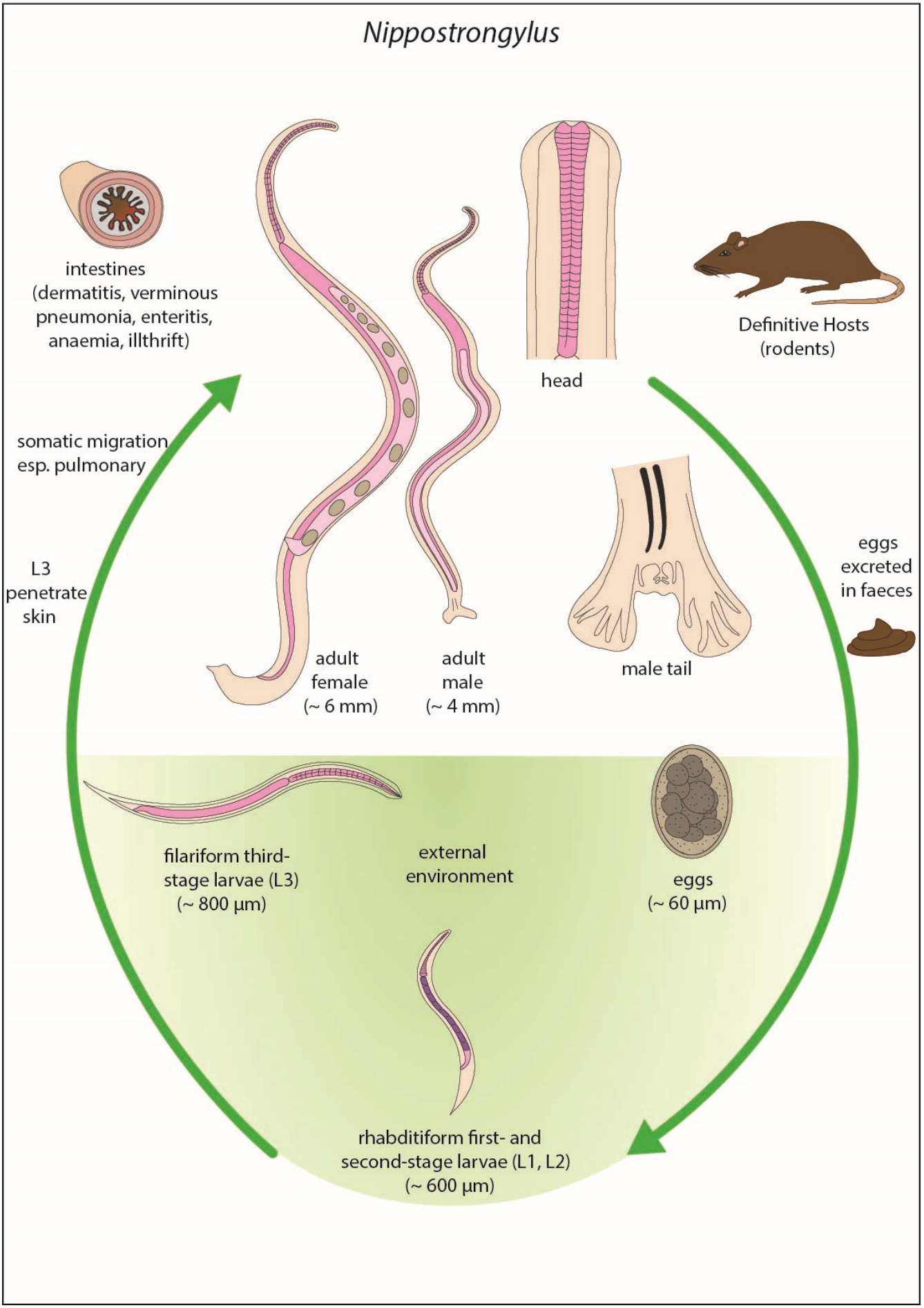
Developmental cycle and mode of transmission: *Nippostrongylus* spp. have direct monoxenous life-cycles involving faecal-oral or transdermal transmission of infective larvae. Gravid female worms may lay up to 1,000 eggs per day which are excreted into the external environment with host faeces. The eggs embryonate in 18-24 hours and then hatch releasing rhabditiform L1 which subsequently moult to rhabditiform L2 and then filariform L3. Free-living larval stages can tolerate a wide range of ambient temperatures (from 7-50°C), but they are highly susceptible to desiccation. In optimal conditions (22-30°C with high humidity), L3 may form as early as 36-48 hours after egg excretion, but take longer in suboptimal conditions (5-6 days). L3 are ensheathed when first formed, but the sheath gradually contracts and splits longitudinally over several days allowing the enclosed larvae egress. Freed L3 are active stages exhibiting both horizontal migration (for dispersal) and vertical migration (ascending vegetation). Infective L3 either ingested orally or they penetrate the skin of the host (transdermal or percutaneous infection). The larvae migrate through superficial tissues and undergo somatic migration via the circulatory and/or lymphatic systems to various tissues (e.g. liver). However, most L3 undertake pulmonary migration to the lungs where they penetrate into alveoli and moult to L4 which ascend the trachea to be swallowed thereby accessing the intestines. Larvae may reach the lungs in as little as 11 hours and reside there up to 72 hours after infection. L4 may reach the intestines in as little as 41 hours but most take at least 72 hours. In the intestinal mucosa, L4 moult to young adults (sometimes designated L5) around 90-108 hours post-infection. They mature over several days and females first produce eggs 6-7 days after infection (= prepatent period) with greatest egg production occurring from 8-10 days. Adult worms usually do not persist long in the intestines as developing host immune responses lead to worm expulsion 7-14 days later, although a few adult worms may sometimes linger in hosts protected against challenge infections. Recurrent infections may become problematic in laboratory animal colonies where sanitation is poor.

