Chapter 14

Stem and gall forming nematodes

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14.1. Introduction

The Anguinidae Nicoll, 1935 includes mycophagous and plant-parasitic nematodes. The latter are obligate specialized parasites of higher plants, mosses and seaweeds on which they often induce swellings and galls. Several species: *Anguina agrostis*, *A. funesta*, *A. pacificae*, *A. paludicola* and *A. tritici*, *Afrina wevelli*, *Ditylenchus africanus*, *D. angustus*, *D. destructor*, *D. dipsaci*, *D. gigas* and *Subanguina radicicola* are considered of economic importance as agricultural and quarantine pests in various countries. The name for the family Anguinidae was first proposed by Nicoll (1935) to replace Anguillulinidae Baylis & Daubney, 1926, and it included genera that are now in other families of the order Tylenchida (Fortuner

& Maggenti, 1987). The modern concept of Anguinidae was first recognized and proposed by Paramonov (1962, 1970) at subfamily level with the genera *Anguina*, *Subanguina*, *Paranguina* and *Nothanguina*. Practically all nematologists recognize the family Anguinidae, discrepancies only concern its composition of subfamilies and genera. The validities and positions of several genera and subfamilies have been the subject of intensive discussions and speculation (Siddiqi, 1971, 1980, 1986, 2000; Brzeski, 1981, 1991; Chizhov & Subbotin, 1985, 1990; Fortuner & Maggenti, 1987) and remain unresolved. Comprehensive morphological and molecular analyses are still needed to understand relationships within these nematodes and replace the present artificial classification by a natural one reflecting evolutionary trends in this group. According to Siddiqi's (2000) classification, the Anguinidae contained two subfamilies Anguininae and Halenchinae, the latter includes exclusively marine, parasitic nematodes forming galls on sea algae.

14.2. Systematics

14.2.1. Subfamily Anguininae Nicoll, 1935

Diagnosis: (modified after Siddiqi, 2000)

Anguinidae. Adults from about 0.4-4.8 mm long, slender or obese. Labial region low and smooth. Muscular valvate median bulb present or absent. Basal pharyngeal bulb small or large, offset from intestine, or dorsal gland may become enlarged and extend over intestine as a lobe. Vulva generally at less than 85% of body length. Post-vulval uterine sac present, rarely absent. Ovary outstretched or flexed. Tail similar between sexes, female tail rarely subcylindrical, never cylindrical or hooked. Bursa variable from adanal to subterminal, never enclosing tail tip. Fungus feeders or parasites of plants, sometimes inducing swellings and galls.

Type genus: *Anguina* Scopoli, 1777

Other genera

Afrina Brzeski, 1981
Diptylenchus Khan, Chawla & Seshadri, 1969
Ditylenchus Filipjev, 1936
Heteroanguina Chizhov, 1980
Indoditylenchus Sinha, Choudhury & Baqri, 1985
Litylenchus Zhao, Davies, Alexander & Riley, 2011
Mesoanguina Chizhov & Subbotin, 1985
Nothanguina Whitehead, 1959

Nothotylenchus Thorne, 1941
Orrina Brzeski, 1981
Pseudohalenchus Tarjan, 1958
Pterotylenchus Siddiqi & Lenné, 1984
Safianema Siddiqi, 1980
Subanguina Paramonov, 1967

Important morphological and biological characters useful for distinguishing the genera are given in Table 14.1.

14.3. Morphology

Nematodes have a vermiform body and, in plant-parasitic species especially as a result of a strong development of the sexual system, the body becomes swollen so much that adult females are practically motionless (Fig. 14.1). The intra-specific variations of body length in plant-parasitic species may be significant; for example, in *A. agropyri* the measurements of adult individuals vary from 1.5 mm in the first generation to 4.8 mm in the second generation. Body length depending on host plant and numbers of specimens in a gall. With temperature fixation, males of almost all species straighten and only individuals more than 3 mm in length are sometimes bent ventrally. Body shape of females varies depending on the species and generation to which the female belongs; almost straight or C-shaped. Cuticle is finely annulated. Lateral field incisures vary from four to six in different species. They are usually constant in young females of the same species, but may disappear completely or reach up to 20 lines in the stretched cuticle of the stout, gravid females of the plant parasites. Labial framework always present but rather weak. Labial region with 3-5 annuli. Stylet short and thin. Procorpus cylindrical. Median bulb present with a valve for most genera but without a valve in *Nothanguina*, *Nothotylenchus* and *Orrina*. Pharyngeal glands may extend over intestine with nucleus lying anterior to pharyngo-intestinal junction or form a long lobe overlapping intestine with nucleus lying posterior to this junction (Table 14.1). Ovary outstretched or with one or two flexures, with one or several rows of oocytes in maturation zone. Crustaformeria of *Ditylenchus*, *Diptylenchus* and other genera forming a quadricolumella of four rows of four cells each, or, for example, in *Anguina* forming a long tube with a large number of cells in multiple irregular rows. Post-vulval uterine sac present in all genera, except for *Diptylenchus*. Bursa adanal to subterminal, never enclosing tail tip. Tail elongate-conoid to subcylindrical or filiform.

14.4. Life cycle

Temperature has a great influence on egg production, hatching and life cycle (Fig. 14.2). *Ditylenchus myceliophagus* thrives and breeds optimally at a temperature of

Table 14.1. Some comparative morphological and biologi cal characters for the genera of Anguininae.

Genus	Feeding	Type of symptoms on plants	Infective stage	Dorsal pharyngeal gland	Median bulb	Incisures in lateral field	Post-vulval uterine sac	Ovary	Oocyte arrangement	Crustaformeria structure	Tail
Afrina	Obligate plant parasites	Galls	J2	Not forming a long lobe overlapping intestine with nucleus lying anterior to pharyngo-intestinal junction	Muscular, valvate	4 or more	Present	Without or with one or two flexures	One row	4 rows, 14 cells per a row	Conoid
Anguina	Obligate plant parasites	Galls	J2	Not forming a long lobe overlapping intestine with nucleus lying anterior to pharyngo-intestinal junction	Muscular, valvate	4 or more	Present	With flexures	Two or more rows	6-12 rows, 20-60 cell per a row	Conoid
Diptylenchus	Fungal feeders	No symptoms	Juveniles and adults	Not forming a long lobe overlapping intestine with nucleus lying anterior to pharyngo-intestinal junction	Muscular, valvate	5	Absent	With flexures	One row	4 rows, 4 cells per a row	Conoid
Ditylenchus	Fungal feeders or plant parasites	Swellings and galls	Juveniles and adults	Not forming a long lobe overlaps intestine with nucleus lying anterior to pharyngo-intestinal junction	Muscular valvate	4-6 or more	Present	Without flexures	One row	4 rows, 4-7 cells per a row	Elongate conoid to filiform
Heteroanguina	Obligate plant parasites	Galls	J4	Not forming a long lobe overlapping intestine with nucleus lying anterior to pharyngo-intestinal junction	Muscular valvate	4 or more	Present	With or without flexures	One row	4-6 rows, 12-18 cells per a row	Conoid
Indoditylenchus	Probably, fungal feeders	No symptoms	Juveniles and adults	Not forming a long lobe overlapping intestine with nucleus lying anterior to pharyngo-intestinal junction	Muscular, valvate	4	Present	Without flexures	One row	No data	Elongate conoid
Litylenchus	Obligate plant parasites	Chlorotic spots on leaves	No data	Not forming a long lobe overlapping intestine with nucleus lying anterior to pharyngo-intestinal junction	Non-muscular, valvate	4	Present	With or without flexures	One row	4 rows, 4 cells per a row	Conoid
Mesoanguina	Obligate plant parasites	Galls	J3	Not forming a long lobe overlapping intestine with nucleus lying anterior to pharyngo-intestinal junction	Muscular, valvate	4 or more	Present	With or without flexures	Two or more rows	4-5 rows, 8-12 cells per a row	Conoid
Nothanguina	Obligate plant parasites	Galls	J2	Not forming a long lobe overlapping intestine with nucleus lying anterior to pharyngo-intestinal junction	Non-muscular, non-valvate	No data	Present	With flexures	Two or three rows	4 rows, more than 4 cells per a row	Conoid
Nothotylenchus	Fungal feeders	No symptoms	No data	Not forming a long lobe overlapping intestine with nucleus lying anterior to pharyngo-intestinal junction	Non-muscular, non-valvate	4-6	Present	Without flexures	One or two rows	4 rows, 4 cells per a row	Elongate conoid
Orrina	Obligate plant parasites	Galls	J4	Forming a long lobe overlapping intestine with nucleus lying posterior to pharyngo-intestinal junction	Non-muscular, non-valvate	4	Present	With flexures	One row	4 rows, 4 cells per a row	Conoid
Pseudohalenchus	No data	No symptoms	Juveniles and adults	Forming a long lobe overlapping intestine with nucleus lying posterior to pharyngo-intestinal junction	Muscular, valvate	4	Present	Without flexures	One row	4 rows , 4 cells per a row	Elongate conoid
Pterotylenchus	Obligate plant parasites	Galls	No data	Forming a long lobe overlapping intestine with nucleus lying posterior to pharyngo-intestinal junction	Non-muscular, non-valvate	4	Present	Without flexures	One row	4 rows, 8-9 cells per a row	Elongate conoid
Safianema	Fungal feeders and associates of insects	No symptoms	Juveniles and adults	Forming a long lobe overlapping intestine with nucleus lying posterior to pharyngo-intestinal junction	Muscular, valvate	6	Present	Without flexures	One or two rows	4 rows, 4 cells per a row	Elongate-conoid to filiform
Subanguina	Obligate plant parasites	Galls	J2	Not forming a long lobe overlapping intestine with nucleus lying anterior to pharyngo-intestinal junction	Muscular, valvate	4 and more	Present	With flexures	One or two row	4-5 rows, 12-20 cell per a row	Conoid

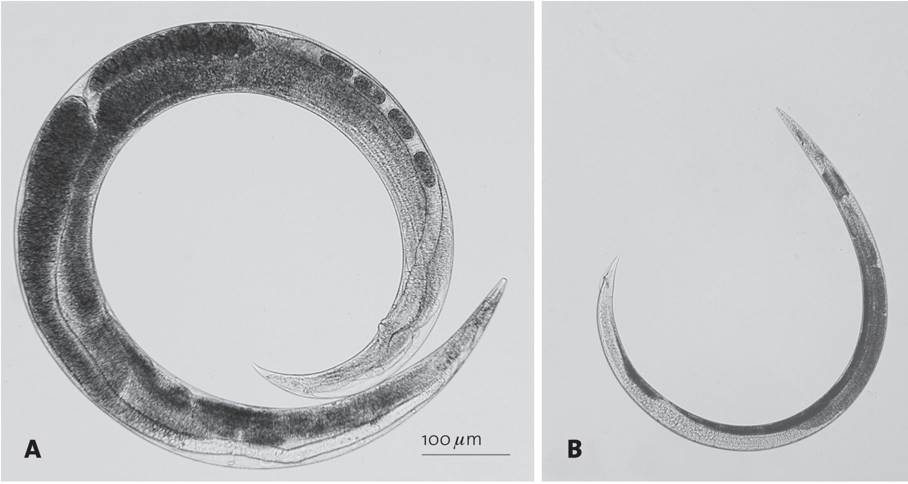

Fig. 14.1. *Anguina pacificae*. A: Female; B: Male. Courtesy of M. McClure.

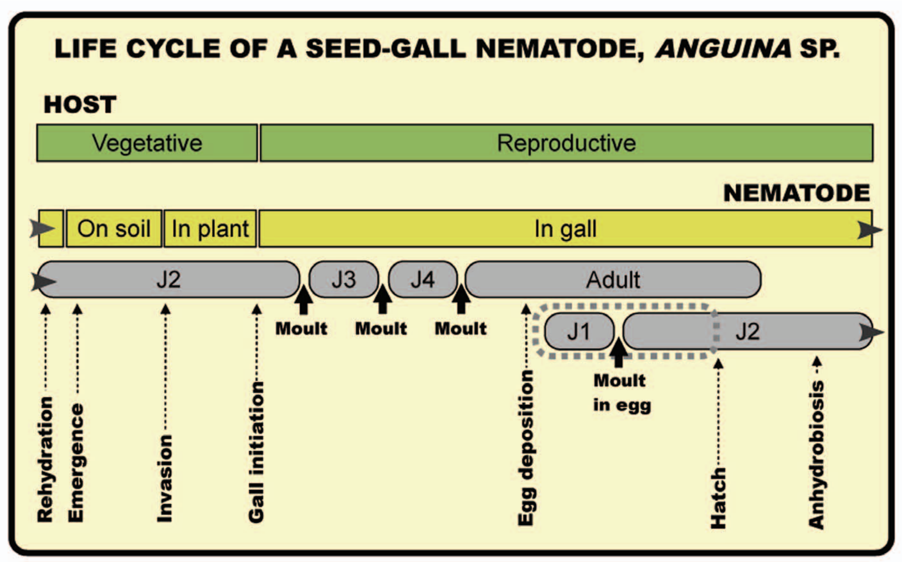

Fig. 14.2. Life cycle of seed-gall *Anguina* species, such as *A. funesta* and *A. tritici*. After Riley and BarbattiBarbetti (2008).

between 18 and 20°C. The life-cycle of *D. myceliophagus* is completed in 40 days at 13°C, 26 days at 18°C, and 11 days at 23°C. The life cycle of *D. dipsaci* ranged from 17 to 23 days at 13-22°C. Development of one generation of *D. destructor* takes 18 days at 27-28°C or 20-26 days, at 20-24°C. Optimal temperature for development of *D. africanus* is 28°C, its life cycle lasting 6-7 days. The life cycle of *A. pacificae* is completed in about 32 days at a mean temperature of 20°C (McClure *et al*., 2008). If the plant-parasitic *Ditylenchus* species complete multiple cycles within a season in plant tissues, the gall forming anguinins may likewise progress through one (most *Anguina* species), or two or more life cycles (*Anguina agropyri*, *Subanguina*, *Heteroanguina*, *Mesoanguina*) per season. Anguinins having several life cycles per season, either with one generation per gall, the progeny then leaving to induce a further cohort of galls (*Subanguina radicicola*, *Heteroanguina*), or with two generations per gall (*Mesoanguina*, *Anguina agropyri*).