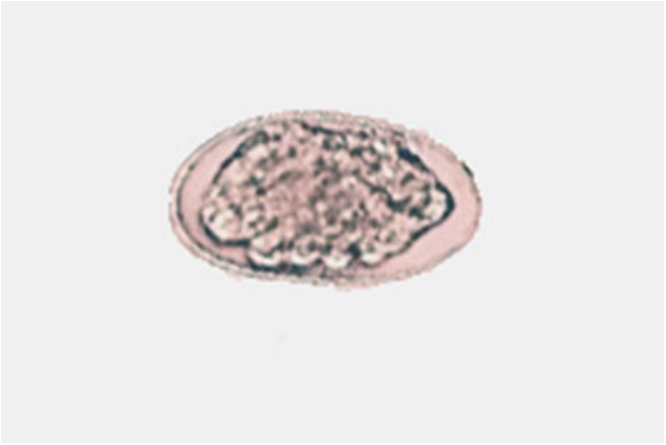
Differential diagnosis: Infections are rarely diagnosed on the basis of clinical symptomatology as many aetiological agents may cause similar clinical conditions (pneumonia, diarrhoea, anaemia, anorexia, failure to thrive). Coprological techniques are therefore used to detect worm eggs in faecal samples, usually following their concentration by sedimentation in water and/or floatation in

saturated sugar or heavy-metal salt solutions. Some techniques may be quantitated by counting the number of eggs in aliquots of diluted faeces in volumetric chambers (McMaster or Whitlock slides) to calculate the number of eggs per gram of faeces (epg). Worm egg counts provide a rough indication of worm burdens, even though they do not account for larval stages, male worms, or immature or senescent females. It is also difficult to differentially diagnose *Nippostrongylus* infections solely on the basis of egg morphology as the trichostrongyle eggs are similar in morphology to those of other nematodes (such as *Nematospiroides*). Recourse is sometimes taken to coprocultures whereby faecal samples are incubated for several days in moist conditions in order to harvest L3 for microscopic examination and identification by their characteristic features (size, longitudinal alae, compressed buccal cavity, filariform oesophagus, short tail). Infections may occasionally be diagnosed by the microscopic detection of developing worms in saline washes of tissue samples (liver, lungs, small intestines) collected at post-mortem. Infections in laboratory animal models have also been diagnosed using alternative technologies: including medical imaging (although X-rays tend to kill worms, especially males): ultraviolet spectroscopy (of faecal and gut samples taken from animals fed fluorescent dyes); and immunoserology (to detect specific host antibodies against worm antigens). Modern molecular biological techniques have also been used to characterize parasites after the polymerase chain reaction (PCR) amplification and sequencing of nuclear gene sequences (ribosomal RNA and internal transcribed spacer regions).

Treatment and control: Considerable experience has been gained in managing *N. brasiliensis* infections in laboratory rodents. Most infections are left untreated because host immune responses lead to the rapid expulsion of worms (self-cure phenomenon) and provide lasting protection against further infection. Nonetheless, a range of anthelmintic drugs have been tested, with benzimidazoles (mebendazole, fenbendazole, albendazole), imidazothiazoles (levamisole, tetramisole), diethylenediamines (piperazine) and tetrahydropyrimidines (pyrantel) found to be effective against enteric stages, and sometimes migrating larvae, without disrupting host immunity. Treatment was most effective when animals were provided with dietary supplements, notably protein and iron. Because hosts acquire solid protective immunity, a range of vaccination studies were conducted, with variable success in that they only stimulated partial protection and some demonstrated toxic side-effects. Various preventive strategies have also been used to help control infections, mostly in laboratory animal colonies where host and environmental management can reduce transmission (through improved hygiene, clean housing and bedding, food hygiene, dietary supplements, preventing unnecessary social interactions between animals and cohorts, and securing facilities against feral rodents).

Nippostrongylus



	
Nippostrongylus adult worm	*Nippostrongylus* worm egg