The plant-parasitic anguinins are largely characterized by life cycles that have particular and robust survival and infective stages, these being consistent and specific for each species. The survival stages are able to enter a cryptobiotic (anabiotic) state and survive long periods of desiccation or freezing. For example, *D. myceliophagus*, *D. dipsaci* and *A. tritici* may remain viable more than 25 years in a cryptobiotic state. In a desiccated state they are heat and chemical resistant. Therefore many species are parasites of above-ground plant parts and are found in semi-arid environments with hot, dry summers. The survival stages normally exhibit only facultative quiescence and are revived by the onset of seasonal rains to become the infective stage. Survival and infective stages may be different in some species. In *A. australis*, which forms leaf galls on a rapidly maturing annual grass, the adults are the survival stage and after the galls re-hydrate in the autumn, eggs and then infective second-stage juveniles (J2) are produced. This is similar to the life history of *Anguina danthoniae*. In *Anguina microleanea* both eggs and J2, and in *Subanguina radicicola*, eggs, J2 and adults, can survive unfavorable conditions, but in these species only the J2 are able to infect plants. The infective stage is always a specific stage for these nematodes, for example, it is the J2 for *Anguina*, *Nothanguina* and *Subanguina radicicola*, the third-stage juvenile (J3) for *Mesoanguina* and the fourth-stage juvenile (J4) for some plant parasitic *Ditylenchus*, *Heteroanguina* and *Orrina*.

14.5. Symptoms

Plant-parasitic representatives of this family cause necrosis, swelling, deformation, distortion or galls on leaves, stems, inflorescence or roots (Figs 14.2-14.8). *Anguinins* forming galls induce intensive plant cell hypertrophy and proliferation. Mature galls contain a cavity lined with parenchymatous nutritive tissue comprising intercellular spaces and actively dividing hypertrophied cells (Fig. 14.9). These cells contained granular cytoplasm, hypertrophied nuclei, and brightly stained large nucleoli. Galls caused by some nematodes are pigmented.

Fig. 14.3. Galls caused by nematodes of the subfamily Anguininae. A: Galls on a stem induced by *Anguina woodi* in *Ehrharta villosa* var. *villosa*; B: Leaf galls induced by *Heteroanguina graminophila* on *Calamagrostis* sp.; C: Leaf galls induced by *Anguina graminis* on *Festuca rubra*; D: Leaf galls induced by *Mesoanguina millefolii* on *Achillea millefolium*; E: Bulbous galls induced by *Anguina agropyri* at the base of the stems of *Agropyron repens*; F: Root galls induced by *Subanguina radicicola* on *Poa annua*. B-F: Courtesy of V.N. Chizhov.

14.6. Karyology

Basic haploid chromosome numbers for *Anguina* and *Ditylenchus* are n = 9 and n = 12, respectively. Other species may be polyploid or aneuploid forms derived from these basic numbers (Triantaphyllou & Hirschmann, 1980; Sturhan & Brzeski, 1991). Haploid chromosome numbers for some species are as follow: *A. paludicola* – 18; *Heteroanguina caricis* – 18-19; *A. tritici* – 19; *A. funesta* – 22; *D. weischeri* – 26; *Ditylenchus gigas* – 24-30; *Mesoanguina plantaginis* – 27.

14.7. Phylogenetic relationships

The phylogenetic relationships between genera and species of Anguinidae have been studied by using morphological, biological and biochemical and molecular

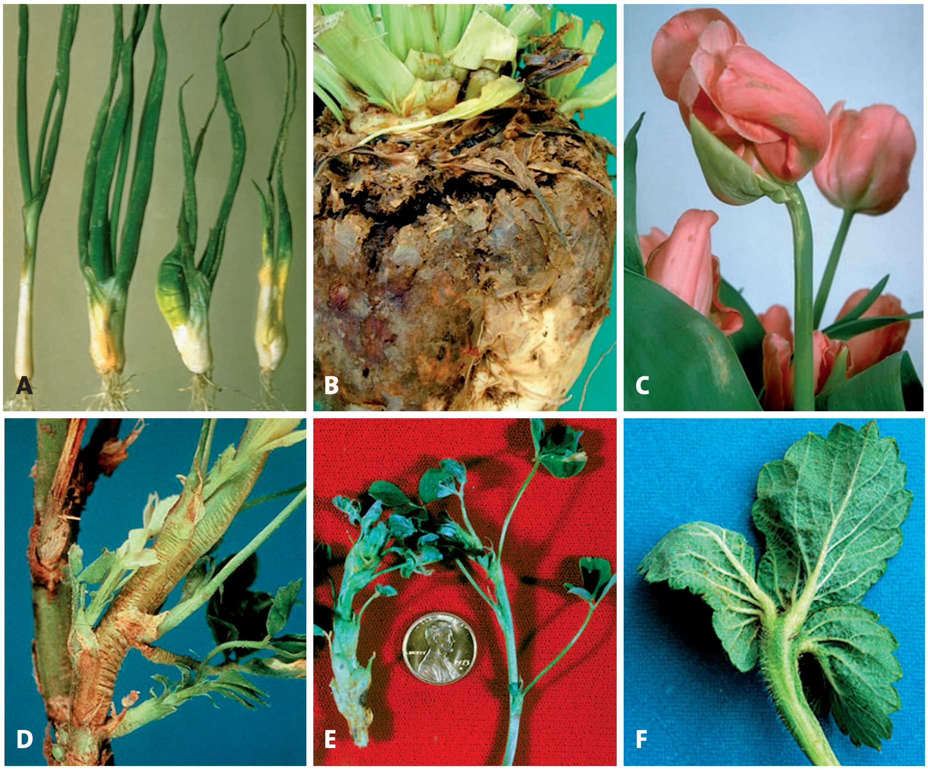

Fig. 14.4. Symptoms of infection of *D. dipsaci sensu lato*. A: Onion; B: Sugar beet; C: Tulip; D: Alfalfa; E: Red clover; F: Strawberry. Courtesy of EPPO (http://photos.eppo.org) (A-C), G.D. Jesperson (D); M. McCLure (E); V.N. Chizhov (F).

datasets. Relationships among some gall forming nematodes have been established on the basis of allozymes (Riley *et al*., 1988) and the rRNA gene sequences (Powers *et al*., 2001; Subbotin *et al*., 2004; Bert *et al*., 2008; Giblin-Davis *et al*., 2010; Zhao *et al*., 2011). Phylogenetic analysis using the ITS-rRNA gene sequence datasets supported, for example, monophyly of *Anguina*, and a narrow concept of *Subanguina* with but a single species, *S. radicicola*, as proposed by Paramonov (1970), and not the broad concept proposed by Brzeski (1981). However, phylogenetic analysis revealed paraphylies for *Mesoanguina* and *Heteroanguina*. Molecular analysis of seed gall nematodes supported the concept of a narrow trophic specialization in this group (Southey *et al*., 1990) and showed that *A. agrostis* occurred only with *Agrostis capillaris*, whereas other seed gall nematodes parasitizing grasses belonged to other, still undescribed, species (Fig. 14.10).

Fig. 14.5. Symptoms of infection caused by *Ditylenchus gigas* on *Vicia faba* L. A: Apical stems and leaves necrotised and deformed by severe attacks; B: Symptomatic range of necrotic areas on stems; C: Longitudinal sections of healthy (first on left) and damaged (two on right) stems showing internal necrosis; D, E: Deformed and undersized pods; F: Deformed (middle and bottom) and uninfected (top) seeds. After Vovlas *et al.* (2011).

14.8. Nematode-bacteria associations

A number of gall forming nematodes are found in association with species of coryneform bacteria of the genus *Rathayibacter*, previously placed in *Clavibacter* or *Corynebacterium*. *Rathayibacter* causes gumming diseases, usually characterized by yellow bacterial slime on seed-heads, stems and leaves of the plant host. In nature, this nematode – bacteria association appears to be specific and coevolved. For example, *Rathayibacter tritici* is found exclusively with *A. tritici* and *R. toxicus* with *A. funesta* and *A. paludicola*. The bacteria adhere to the cuticle of infective-stage juveniles in compatible interactions of bacterial adhesins and nematode receptors. However, cross-inoculation experiments and *in vitro* adhesions test show that, although some degree of specificity occurs, associations not found in nature can occur: *Anguina*

Fig. 14.6. Symptoms of infection caused by *Ditylenchus destructor* on potato (A). sweet potato (B) and carrot (C, D). A, C, D, - Courtesy of V.N. Chizhov.

australis and *A. tritici* have been shown to be potential vectors of some strains of *R. toxicus*. *Rathayibacter* spp. seem to be fully dependent on a nematode vector and direct inoculation attempts have only rarely been successful.

Although *Anguina* species can have an economic impact as agricultural pests in their own right, when they carry *Rathayibacter* the impact is increased. *Anguina tritici* reduces the yield of wheat and barley by replacing seed with galls. The admixture of galls also reduces grain quality, adding cleaning costs and limiting market access.

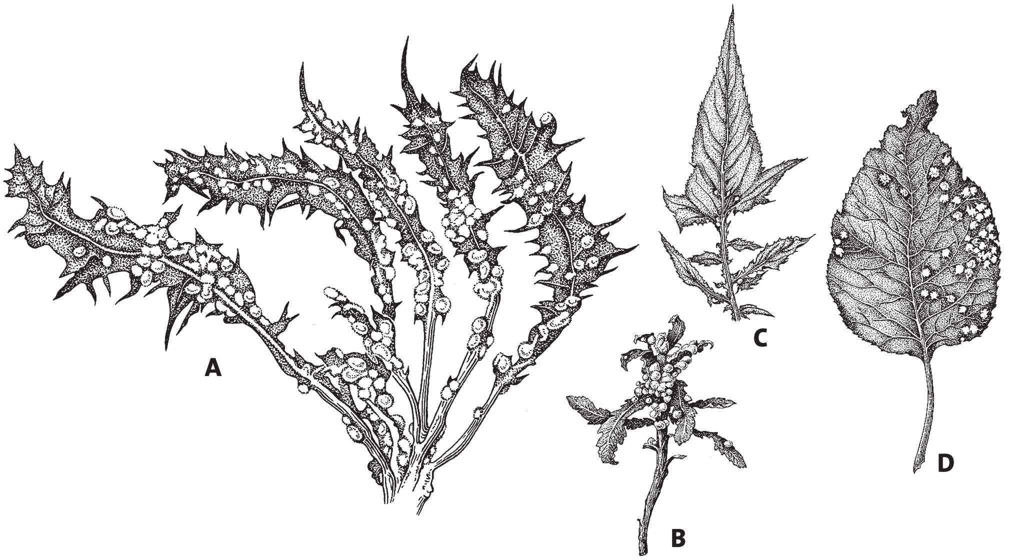

Fig. 14.7. Symptoms of infection caused by *Mesoanguina picridis*. A: *Cousinia onopordioides*; B: *Acroptilon picridis*; C: *Cousinia radians*; D: *Cousinia refracta*. After Kirjanova and Krall (1971).

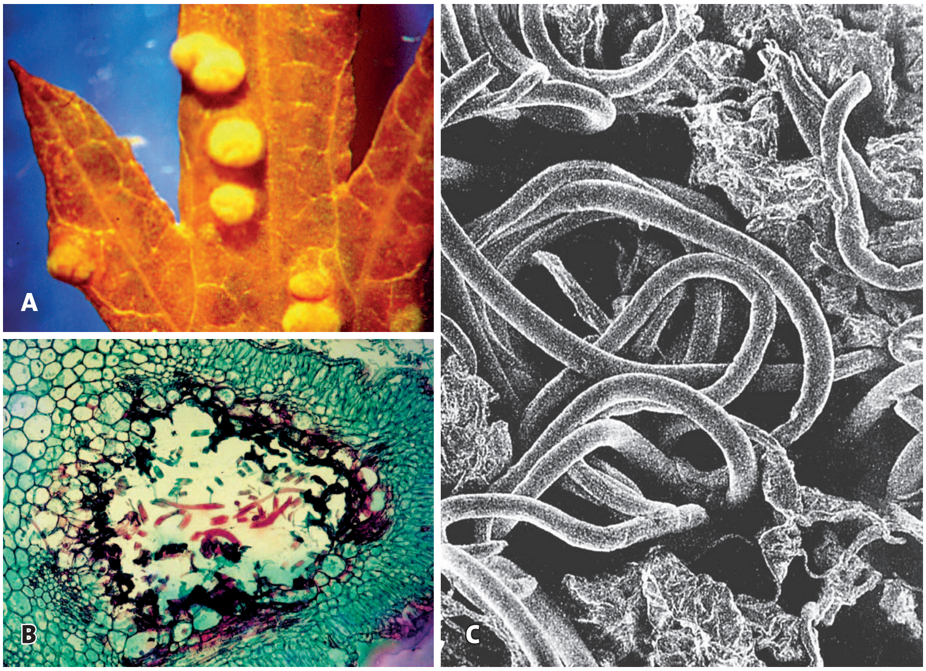

Fig. 14.8. *Mesoanguina moxae* on *Artemisia asiatica*. A: Leaf galls; B: Cross section of a gall; C: SEM of third-stage juveniles in galls. Courtesy of N. Vovlas.

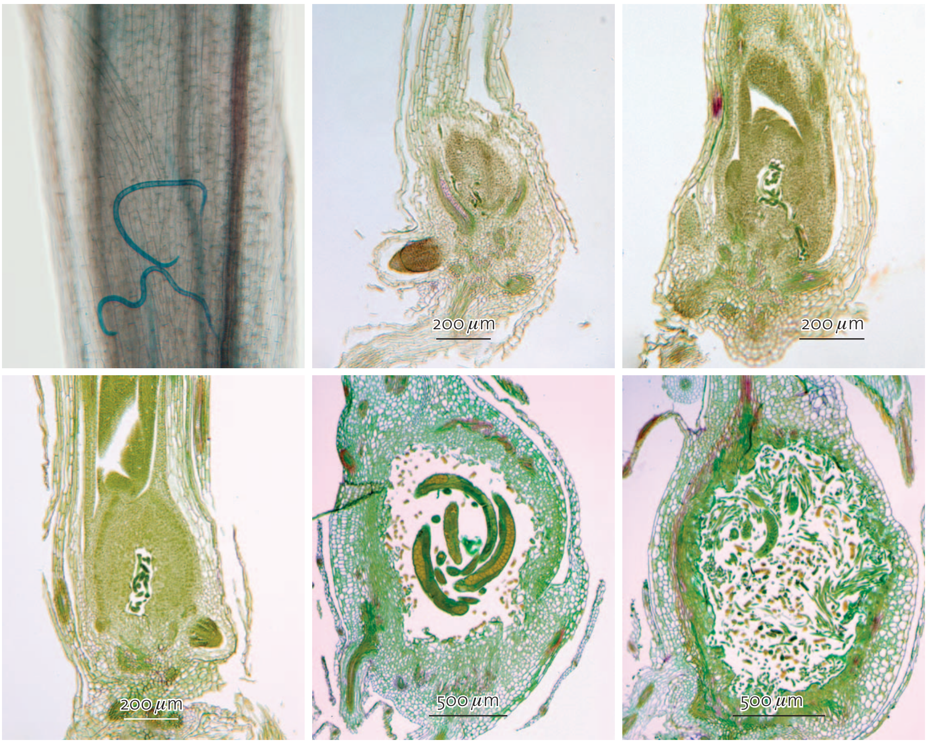

Fig. 14.9. Histopathology of *Anguina pacificae* on *Poa annua*. A: Infective J2 passing between leaves of the shoot, 2 mm above crown, 2nd day; B-F. Longitudinal sections through crown of plant; B: Fourth day, J2 have initiated a cavity just below the shoot meristem; C, D: Hyperplasia of shoot parenchyma cells, sixth and 15th days, results in visible swelling at base of stem. Cavity volume increases; E: Adults in cavity at onset of egg laying, 29th day. Cavity contains loose parenchyma cells and a few eggs; F: Eggs begin to hatch, 50th day, and a layer of cells, filled with cytoplasm and 3 to 6-cells thick, surrounds the cavity. After McClure *et al*. (2008) with modification.

However, in seed-heads concomitantly infected with *R. tritici*, seed set in florets not infested by the nematode will be prevented if the bacterial gummosis affects the whole inflorescence. A similar situation occurs for commercial seed production of the forage grass, *Dactylis glomerata*, when colonized by *R. rathayi* carried by its associated *Anguina* sp. However, in the case of *R. toxicustoxius* its vectors are of no direct economic impact, although when the bacterium colonizes the host grass it produces toxins that affect grazing animals. The bacterium *R. toxicus* is responsible for a gumming disease and ryegrass toxicity resulting in an often fatal poisoning of livestock in Australia. In addition to anguinid-*Rathayibacter* associations, other bacteria from *Curtobacterium*, *Microbacterium*, *Agreia* and *Leifsonia* belonging to the *Microbacteriaceae* have been isolated from nematode galls (Evtushenko *et al*.,

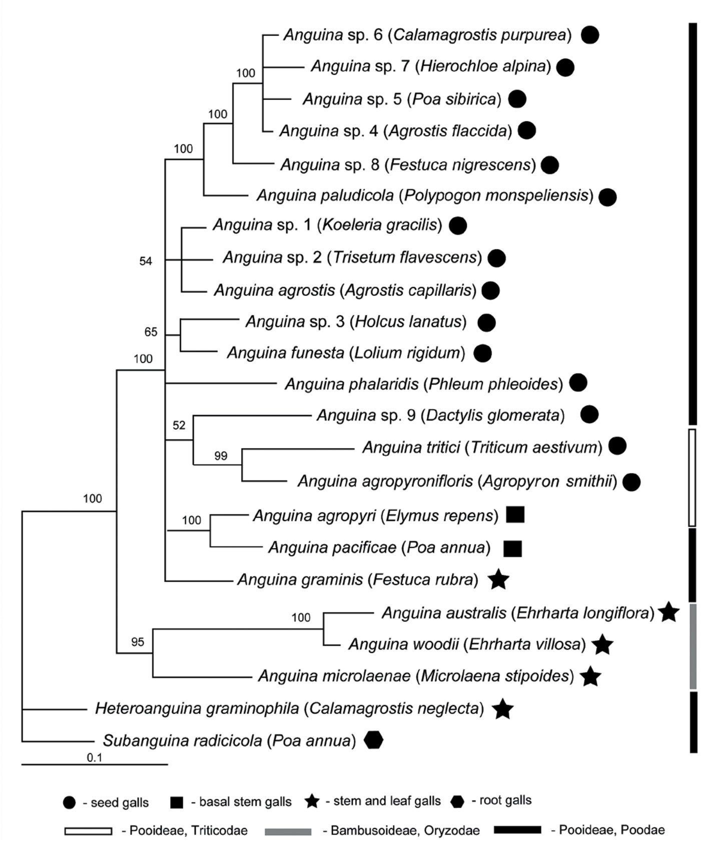

Fig. 14.10. Phylogenetic relationships within the genus *Anguina* as inferred from the Bayesian analysis of the ITS1 rRNA gene sequences with mapping gall types and systematic position of host plants (subfamily, tribe). Posterior probabilities are given on appropriate clades.

1994, 2001). The stem nematode, *D. dipsaci*, is known to increase the transmission of the actinobacterium, *Clavibacter michiganensis* subsp. *insidiosum*, responsible for a bacterial wilt in *Medicago sativa*, although it is not an obligate vector.

14.9. Nematode-fungal associations

Just as *Rathayibacter* spp. adhere to the cuticle of *Anguina*, so do the spores of the plant pathogenic fungus, *Dilophospora alopecuri*. Unlike the bacterium this fungus appears to be a true plant pathogen able to directly infect a host, albeit with limited aggressiveness. However, when spores are carried into the apex of the plant the infection is much more effective and the impact can be severe, completely inhibiting any seed set in the affected tiller and preventing the nematode from completing its life cycle. Although populations of *Anguina* vary in their receptiveness to adhesion by spores of *D. alopecuri*, there appears to be no species level specificity. *Dilophospora* is a monotypic genus and, although genetically variable, this is not strongly associated with vector specificity.

The fungal disease was important in wheat in Europe when *A. tritici* was common. Nowadays it is of limited importance, although it still occurs in some parts of the current geographic range of the vector, exacerbating its impact. Because the fungus is more aggressive than *R. toxicus* in its colonization of the host, it also acts as a more effective regulator of vector populations. It has been observed as a strong contributing factor to the decline of *A. funesta* populations in Australia and has been mass produced and applied as a biocontrol agent in areas where natural decline had not occurred.

14.10. Control

The gall forming nematodes have both a direct and indirect economic impact and so approaches to control vary depending on the species and the context. Galls of *A. tritici* are readily cleaned from cereal grain by the normal seed grading practised in mechanized agriculture. This, in conjunction with crop rotation, has provided highly effective control and has made the nematode effectively extinct over much of its former range. In small scale, traditional, farming systems in west and south Asia, the nematode persists as this control method cannot be as efficiently applied. Some work in India has focused on selecting resistant cultivars and on alternative seed cleaning methods.

For grass seed production, for example *Agrostis* and *Dactylis*, the impact of *Anguina* is mostly through reduction in seed germination, quality and market access (quarantine restrictions). In some grass-*Anguina* combinations, the infested diaspores are the same size and only of slightly lower mass than those with normal seed, so cleaning is unlikely to be fully effective. Therefore, to eliminate the nematode, complete removal of the host for a period and re-sowing with uninfested seed would be needed. Alternatively, early mowing or crash grazing can be effective because the nematodes do not reach their survival stage until well after initial tiller elongation. This is why grass seed production systems favor build up of the nematode, as the nematode is protected through to maturity. In contrast, in grazed pasture or hay production systems, development of the majority of nematodes is prevented.

For *A. funesta*, the host is a weed and so the nematode is able to reach maturity protected within the crop (Riley & BarbattiBarbetti, 2008). In this case, control has focused on eliminating the host from the crop with herbicides, but herbicide resistant populations of *Lolium rigidum* constrain this approach. Where the cropping system includes pasture leys, heavy grazing, mowing and/or herbicides in the pasture phase are used to control the nematode. As mentioned above, *D. alopecuri* is exploited for its biocontrol potential. Also, resistant *L. rigidium* cultivars are available but these are only adopted where the value of the grass as a pasture component is greater than its impact as a crop weed, and effective deployment depends on highly effective initial control of the naturalized host ecotypes.

Other gall nematodes have had minor, localized, inconsistent or transient impact, so control options have not been give as much attention as the *Anguina* spp. discussed above.

Most *Ditylenchus* species are fungal-feeders and considered to be beneficial members of soil ecosystems, however several are problematic plant pathogens and need to be managed in cropping systems. As these species can infest seeds and bulbs, primary control is based on selecting uninfested propagating material, especially, but not only, for planting in uninfested fields. Various treatments, such as hot water dips, fumigation and other chemical treatments, can reduce infestation levels and provide worthwhile control, but are unlikely to eliminate the risk of introducing these pests to uninfested fields. Where quarantine is important, a combination of sourcing propagating material from uninfested fields/regions, testing and treatments is necessary to provide an acceptable level of risk. In some host species, seeds are not infested, but harvested fodder (as in the case of *D. dipsaci* in *Avena sativa*) can be a means of spread, as can infested crop residues in other hosts and contaminated farm machinery. Irrigation water can also be an effective means of spread, as is the case with *D. angustus* in deep-water and low-land rice production.

In infested fields, crop rotation would be the first choice for control, but this relies on control of all potential hosts, including volunteers and weeds, in the non-host phase. For example, *Gallium aparine* (bedstraw) is a weed that can maintain *D. dipsaci* populations at problem densities between host crops. Where populations have been lowered by crop rotation, reinfestation from adjacent fields should be considered, as anguinid nematodes can be located on the soil surface and in above-ground crop residues, facilitating their dispersal by surface water flows and wind.

Given that field applied fumigants and nematicides are limited by efficacy, cost and environmental/regulatory concerns, breeding/selecting resistant crop cultivars is the other important approach to control plant-pathogenic *Ditylenchus* spp. Resistance to *D. dispsaci* has been identified and deployed in commercial cultivars for crops such as alfalfa (lucerne) and oats, but is complicated by variation in pathogenicity of the populations and races of the nematode. For the other plant-pathogenic species, progress has been more variable. Resistance found to *D. africanus* appears to fail in heavy infestations, indicating that tolerance and resistance may be needed for workable control. Therefore, incorporation of all or several (depending on the crop and pest species) of the above approaches into an integrated control program is likely to provide the most effective control.

14.11. Genera and major species

14.11.1. Genus *Anguina* Scopoli, 1777

= *Anguillulina* Gervais & Van Beneden, 1859
= *Paranguina* Kirjanova, 1955
= *Cynipanguina* Maggenti, Hart & Paxman, 1974

Diagnosis: (modified after Chizhov and Subbotin, 1990)

Female after heat relaxation crescentic or spirally coiled. Ovary reflexed two or three times. Oocytes in zone of maturation usually in two or more rows (in second-generation female of *A. agropyri* in a single row). Crustaformeria gland irregular, consisting of 180-600 cells in 6-12 rows (20-60 cells in each of it). Up to 18 synchronous eggs present in crustaformeria gland. Infective juveniles represented by J2. Inducing usually pigmented galls with an internal cavity on above-ground parts or sometimes on the rhizomes (underground stems) of monocotyledons of the family Poaceae (grasses), subfamilies Pooideae and Bambusoideae.

Type species: *A. tritici* (Steinbuch, 1799) Filipjev, 1936

Number of other species: 20

Adults of *Anguina* species are morphologically almost indistinguishable, although they develop on different hosts in widely separated geographical areas. A key to species identification is given by Chizhov and Subbotin (1990).

***Anguina tritici* (Steinbuch, 1799) Chitwood, 1935 – the wheat seed gall nematode**
(Figs 14.11; 14.12)

The wheat seed gall nematode was the first plant-parasitic nematode to be described in the literature. In 1743, the English theologian J.T. Needham examining seed galls of wheat observed a soft white fiber substance within the galls which, after adding water, started moving in a disorderly fashion. Needham named these unfamiliar animals as "worms, eels or serpents". J.G. Steinbuch was the first to

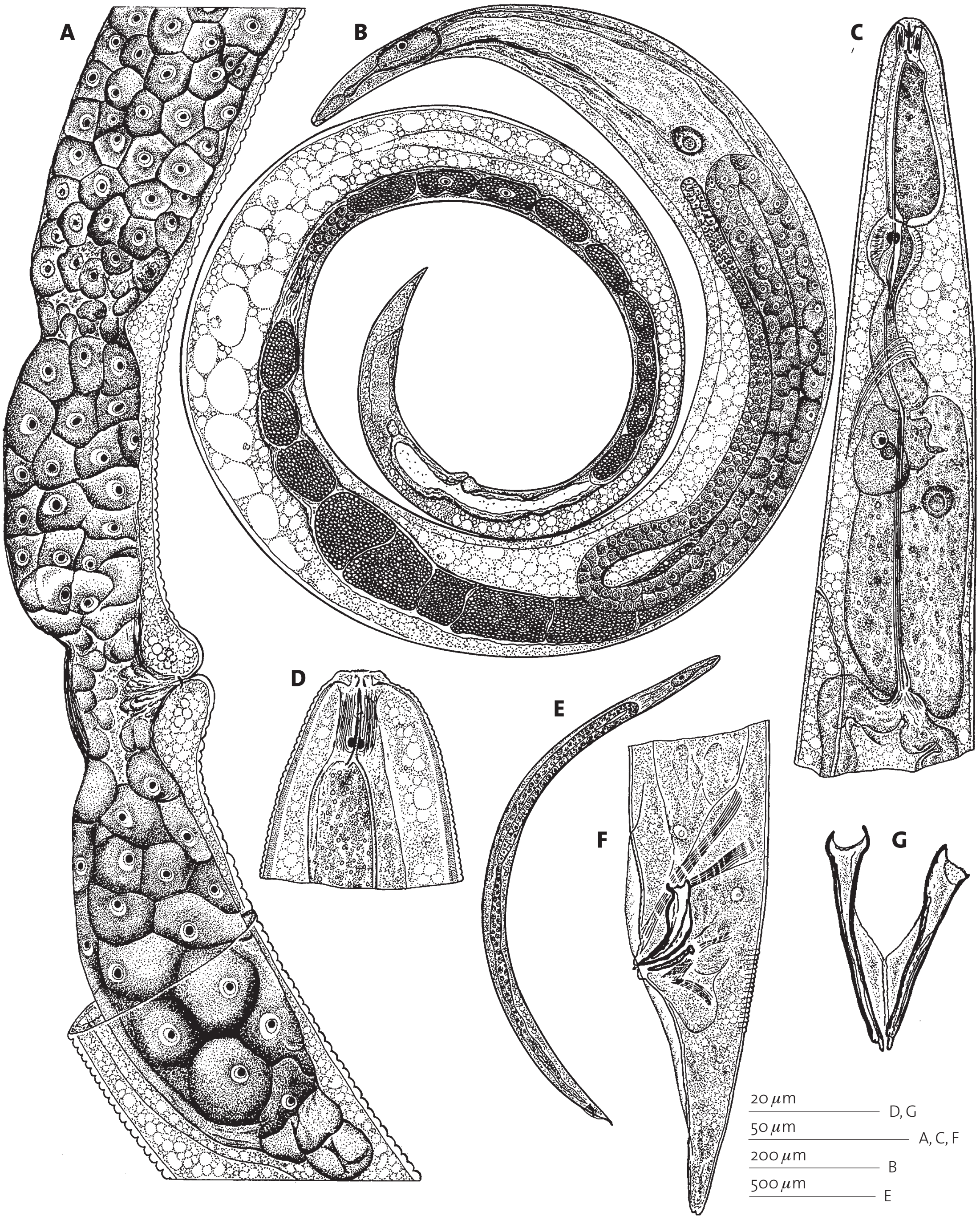

Fig. 14.11. *Anguina tritici*. A: Female; B: Male; C: Female pharyngeal region; D: Male tail; E: Spicules and gubernaculum. After T. Goodey (1932).

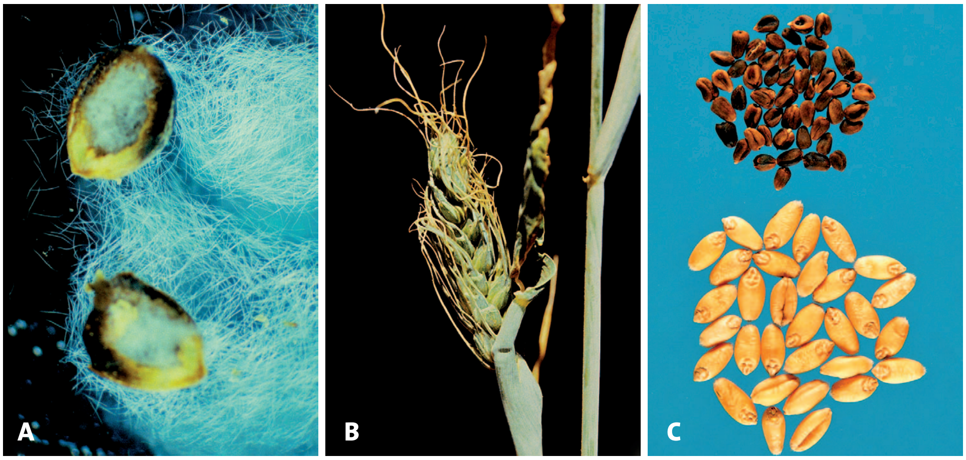

Fig. 14.12. *Anguina tritici*. A: Seed galls with J2; B: Infected wheat seed head; C: Seed galls and healthy seeds of wheat. A, B - Courtesy of M. McClure and V.N. Chizhov.

name this species, although an earlier description of this nematode was made by Roffredi in 1775. *Anguina tritici* has been recorded in major wheat-growing areas of all five continents, where it causes a disease in wheat and rye called 'ear-cockle' or seed gall. Application of effective mechanical seed-cleaning methods have practically eradicated this nematode from many countries where it was once common.

Measurements

After Goodey (1932), Kirjanova and Krall (1971) and Southey (1972).

Female: L = 2.6-5.2 mm; a = 13-30; b = 9.8-25.0; c = 24-63; V = 70-95; stylet = 8-11 μm.

Male: L = 1.9-2.5 mm; a = 21-30; b = 6.3-13.0; c = 17-28; stylet = 8-11 μm; spicules = 35-40 μm; gubernaculum = 10 μm.

J2 infective: L = 0.75-0.95 mm; a = 47-54; b = 4.0-6.3; c = 23-28; stylet = 10 μm.

Description: Female body obese, spirally coiled ventrally when relaxed by heat. Cuticular annules very fine, usually visible only in pharyngeal region; lateral fields of adults with four or more lines, visible only on young specimens. Procorpus swollen but constricted at junction with median bulb. Isthmus sometimes posteriorly swollen, then offset from glandular basal bulb by a deep constriction. Anterior branch of genital tract greatly developed; ovary with two or more flexures and very many oocytes in multiple rows around a rachis. Simple post-vulval sac present. Tail conoid, tapering to an obtuse or rounded tip, not mucronate. Male body sometimes curved dorsally when killed by heat. Bursa extending from just anterior to spicules to just anterior to tail tip.

This nematode is characterized by the large size of the adults and form of the procorpus and median bulb of the pharynx compared with related species.

Type host: wheat, *Triticum aestivum*.

Other hosts: *Secale cereale*, *T. dicoccum*, *T. durum*, *T. monococcum*, *T. spelta*, *T. ventricosum*. Oats and barley are poor hosts (Krall, 1991).

Symptoms and pathogenicity: Symptoms in infected plants are illustrated in Figure 14.12. The infective juveniles enter young seedlings and move upward within the plant to the meristem or concentrate in the leaf axis of shoots where they remain until the ears develop. Infected plants become stunted and bear shorter and deformed stems and leaves. Severely infested plants do not form ears or form only stunted ears on stunted stems. Infested ears are generally smaller, shorter and thicker than healthy ones, and have abnormally spreading glumes. Some or all of the grains are replaced by galls. The galls are light to dark brown to almost black in color, are hard to the touch and are filled with a mass consisting of infective juveniles. They may remain in anhydrobiosis for decades (Fig. 14.12A). Each gall initially contains 40 or more adults of each sex which produce 30,000 or more eggs and/or juveniles. Yield loses up to 70% have been reported in wheat grain.

Distribution: Formerly widespread in wheat-growing areas of Asia, Australia, Europe, North Africa, and North America. Earcockle disease has almost been eliminated in most countries. Outbreaks of the disease have been reported more recently in India, north Africa and west Asia.

Disease complex: Wheat seed gall nematode infection is associated with a bacterial yellow gumming disease caused by *Rathayibacter tritici*, which is especially important in India, where it has been known for many years as 'tundu'. The characteristic symptom is the presence of a bright yellow slime, consisting of massed bacteria, on the abortive ears and unsheathing leaves while the ears are still in boot. The fungal pathogen, *Dilophosphora alopecuri*, is also associated with *A. tritici* and causes diliophosphorosis of wheat, which is characterized by the spike being enveloped in a sticky black mass.

Control: The disease may be effectively controlled by seed cleaning and crop rotation.

Anguina agrostis (Steinbuch, 1799) Filipjev, 1936 – the bentgrass seed gall nematode

The bentgrass seed gall nematode was described by J.G. Steinbuch in 1799, from specimens of a grass collected in a wood in the vicinity of Erlangen, Bavaria, Germany. He thought that that grass belonged to *Agrostis silvatica* Huds., but after dissection of the enlarged structures of the inflorescence he discovered parasitic nematodes and came to the conclusion that the grass was actually *Agrostis capillaris*. Another species, now known as *Anguina phalaridis*, and from *Phleum phleoides*, was also described together with the bentgrass seed gall nematode. There are numerous reports of galls found in various species and genera of grasses, but without satisfactory diagnostic descriptions of the associated nematodes. The grasses *Apera*, *Arctagrostis*, *Calamagrostis*, *Dactylis*, *Eragrostis*, *Festuca*, *Hordeum*, *Koeleria*, *Lolium*, *Phalaris*, *Phleum*, *Poa*, *Puccinellia*, *Sporobolus* and *Trisetum* have been included in the list of host plants for *A. agrostis* (Kirjanova & Krall, 1971). Southey (1973), after reviewing literature on the host specificity of seed-gall nematode populations and their morphological and morphometric differences from the type host, suggested that a thorough revision of species causing galls in the flowers of grasses was required. He suggested that *A. agrostis sensu stricto* appeared to be restricted to populations causing characteristic elongate galls and abnormally elongated floral structures in *Agrostis* grasses. Analysis of the ITS rRNA gene sequences supported Southey's suggestion and the concept of a narrow specialization of seed-gall nematodes and confirmed that *A. agrostis* infected *Agrostis capillaris* only. Other grasses known previously as hosts for *Anguina agrostis* are actually hosts for several other, still undescribed, species of *Anguina* (Subbotin *et al*., 2004). *Anguina agrostis* is considered to be a serious or potentially important nematode pest of bent grass, especially in the Pacific Northwest, USA, and New Zealand (Southey, 1973).

Measurements

After Goodey (1932), Southey (1973) and Chizhov (1980).

Female: L = 1.39-2.60 mm; a = 13.8-25.4; b = 8-28.7; c = 25.2-44.0; V = 87-92; stylet = 8-12 μm.

Male: L = 1.05-1.68 mm; a = 23.8-38.0; b = 6.5-9; c = 20.5-28.4; stylet = 10-12 μm; spicules = 25-40 μm; gubernaculum = 10-14 μm.

J2 infective: L = 0.55-0.82 mm; a = 47.2-65.0; b = 3.2-4.5; c = 11.7-20.0; stylet = 10 μm.

Description: Female body obese, coiled or crescentic when relaxed by heat with ventral surface inwards; tapered towards both ends. Procorpus cylindrical, somewhat enlarged in middle part with a constriction before joining median bulb. Ovary reflexed two or three times. Post-vulval uterine branch reaching halfway from vulva to anus, filled with sperm. Tail short, conoid, with acute terminus. Male smaller and more slender than female, body more or less arcuate ventrally when heat-relaxed, *i.e.*, dorsal surface outermost. Bursa subterminal, not reaching tail tip.

Type host: *Agrostis capillaris*.

Other hosts: In experimental condition *A. agrostis* infected several *Agrostis* species.

Life-cycle: Once in the plant crown, J2 remain between the leaf sheaths or rhizomes until spring, when floral primordia form. Upon invading the floret ovaries, J2 rapidly develop to adults and reproduce. Juveniles molt once in the egg stage, hatch quickly as J2 and become resistant before the galls dry. The life cycle requires 3-4 weeks and is often completed before the inflorescence opens.

Symptoms and pathogenicity: Forms characteristic cigar-shaped galls, at first green, later dark purple-brown, in florets of various species of bent grass (Fig. 14.13). Galled florets have the floral parts (glumes, lemma and palea) greatly elongated to three to five or more times normal length, the lemma projecting beyond the glumes as a sharp point (Hooper & Southey, 1978). Fresh galls of *A. agrostis* associated with *Rathayibacter* from *Agrostis capillaris capillaris* have been found to be poisonous to some animals.

Anguina agropyri **Kirjanova, 1955 – the couch grass gall nematode**

The couch grass nematode was described from *Elytrigia repens* (L.) Desv. ex Nevski. This nematode is distributed in several European countries and may damage some cereals.

Measurements

After Kirjanova and Krall (1971), Chizhov and Berezina (1986) and Krall (1991).

Female (*first generation*): L = 2.23-4.78 mm; a = 16.6-25.7; b = 14.5-24.4; c = 21.2-47.7; V = 84-90; stylet = 10-12 μm.

Male (*first generation*): L = 1.91-3.11 mm; a = 22.7-29.5; b = 10.6-21.3; c = 18.6-34.0; stylet = 10-12 μm; spicules = 33-40 μm; gubernaculum = 13-15 μm.

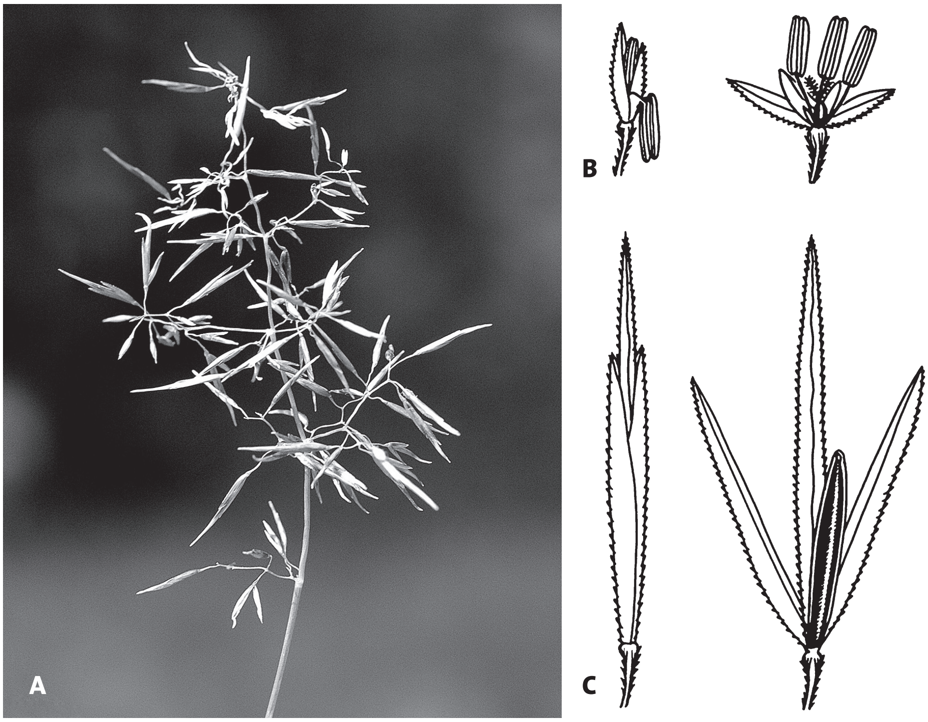

Fig. 14.13. *Anguina agrostis*. A: Seed galls induced by *Anguina agrostis* on *Agrostis* sp. Courtesy of V.N. Chizhov; B: Florets of *Agrostis stolonifera* showing effects of infestation by *Anguina agrostis*. Normal floret, unopened and normal floret, opened; C: Galled floret, unopened and galled floret opened to show dark cigar-shaped gall. After Goodey (1930).

J2 infective (*first generation*): L = 0.81-1.13 mm; a = 51.5-75.5; b = 3.8-5.2; c = 9.7-11.8; stylet = 10-12 μm.

Female (*second generation*): L = 1.94-3.23 mm; a = 26.0-36.5; b = 9.3-14.6; c = 16.9-23.2; V = 77-84; stylet = 10-12 μm.

Male (*second generation*): L = 1.53-2.27 mm; a = 28.6-44.0; b = 7.2-11.4; c = 14.0-21.0; stylet = 10-12 μm; spicules = 31-37 μm; gubernaculum = 13-15 μm.

J2 infective (*second generation*): L = 0.83-1.15 mm; a = 52.3-67.4; b = 4.0-5.2; c = 10.0-12.6; stylet = 10-12 μm.

Description: Female of first generation considerably more obese than vermiform adults of second generation. Cuticle with fine striations intercepting folds of the lateral fields. Procorpus swollen. Pharyngeal glands overlapping intestine only slightly. Ovaries reflexed. Post-vulval uterine branch long, extending up to halfway or more from vulva to anus. Tail conoid, tapering to a finely rounded tip. Male considerably more slender than females. Spicules elongate, almost straight. Bursa narrow, seeming to extend to tail tip.

Type host: couch grass, *Elytrigia repens*.

Other hosts: *Secale cereale*, *Triticum vulgare*, *Hordeum sativum* and *Elymus arenarius*.

Symptoms and pathogenicity: *Anguina agropyri* induces bulbous galls at the base of the stems of couch grass, and sometimes also on rhizomes (Fig. 14.3E). After nematode infection of cereals, the lower parts of the stems of wheat and rye become stout and swollen. In rye, small bulbous thickenings at the base of the stem may also be present. In the phase of development when the grains emerge in the tubule, the lower internodes of infested plants remain abnormally short and thickened. As a result of the severe reduction of the lower internodes, the leaves of infested cereals become bunched and rosette-like. Depending on the degree of infestation, the plants either do not form ears or give an extremely low yield, the number and weight of grains being much smaller than from uninfested plants. As a result of their stoutness, the infested shoots are easily broken. The grain yield can be estimated up to 46% of the average harvest in infested areas (Krall, 1991).

Anguina funesta Price, Fisher & Kerr, 1979 – the ryegrass seed gall nematode

Anguina funesta is a seed gall nematode found in *Lolium rigidum* and associated with annual ryegrass toxicity (ARGT) in Australia and South Africa. Although described as a new species by Price *et al*. (1979), Stynes and Bird (1980) considered that it was not morphologically distinguishable from *A. agrostis*. A growing body of biological, ecological, isozyme and molecular data eventually confirmed the validity of *A. funesta* as a distinct species. *Anguina funesta* is a vector for *Rathayibacter toxicus* and a key component of the multi-partite interactions that cause ARGT. The earliest known outbreaks of ARGT were in South Australia in the mid-1950s and it has remained an economic problem for over five decades. The nematode and associated organisms spread to Western Australia and South Africa and generally follow a pattern of build-up, impact, then decline. The decline seems to be associated with a subsequent build-up of *Dilophosphora alopecuri* and changes in land management practices that had supported high host densities protected through to seed set. As an economic problem, it now largely persists in the northern cropping areas of Western Australia.

Measurements

After Price *et al*. (1979), Stynes and Bird (1980) and Bird and Stynes (1981).

Female: L = 1.65-2.44 mm; a = 16.8-20.1; b = 9.3-34.0; c = 18.1-41.2; V = 87-94; stylet = 7-10 μm.

Male: L = 0.78-1.52 mm; a = 20.3-30.9; b = 6.3-9.5; c = 16.1-24.9; stylet = 7-10 μm; spicules = 16-28 μm; gubernaculum = 9-14 μm.

J2: L = 0.76-0.91 mm; a = 48-53; b = 4.2-5.3; c = 12-14; stylet = 9-11 μm.

Description: Female obese, coiled with ventral surface inwards; head narrower than body and offset. Wide procorpus opening to a muscular median bulb, ovate to spheroid, with three crescentic thickenings. Prodelphic ovary with one or two flexures. Post-vulval sac similar length to uterus, sometimes muscular or with small straight process. Male smaller than female, cylindrical, slender, straight to slightly curved dorsally when heat relaxed, with a single reflexed testis.

Type host: annual ryegrass, *Lolium rigidum*.

Other hosts: Inoculation experiments demonstrated that the host range includes various *Lolium*, *Festuca* and *Vuplia* species, but only *L. rigidum* and *V. myuros* are known as hosts from natural infestations.

Symptoms and pathogenicity: The J2, having over-summered anhydrobiotically in last season's galls, invade host seedlings following the onset of autumn rains. The juveniles congregate at the vegetative meristem and induce galls after floral initiation. The galls are smaller and lighter than the seed they replace and there are no morphological changes to the inflorescence as seen with *A. agrostis* and *A. paludicola*. Infested florets can only be detected by examining diaspores with transmitted light under a microscope or on a light table. Although *A. funesta* will reduce seed set in the host, galled florets are mostly removed with normal seed cleaning procedures so direct economic impact comes through quarantine restrictions on trade (indirect impact is discussed below).

Distribution: *Anguina funesta* is found in winter rainfall areas of southern Australia, especially in pasture-ley farming systems with *Lolium rigidum* as the main grass weed in the cereal cropping phase. It is most common in South Australia and Western Australia, but also occurs in Victoria. It spread to South Africa, most likely in infested seed from Australia, but did not spread as quickly or cause as much economic impact. Given that known hosts are largely of Mediterranean origin, it is likely *A. funesta* evolved with these hosts in the same region.

Disease complex: *Anguina funesta* is vector for *R. toxicus* and *D. alopecuri*. *R. toxicus* colonizes the galls and gums the seed head of the host, causing distortions. On senescence of the host, the bacterium produces toxin, paralleled by bacteriophage multiplication. The glycolipid toxins inhibit glycosylation of proteins, which compromises membrane integrity leading to neurological symptoms and finally death of the grazing animal. The toxins are cumulative and, as symptoms are not usually observed in livestock before a lethal dose is consumed, mortality rates can be high.

Control: Control has focused on the host. The nematode invades tillers produced early in the growing season so effective control is achieved by preventing these tillers from maturing by crash grazing, mowing or herbicide in the pasture phase. In cereal crops, herbicides often only reduce the host density, so that the nematode population is concentrated in fewer, larger plants and thus nematode control is only partially achieved. Nematode resistant ryegrass cultivars have been released but are not widely adopted. *Dilophosphora alopecuri* appears to contribute to natural decline in ARGT outbreaks in some areas and has been mass produced for application in other areas in order to accelerate this decline.

Anguina pacificae **Cid del Prado & Maggenti, 1984 – the Pacific shoot gall nematode,** (Fig. 14.1)

The Pacific gall nematode was found on grasses on *Poa annua* in 1978 from several golf courses along the Coast of Central California, USA and was initially identified as *S. radicicola*. Subsequently, it was characterized morphologically and described as a new species by Cid del Prado Vera and Maggenti (1984). *Anguina pacificae* is distributed along a narrow strip on the Pacific coast of Northern California where it forms galls on the shoots of *Poa annua* and causes significant damage to golf course greens.

Measurements

After Cid del Prado Vera and Maggenti (1984).

Female: L = 1.44-2.58 mm; a = 19.4-33.2; b = 8.1-13.1; c = 19.9-34.4; stylet = 8.9-12.4 μm; V = 82-89 μm.

Male: L = 1.22-1.84 mm; a = 21.1-33.0; b = 5.8-10.4; c = 19.8-29.8; stylet = 9.3-12.7 μm; spicules = 26.2-40.7 μm; gubernaculum = 10.6-15.5 μm.

J2 infective: L = 0.63-0.97 mm; a = 37.0-56.4; b = 3.5-7.3; c = 8.8-11.4; stylet = 10.0-12.7 μm.

Description: Body cylindrical, assuming C-shape after death. Labial region without visible annuli by light microscope, not offset. Lateral field with four main distinct lines, and many longitudinal fine lines between them. Ovary with one or two flexures, sometimes straight and extending to pharynx Post-uterine sac containing degenerate cells. Vulval lips distinct. Tail conical, sharply pointed tip. Male body cylindrical, arcuate with ventral surface outermost after killing. Bursa beginning anterior to spicules and ending before tail tip. Tail with acute terminus.

Anguina pacificae differs from *A. agropyri* by the pharyngeal glands being well separated from the median bulb in the females. They also differ in the position of the excretory pore; in *A. agropyri* it is situated a little anterior to the pharyngo-intestinal valve, and in *A. pacificae* the excretory pore is situated at the level of posterior end of the isthmus. *Anguina pacificae* has a sharply pointed tail tip, while *A. agropyri* has a conical tail tip.

Type host: *Poa annua* L.

Other hosts: *Poa trivialis* and several *Agrostis* cultivars proved to be poor hosts for *A. pacificae* (McClure *et al*., 2008).

Symptoms and pathogenicity: The nematode induces galls on stems. Initial symptoms on turf consist of small yellow patches, 25-75 cm in diameter, which enlarge and may coalesce as the nematodes spread. Young, infected plants may die and, when the infestation is severe, a rough, uneven putting surface results. *Anguina pacificae* is considered as a major threat to turf in coastal areas of California (McClure *et al*., 2008).

Anguina paludicola Bertozzi & Davies, 2009

Anguina paludicola is a seed gall nematode described by Bertozzi and Davies (2009) from two hosts from areas subject to occasional seasonal flooding in eastern Australia. In *Polypogon monspeliensis* (type host, non-native grass), it occurs in the southeast of South Australia where pastures can be subject to extended, annual flooding. In this host, shoot meristem galls are also formed in inundated plants. In *Lachnagrostis filiformis* (an Australian native grass), it occurs along the inland river systems of northern New South Wales in semi-arid pastures subject to transient, infrequent flooding. Morphological differences occur between the populations but are variable and not considered as justification to separate at the species level. In both hosts and areas, the nematode acts as a vector for *Rathayibacter toxicus*, which colonizes the nematode galls and grass seed heads, producing a toxin responsible for poisoning and death of livestock.

Measurements

After Bertozzi and Davies (2009)

Female: L = 1.51-4.40 mm; a = 18.8-33.2; b′ = 8.5-22.1; c = 21.8-54.0; V = 81-94; stylet = 9-13 μm.

Male: L = 1.50-2.49 mm; a = 18.3-37.3; b′ = 8.9-14.4; c = 18.0-35.3; stylet = 8-12 μm; spicules = 27-37 μm; gubernaculum = 7-13 μm.

Second stage juveniles: L = 603-771 mm; a = 49.0-60.8; b′ = 3.3-5.1; c = 9.9-15.2; stylet = 9-11 μm.

Description: Female obese, coiled with ventral surface inwards; head narrower than body and offset. Procorpus wide, metacorpus oval with refractive thickenings slightly anterior to middle, isthmus shorter and narrower than procorpus. Ovary usually reflexed twice. Post-vulval uterine sac similar in length to uterus, frequently with a reflexed process. Tail conical with irregular and variable terminus, appearing lobed in some specimens (diagnostic feature of the species). Male smaller than female, cylindrical, slender, C-shaped when heat relaxed with dorsal surface inwards, with a single reflexed testis.

Type host: *Polypogon monspeliensis*.

Other hosts: *Lachnagrostis filiformis*.

Symptoms and pathogenicity: In populations of *P. monspeliensis*, *A. paludicola* juveniles invade the host with the onset of autumn rains and rapidly forms galls in the shoot meristems, a complete life cycle being completed before the field is flooded in winter. The new cohort of juveniles invades tillers that develop as flooding recedes to form seed galls and a small proportion of panicle branch galls. When these galls mature they contain a second cohort of J2 that over-summer in an anhydrobiotic state. In *L. filiformis*, *A. paludicola* only induces seed galls and has one cycle per year. In both hosts, the seed galls formed are larger than normal seed and the floral bracts are enlarged five times or more (McKay *et al.*, 1993).

Distribution: This species is found in Australia but only in situations where flooding allows its host grass to become the dominant species in the sward. It is found in South Australia in the introduced weedy grass, *P. monspeliensis*, and in New South Wales in the native grass, *L. filiformis*.

Disease complex: *Anguina paludicola* acts as a vector for both *R. toxicus* and *D. alopecuri*. As with the association of *A. funesta* and *R. toxicus*, toxins are produced and grazing animals poisoned (the syndrome in this case is called flood plain staggers or FPS). The bacterium adheres to invasive juveniles and is carried into the host, colonizing nematode galls and whole inflorescences, and in the latter situation causing gummosis and distortion. In the early 1990s, serious stock losses were recorded, which prompted research to identify the cause and develop controls. It now appears that conditions favoring the high nematode population densities and bacterial infection are relatively uncommon, and major stock losses have not been recorded since. *Dilophospora alopecuri* is likewise carried into the plant with *A. paludicola*, producing leaf lesions and distorted floral development.

Control: Bertozzi and Davies (2009) investigated control options, mostly focused on host control and some consideration of the biological control potential of *D. alopecuri*. However, given the infrequent occurrence of major outbreaks of toxicity, there has been no impetus to apply or further develop control measures. Also, in the southeast of South Australia, drainage schemes have reduced the winter flooding of pasture and hence the conditions that favor high population densities of *A. paludicola*. In northern New South Wales, extended periods of drought have dominated climatic patterns for a decade or more.

14.11.2. Genus *Subanguina* Paramonov, 1967

Paramonov (1967, 1970) proposed *Subanguina*, with *S. radicicola* as type species, and stated that it retained the ancestral type of ontogeny. *Subanguina radicicola* is the only species of the Anguinidae that forms galls on roots of many grass species. Based on similarity in form of the crustaformeria structure, Brzeski (1990) transferred 17 species to this genus. However, molecular phylogenic analysis using the ITS rRNA sequence data (Subbotin *et al*., 2004) supports the narrow concept of *Subanguina* by Paramonov (1970) and Chizhov and Subbotin (1990) and not the broad concept proposed by Brzeski (1990).

Diagnosis: (after Chizhov and Subbotin, 1990)

Adult specimens almost straight after heat relaxation. Ovary outstretched or with one to two flexures. Oocytes in the zone of maturation in a single row. Crustaformeria consisting of 48-80 cells in four to five rows, 12-20 cells in each, and may contain up to four synchronous eggs. Infective juveniles of the second stage. Induce hook-like galls with well-developed internal cavity on roots of grasses. Only one generation developing in gall.

Type and only species: *S. radicicola* (Greeff, 1872) Paramonov, 1967.

***Subanguina radicicola* (Greeff, 1872) Paramonov, 1967 – the grass root gall nematode** (Fig. 14.14)

Subanguina radicicola causes characteristic root galls on graminaceous hosts. It damages golf courses sown with *Poa* species, and barley, rye and other cereals. It also occurs on grasses in various regions of the world.

Measurements

After Goodey (1932) and Chizhov and Marjenko (1984).

Female: L = 1.11-3.15 mm; a = 26.2-37.6; b = 6.8-13.8; c = 13.5-21.5; V = 75-81; stylet = 12-16 μm.

Male: L = 0.86-2.04 mm; a = 26.0-36.9; b = 6-10.1; c = 13.6-20.8; stylet = 12-16 μm; spicules = 25-31 μm; gubernaculum = 6-11 μm.

J2 infective: L = 390-530 mm; a = 24.7-40.0; b = 3.4-4.1; c = 6.4-7.7; stylet = 10-11 μm.

Description: Female labial region offset. Cuticle finely annulated with 10-12 incisures in lateral field. Procorpus cylindrical, median bulb ovoid. Isthmus short and narrow. Pharyngeal glands as a wide bulb slightly overlapping the intestine dorsally. Ovary simple or once or twice reflexed. In maturation zone, oocytes arranged in one row. Post-vulval uterine branch long, more than half vulva-anus distance. Tail tip sharply pointed. Male testis outstretched, sometimes with one flexure. Bursa not extending to sharply pointed tail tip.

Type host: meadow grass, *Poa annua*.

Other hosts: This species parasitizes roots of grasses belonging to 20 genera and more than 25 species, including rye, wheat, oats and barley.

Symptoms and pathogenicity: The J2 attack root tips of grasses and induce hypertrophy of parenchymatous tissue with subsequent formation of a cavity

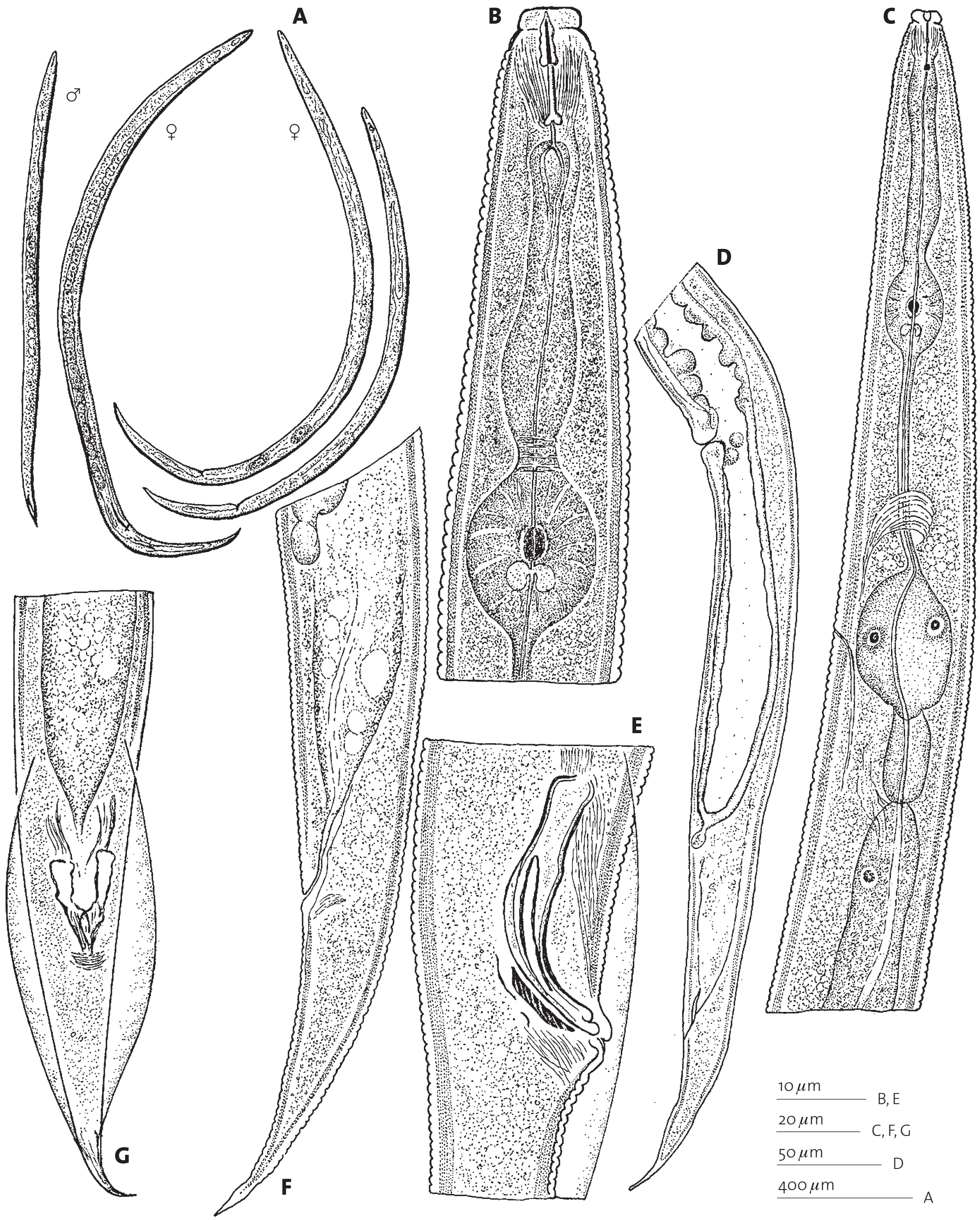

Fig. 14.14. *Subanguina radicicola*. A: Adults; B, C: Female pharyngeal region; D, F: Female tail end; E: Spicular region; G: Mail tail end. After Siddiqi (2000).

between the parenchyma and the root central cylinder. Extensive galling can occur throughout the plant roots, galls being slender and twisted in appearance and up to several millimeters long (Fig. 14.3F). Chlorotic patches may be observed on newly established *P. annua* golf courses. Only one generation develops in each gall but several subsequent generations may be observed per growing season.

Races: Five races are presently recognized: barley race; *Elymus* race; Saskatchewan race parasitizing Smith's couch grass; *Poa* race; Rhode Island race parasitizing *Ammophila*; and *Phalaris* race. The *Poa* race is the most widespread in Europe and New Zealand. A New Zealand population heavily infesting *P. annua* did not infect barley or wheat, whereas a Dutch population caused only a few galls on barley roots. The barley race and a Scandinavian population have been reported as pathogenic to cereals in northern Europe. The *Elymus* race attacks *Elymus arenarius* and may be transferred to barley, rye, wheat, and oat (Krall, 1991).

Distribution: Europe, North America and New Zealand.

14.11.3. Genus *Afrina* Brzeski, 1981

Brzeski (1981) proposed this genus to accommodate nematodes having a crustaformeria of four rows with 14 cells, a lobed bursa and mucronate tail in both sexes. He considered two species to belong to this genus: *A. hyparrheniae* and *A. tumefaciens*. Later, Van den Berg (1985) described *A. wevelli* and transferred *Anguina spermophaga* (Steiner, 1937) to Afrina.

Diagnosis: (After Brzeski, 1981, with modifications).

Mature Female swollen. Median bulb with refractive thickenings. Isthmus separated by a constriction from terminal bulb. Crustaformeria formed by four rows of cells, 14 cells in a row. Testes often with two flexures. Tail of both sexes conical, tail tip mucronate. Bursa notched at posterior end, almost reaching tip but does enclose a mucro. Forms seed and leaf galls on grasses of subfamily Panicoideae and Chloridoideae. Distributed in Africa, Australia and Asia.

Type species: *A. hyparrheniae* (Corbett, 1966) Brzeski, 1981.

Number of other species: 3

Afrina hyparrheniae (Corbett, 1966) Brzeski, 1981

This species was described from Malawi and was associated with several *Hyparrhenia* species.

Measurements

After Corbett (1966) and Van den Berg (1985).

Female: L = 1.51-1.84 mm; a = 18-29; b = 7.0-11.3; c = 25-34; V = 90-92; stylet = 6.6-8 μm.

Male: L = 1.26-1.39 mm; a = 31-39; b = 6.8-9.6; c = 27-35; stylet = 6-7 μm; spicules = 32-40 μm; gubernaculum = 13-18 μm.

J2 infective: L = 0.66-0.80 mm; a = 43-56; b = 4.9-6.5; c = 12.9-16.6; stylet = 6-8 μm.

Description: Female body tightly coiled ventrally. Lips flattened, smooth. Lateral field rarely seen, with four to up to 10 incisures. Procorpus wide, median bulb ovate. Cardia distinct. Genital tract outstretched, rarely with one small flexure. Post-vulval uterine branch large, 1-1.5 vulval body width long. Tail terminus acutely pointed. Spicules massive, curved. Gubernaculum trough-like, curved. Bursa rejoining tail tip after forming two lobes which may overlap conspicuous ventral process beyond terminus.

Type host: *Hyparrhenia collina* (Pilger).

Other hosts: Tropical grasses of the genus *Hyparrhenia*.

Distribution: Southeastern part of central Africa: Malawi, Mozambique, Zambia.

Symptoms: The earliest sign of the disease is a crinkling of the edge of the young leaves followed by initiation of seed gall formation. Galls are shriveled, light greenish brown to a dark olive in color, and much smaller than the light brown seeds. It is most easily recognized later by the severe clumping of the inflorescences into 'witches' brooms'.

***Afrina tumefaciens* (Cobb, 1932) Brzeski, 1981 – the Bradley grass gall nematode**

The Bradley grass nematode was described from *Cynodon transvaalensis* collected from lawns in the vicinities of Johannesburg and Pretoria, Transvaal, South Africa.

Measurements

After Van den Berg (1985).

Female: L = 1.38-2.06 mm; a = 20.1-39.3; b = 6.7-10.9; c = 17.0; V = 86-92; stylet = 8.8-10.7 μm.

Male: L = 1.35-2.03 mm; a = 23.7-34.6; b = 7.1-8.0; c = 15.0-24.6; stylet = 8.0-11.1 μm; spicules = 36.8-47.4 μm; gubernaculum = 18.0-21.3 μm.

Second stage juveniles: L = 743-775 mm; a = 38.9; b = 5.9-6.0; c = 13.5; stylet = 9.2-9.6 μm.

Description: Female body posture ranging from an open C to a complete circle. Body narrowing rapidly posterior to vulva. Labial region flattened with indistinct annulation. Procorpus enlarged, median bulb rounded. Basal bulb large and irregular, slightly overlapping intestine. Ovary single or double reflexed. Short post-vulval uterine branch present. Tail tapering to an acute terminus. Bursa extending from about one cloacal body width anterior to cloaca to tail tip. Male tail ending in an irregular process.

Type host: Bradley grass, *Cynodon transvaalensis*.

Distribution: South Africa.

Symptoms: Induces ovoid galls 2-8 mm long on stems, leaves, and in flower heads. Infested plants are stunted and may die out in lawns.

***Afrina wevelli* Van den Berg, 1985 – the weeping lovegrass seed gall nematode**
(Fig. 14.15)

Afrina wevelli was described from South Africa in 1985, from seed galls on weeping lovegrass (*Eragrostis curvula*), a very important forage grass in semi-arid regions. This nematode became an important nematode pest of this grass in South Africa and significantly affected the seed export industry as seed must be free of nematodes.

Measurements

After Van den Berg (1985).

Female: L = 1.61-1.95 mm; a = 19.9-26.9; b = 8.1-8.7; c = 27.0-42.2; V = 90-94; stylet = 8.1-10.3 μm.

Male: L = 1.01-1.42 mm; a = 30.0-41.9; b = 5.9-8.1; c = 20.1-25.7; stylet = 8.0-11.8 μm; spicules = 34.2-40.8 μm; gubernaculum = 15.8-19.9 μm.

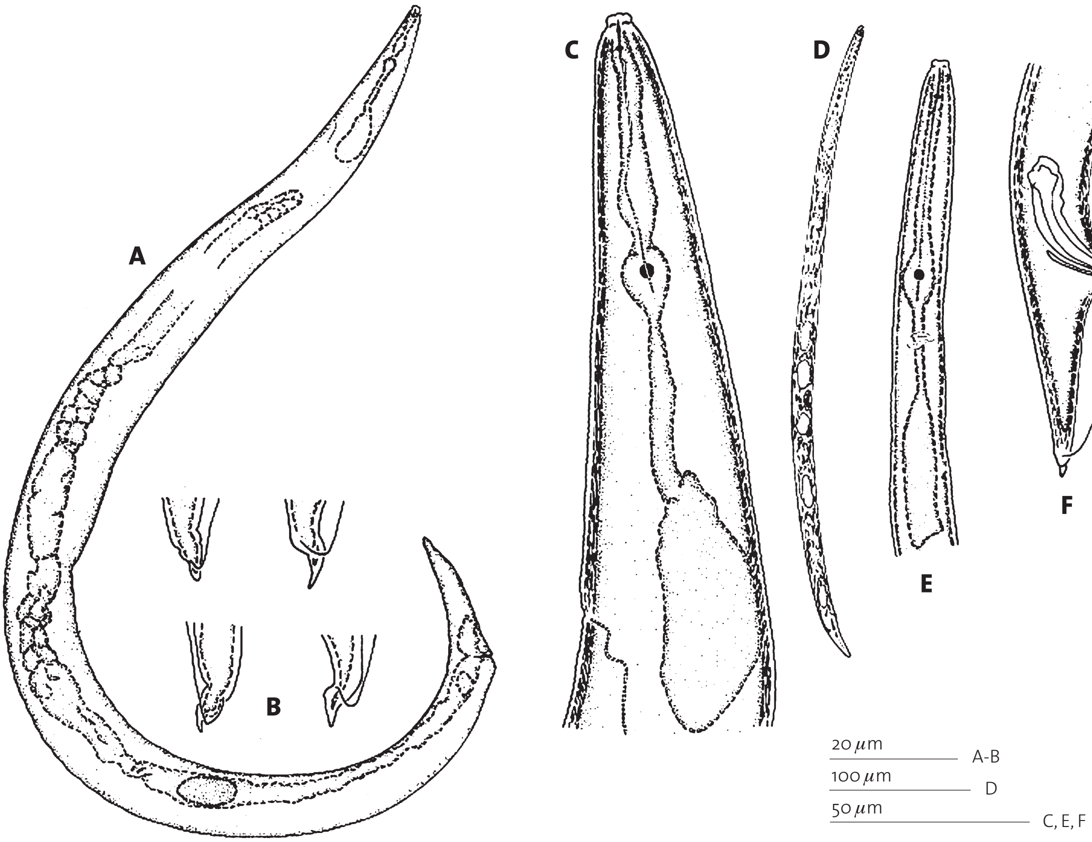

Fig. 14.15. *Afrina wevelli*. A: Female body; B: Variations in male tail tip; C: Anterior part of female; D: Juvenile; E: Anterior part of juvenile; F: Male tail. After Van den Berg (1985).

Second stage juveniles: L = 675-781 mm; a = 36.7-43.9; b = 4.7-5.3; c = 13.1-15.8; stylet = 8.0-9.6 µm.

Description: Female body posture ranging from an open circle to tightly coiled. Labial region flattened. Basal bulb large. Tail long, tapering to narrow rounded tip with slight irregularities just anterior to tip of most specimens. Bursa extending from about one and a half times cloacal body width anterior to cloaca to tail tip. Tail ending in an irregular projection.

Type host: *Eragrostis curvula*.

Distribution: South Africa.

Symptoms: Nematodes induced seed galls which are slightly larger than the normal seeds. Galls are shriveled and mostly a very dark brown, almost black in color.

14.11.4. Genus *Mesoanguina* Chizhov & Subbotin, 1985

Diagnosis: (modified after Chizhov and Subbotin, 1985)

During each growing season, usually two generations of nematodes develop in each gall. Adults of the first generation large, after heat relaxation crescentic or spirally coiled. Adults of second generation smaller, not curved remarkably. Ovary in female of first generation with two or three flexures. Oocytes in several rows. Crustaformeria consisting of 32-60 cells in four to five rows, 8-12 eggs in each, and may contain up to 16 synchronous eggs. Ovary in female of second generation outstretched or with only one flexure. Oocytes in zone of multiplication generally in single file. Crustaformeria shorter than in female of first generation and may contain up to four synchronous eggs. Infective juvenile of third stage. Induce unpigmented galls with well-expressed internal cavity on above-ground parts of dicotyledons, mostly on species of the family Asteraceae (Compositae) (Fig. 14.8), but also on Boraginaceae and Plantaginaceae.

Type species: *M. millefolii* (Low, 1874) Chizhov & Subbotin, 1985 (Fig. 14.3D).

Other species: 6.

Mesoanguina picridis **(Kirjanova, 1944)**
Chizhov & Subbotin, 1985 – the knawel cyst nematode

Mesoanguina picridis is the only plant pathogen, along with the fungus *Puccinia acroptili*, known to be effective as a biological control agent against Russian knapweed, *Acroptilon repens* (L.) DC. *Acroptilon repens* is a persistent herbaceous perennial. The weed is toxic to horses and difficult to control with herbicides. The nematode is endemic to Asia where its use for biocontrol was pioneered (Ivanova, 1966) *Mesoanguina picridis* is not restricted to Russian knapweed and some members of the Cynareae tribe of the Asteraceae family are also hosts.

Measurements

Dimensions of *M. picridis* from different hosts varied considerably.

After Kirjanova and Ivanova (1968) and Watson (1986a).

Female: L = 1.5-2.1 mm; a = 22-28; b = 6.0-12.5; c = 14.0-25.0; V = 81-84; stylet = 12 μm.

Male: L = 1.3-1.8 mm; a = 36-40; b = 6.7-7.0; c = 14.0-18.7; stylet = 12 μm; spicules = 40-43 μm; gubernaculum = 10-15 μm.

Description: Head region with three annules. Lateral field variable within a single specimen, in middle-body region smooth or with six or more up to 20 incisures. Procorpus slightly swollen posteriad with a constriction before spherical median bulb. Ovary with two flexures. Post-vulval uterine branch long, with reduced ovary consisting of one to two cells. Tail conoid with pointed terminus.

Type host: Russian knapweed *Acroptilon repens*.

Other hosts: It has a wide range of host plants within the tribe Cynareae of the Asteracea, including *Centaurea squarrosa* Willd., *Chartolepis biebersteinii* J. & Sp., *Cousinia* spp. and *Centaurea leucophylta* Bied.

Symptoms: The nematode induces galls on the stems, leaves and root collars causing reduction in plant growth and seed production (Fig. 14.7).

Distribution: Tadjikistan, Turkmenistan, Iran, Russia, Armenia, Turkey. Introduced into Canada.

Biological control potential: Ivanova (1966) reported that over 90% of stems of *A. repens* inoculated with crushed gall material (100 g/m^2) were galled, nearly 20% of them died, and there was severe damage to another 30%. *Mesoanguina picridis* has been little used for biocontrol in North America, but is promising in sites that are irrigated or naturally moist in the spring. The nematode can be used as a classical biocontrol agent or applied as a bioherbicide and large numbers of nematodes can be obtained from field plots or increased 140 to 200-fold in 3 months on shoot tips of *M. repens* in tissue culture (Watson, 1986b; Ou & Watson, 1992).

14.11.5. Genus *Ditylenchus* Filipjev, 1936

Since Filipjev proposed *Ditylenchus*, this genus has remained one of the most problematic in systematics. The systematic position of the genus within the Tylenchida has changed several times and it has been treated differently by recent authors. Fortuner and Maggenti (1987) and Sturhan and Brzeski (1991) considered *Nothotylenchus* Thorne, 1941, *Diptenchus* Khan, Chawla & Seshadri, 1969, *Safianema* Siddiqi, 1981 and *Orrina* Brzeski, 1981 as synonyms of *Ditylenchus*, whereas Siddiqi (2000) kept these genera valid.

Diagnosis: (after Chizhov and Subbotin, 1985, and Siddiqi, 2000)

Body slender, not more than 2 mm, not curving strongly when relaxed, mature adults slender. Lateral field with four or six incisures, which may be indistinct. Median bulb muscular or non-muscular, with or without refractive thickenings. Basal bulb a thin elastic sac containing pharyngeal glands. Ovary outstretched, oocytes in the zone of maturation in a single row. Crustaformeria in form of a quadricolumella, with 4 rows with 4-7 cells per row; not longer than the spermatheca. Post-vulval uterine sac present. Bursa adanal to subterminal, never enclosing tail tip. Tails elongate-conoid to subcylindrical or filiform. Fungal feeders and parasites of higher plants. Plant parasite species of *Ditylenchus* induce stunting and swellings on plants, but no galls.

Type species: *Ditylenchus dipsaci* (Kuhn, 1957) Filipjev, 1936. The genus contains near 60 species.

Fortuner (1982) and Brzeski (1991) showed that only a few characters are useful in differentiating *Ditylenchus* species, including: stylet length, tail shape, number of lateral incisures, vulva position, post-vulval uterine sac length, bursa size and spicule length. Dichotomous and tabular keys for identification are given by Fortuner (1982), Brzeski (1991) and Sturhan and Brzeski (1991).

Ditylenchus dipsaci **(Kuhn, 1857) Filipjev, 1936 – the stem and bulb nematode**
(Fig. 14.16)

Ditylenchus dipsaci is among the phytonematodes of greatest economic significance and is on the list of quarantine organisms of many countries around the world. This nematode is distributed worldwide, especially in temperate regions. It is known to attack over 450 different plant species and cause serious problems on onions, garlic, cereals, legumes, strawberries, sugar beet, carrot and many other agricultural and vegetable crops, as well as many horticultural plants, especially phlox, flower bulbs, such as narcissus, hyacinth and tulip. It occurs in several biological races, some of which have a limited host range (Hooper, 1972). According to present morphological, karyological and molecular analyses, *D. dipsaci* has to be considered a species complex. Two species, *D. weischeri* and *D. gigas*, parasitize *Cirsium* spp. and *Vicia faba*, respectively. *Ditylenchus gigas*, formerly considered as a 'giant race' of the stem and bulb nematode, was recently designated and described (Vovlas *et al*., 2011). Several highly specialized races and populations parasitizing *Plantago maritima*, *Taraxacum officinale*, *Crepis praemorsa*, *Pilosella* spp. and other plants are still grouped under the nominal name *D. dipsaci* sensu lato, although they actually represent separate species.

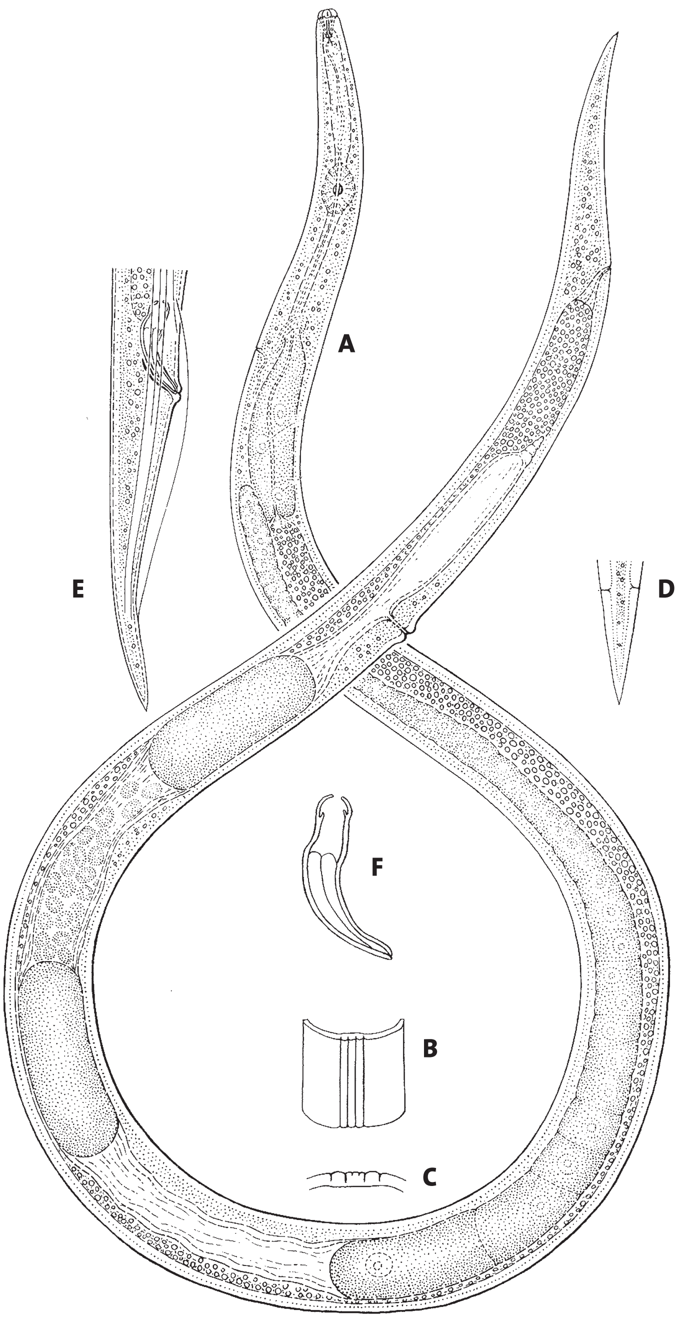

Fig. 14.16. Ditylenchus dipsaci. A: Female; B: Section of cuticle at mid-body showing four incisures; C: Cross section of lateral field; D: Tail; E: Posterior portion of male; F: Spicules. After Thorne (1945).

Measurements

After Sturhan and Brzeski (1991).

Female: L = 1.1-1.7 mm; a = 30.2-64; b = 6.0-8.8; c = 13.5-19.5; V = 79-86; stylet = 9-12 μm.

Male: L = 1.0-1.5 mm; a = 35.4-53.5; b = 5.5-7.8; c = 12-17.3; stylet = 9-12 μm; spicules = 20-28 μm; gubernaculum = 8-11 μm.

Description: Female body almost straight when killed by heat. Labial region low, unstriated, slightly flattened, barely set off from the body, squarish in outline, medial lips large, each subdivided into two submedial lobes by a small, medial bulging area (Fig. 14.17D). Lateral field with four lines (Fig. 14.17H). Basal bulb offset or overlapping intestine. Vulva distinct, anterior ovary outstretched with oocytes usually in a single, occasionally double, row that sometimes reaches the pharyngeal region. Post-vulval part of uterine sac about half of vulva-anus distance long or slightly more. Tail of both sexes conical, always pointed.

Type host: Fuller's Teasel *Dipsacus fullonum* L.

Other hosts: More than 500 plant species from over 40 angiosperm families are known as hosts for this nematode.

Symptoms and pathogenicity: *Ditylenchus dipsaci* lives mostly as an endoparasite in aerial parts of plants (stems, leaves, and flowers) but also attacks bulbs, tubers and rhizomes. Common symptoms of infestation are swelling, distortion, discoloration and stunting of above-ground plant parts and necrosis of bulbs and tubers (Fig. 14.4). The economic thresholds of *D. dipsaci* are mostly very low and even population densities of only 20 nematodes per kg of soil may lead to significant crop losses.

Distribution: Cosmopolitan.

Ditylenchus africanus **Wendt, Swart, Vrain & Webster, 1995 – the peanut pod nematode**

The peanut pod nematode was first reported as *D. destructor* from infested peanut fields in South Africa in 1987. Populations of this nematode were tested on South African potato cultivars but no damage was done to the potato tubers and potato was a poor host. Consequently, the populations parasitizing peanut were considered a distinct race of *D. destructor* with a limited host range (De Waele *et al*., 1989, 1991).

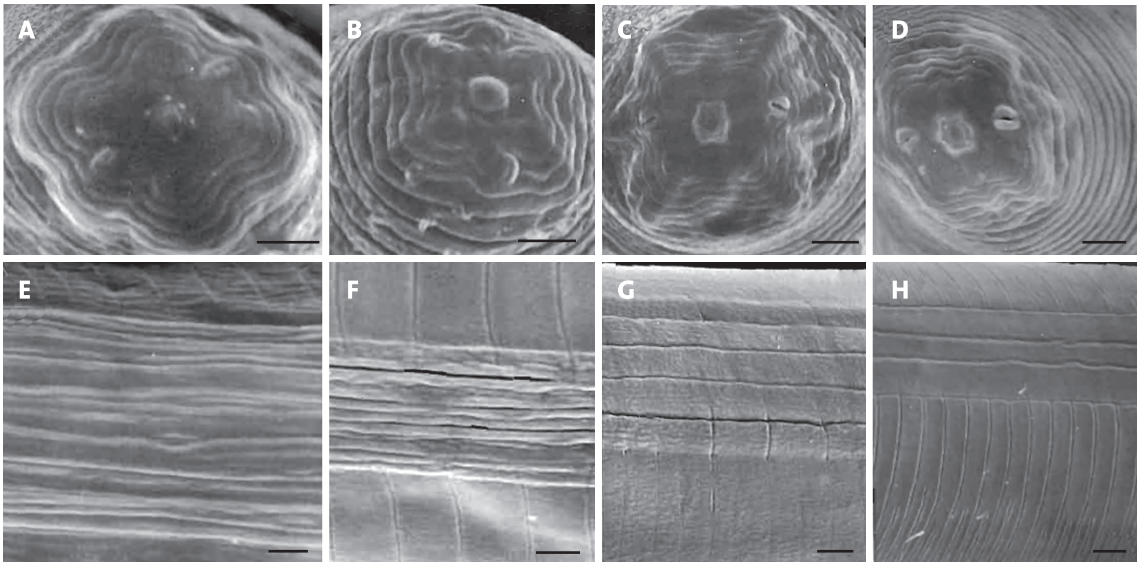

Fig. 14.17. SEM photos of *Ditylenchus* species. A-D: Female head; E-H: Lateral field; A, E: *Ditylenchus africanus*; B, F: *D. myceliophagus*; C, G: *D. destructor*; D, H: *D. dipsaci*. Scale bar = 1 mm. After Wendt *et al*. (1995).

After analysis of rDNA it was established as a separate species (Wendt *et al*., 1995). *Ditylenchus africanus* has been found in all the major peanut production areas of South Africa and can destroy 40-60% of the seeds in heavily infested fields.

Measurements

After Wendt *et al*. (1995).

Female: L = 0.7-1.14 mm; a = 24.2-40.4; b = 7.1-11.8; c = 8.8-16.9; stylet = 8-10 μm; V = 77-81.

Male: L = 0.8-1.0 mm; a = 31-42.4; b = 7.4-10.1; c = 13-15.4; stylet = 8-9.5 μm; spicules = 17-21 μm; gubernaculum = 6-8 μm.

Description: Female head flattened, hexagonal in outline, medial lips large each indented to form two submedial lobes. Stylet delicate, knobs distinct, separated, sloping backwards (Fig. 14.17A). Lateral field with six lines, becoming subdivided to a maximum of 15 lines (Fig. 14.17E). Post-vulval uterine sac 50-143 μm. Tail elongate conoid, tapering in posterior one-third to a finely rounded terminus. Bursa covering 48-66 % of tail length.

Type host: peanut, *Arachis hypogaea* L.

Other hosts: survives on a variety of crops without causing damage.

Symptoms and pathogenicity: Soon after the peg enters the soil and pod formation is initiated, the nematodes enter the plant tissues at the base of the pod near the point of connection with the peg. The first symptom is the appearance of dark brown tissues with a corky appearance at the pod base. The pod may break off during harvesting. In the advanced stage of infection, longitudinal stripes of black or brown tissues appear on the side of the pods and expand into large dark areas. Infected seeds are shrunken. *Ditylenchus africanus* can survive in stored seeds, which may be symptomless (De Waele *et al*., 1997).

Distribution: South Africa and other southern African countries.

***Ditylenchus angustus* (Butler, 1913) Filipjev, 1936 – the rice stem nematode**
(Fig. 14.18)

The rice stem nematode was first described by E.J. Butler causing 'ufra' disease of rice in the territory of modern Bangladesh. *Ditylenchus angustus* is known to occur in rice-growing regions in Asia and Africa, where is considered as an important pest.

Measurements

After Seshadri and Dasgupta (1975).

Female: L = 0.7-1.2 mm; a = 47-62; b = 6-9; c = 15-24; stylet = 9 = 11 μm; V = 70-80.

Male: L = 0.6-1.2 mm; a = 36-62; b = 7-8; c = 17-26; stylet = 9-10 μm; spicules = 16-21 μm; gubernaculum = 6-9 μm.

Description: Female body slender, almost straight to slightly arcuate ventrally when relaxed. Lateral field with four incisures, outer incisures more distinct than inner ones. Median pharyngeal bulb oval, with a distinct valvular apparatus anterior to centre. Vulva a transverse slit, vaginal tube somewhat oblique, reaching more than halfway across body. Spermatheca very long, vagina somewhat oblique, more than half of vulval body width long. Post-vulval uterine sac 2-2.5 vulval body widths or 50-67% of vulva-anus distance long. Tail conoid, tapering to a sharply pointed terminus resembling a mucro. Male morphology similar to female. Bursa narrow extending almost to tail tip. Spicules curved ventrally. Gubernaculum short and simple.

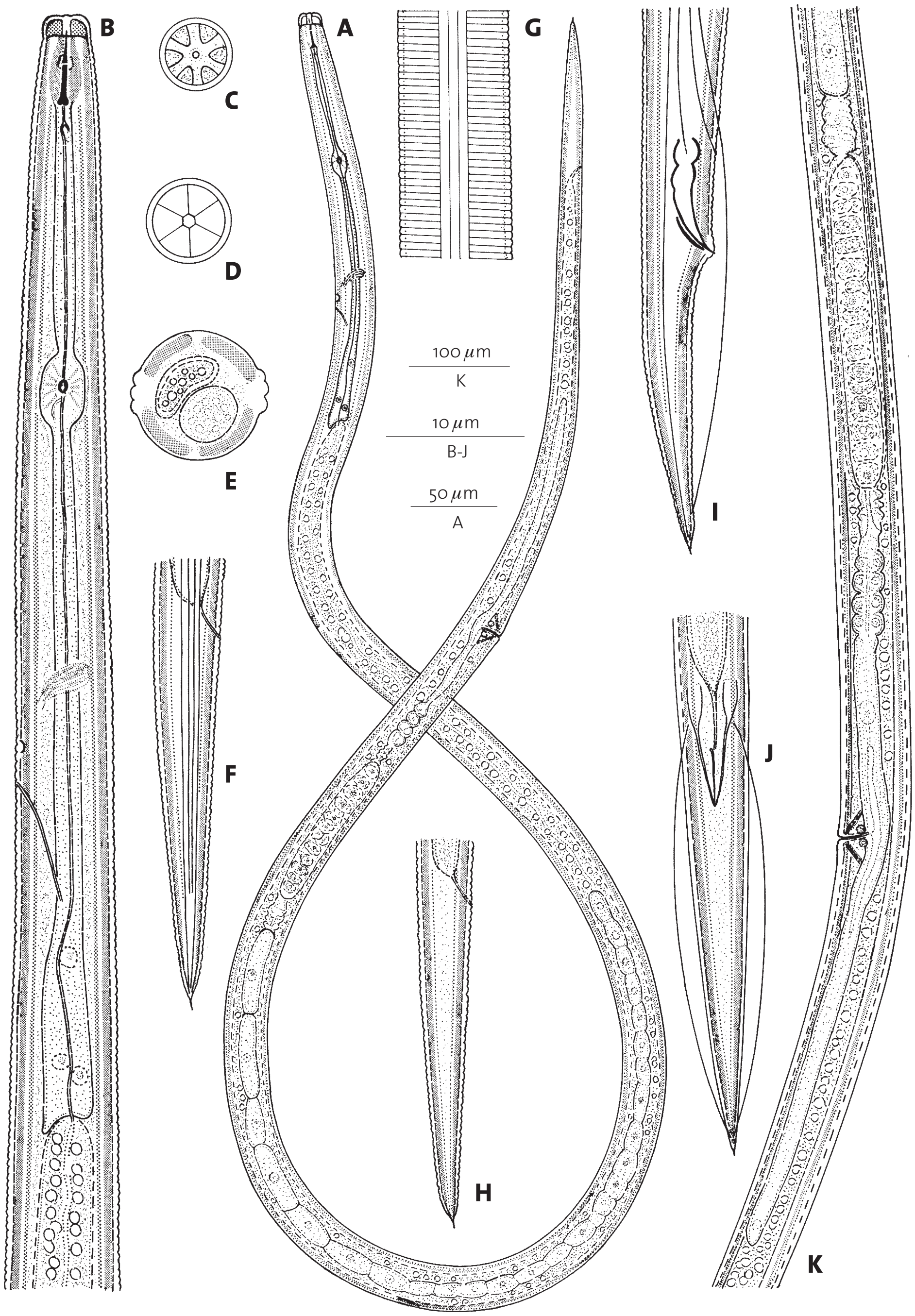

Fig. 14.18. *Ditylenchus angustus*. A: Female, entire; B: Female pharyngeal region; C: En face view; D: Labial framework; E: Female, cross section at mid-body; F: Female tail; G: Lateral field at mid-body; H: Juvenile tail; I: Male tail, lateral view; J: Male tail, ventral view; K: Female vulval region. After Seshadri and Dasgupta (1975).

Type host: Rice, *Oryza sativa*.

Other hosts: *Oryza alta*, *O. cubensis*, *O. eichingeri*, *O. globerrima*, *O. latifolia*, *O. meyriana*, *O. minuta*, *O. nivara*, *O. officinalis*, *O. perennis*, *O. rufipogon* and *O. spontanea*. The wild rice *Leersia hexandra* has been recorded as a host in Madagascar and Burma, and the graminaceous hosts *Echinochloa colona* and *Sacciolepis interrupta* were reported from rice fields in Vietnam. Propagation of the rice stem nematode is also possible on various fungi (Sturhan & Brzeski, 1991).

Symptoms and pathogenicity: *Ditylenchus angustus* occurs in the leaves, inflorescences, young seeds and rolled stems of growing plants. The nematodes feed ectoparasitically causing malformation of host tissues. Infected plants are stunted and the leaves often wilt and become chlorotic. Necrotic brown strains may develop on leaves and leaf sheaths. The leaf sheath and the limb above the last internode become distorted and crimped. The peduncle coils and pedicels bear distorted sterile spikelets. Dark brown patches of infected plants can be observed within fields, normally after panicle initiation. Yield loss caused by this disease has been reported up to 10-15% in India, 20-90% in Thailand, 50-100% in Vietnam and 40-60% or occasionally 100% in Bangladesh deepwater, irrigated and lowland rice. *Ditylenchus angustus* can survive for several months in plant material and seeds (Ibrahim & Perry, 1993).

Distribution: Found in several Asian counties (Burma, Bangladesh, India, Malaysia, Thailand, Vietnam) and Madagascar.

Ditylenchus destructor **Thorne, 1945 – the potato rot nematode**
(Fig. 14.19)

Before *D. destructor* was described in 1945 it was regarded as a race of *D. dipsaci*. Potato rot nematode is of an economic importance and causing great losses in potato, sweet potato, flower bulbs and tubes. It is on the list of quarantine organisms of many countries and organizations (Esser, 1985).

Measurements

After Hooper (1973).

Female: L = 0.69-1.89 mm, a = 18-49, b = 4-12, c = 9-30, V = 73-90, stylet = 10-15 μm.

Male: L = 0.63-1.35 mm, a = 24-50, b = 4-11, c = 11-21, stylet = 10-12 μm, spicules = 24-27 μm, gubernaculum = 9-12 μm.

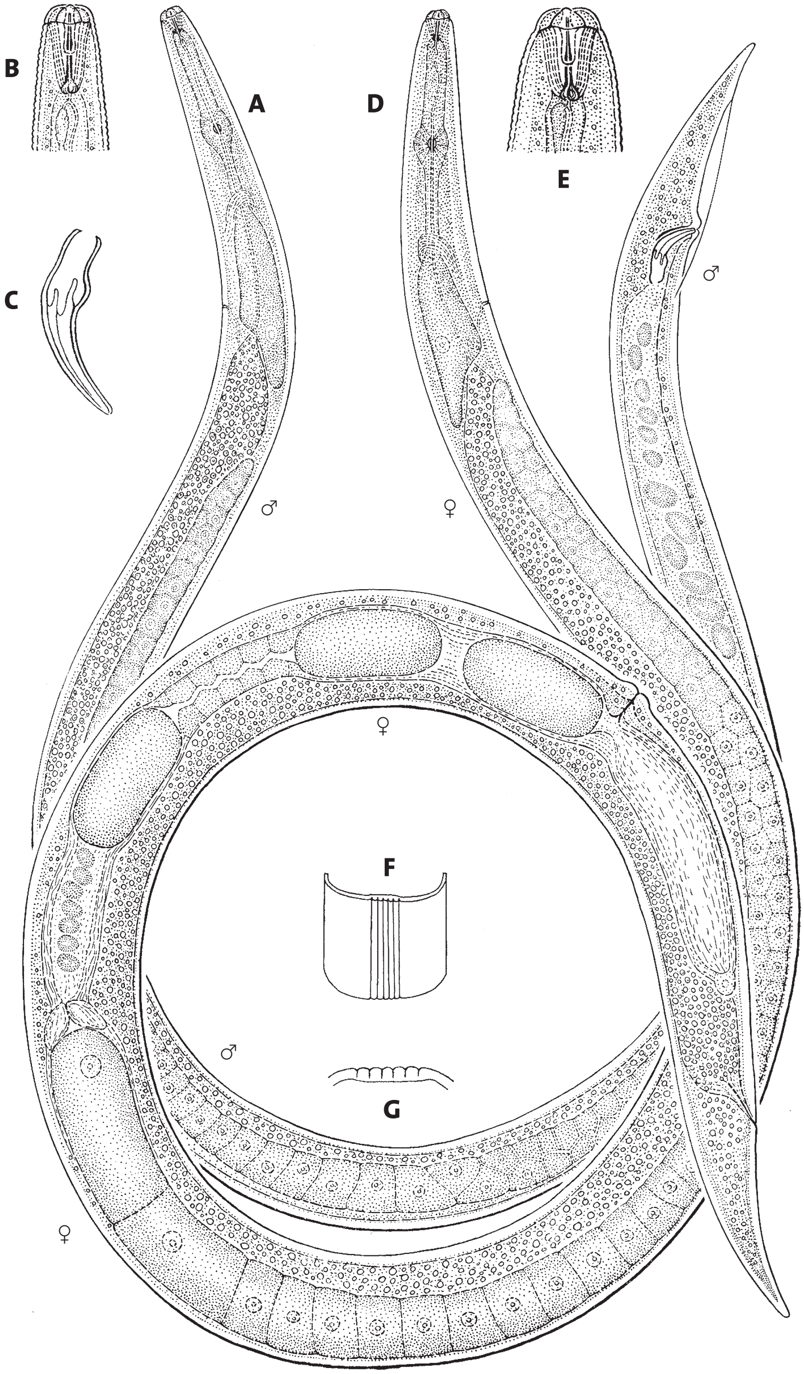

Fig. 14.19. *Ditylenchus destructor*. A: Male; B: Head of male; C: Spicule; D: Female; E: Head of adult female; F: Section of cuticle at mid-body showing six incisures in lateral field; G: Cross-section of lateral field. After Thorne (1945).

Description: Female body with slight ventral curvature when killed by heat. Labial region low and flattened, slightly set off from body, hexagonal to squarish in outline, medial lip large, subdivided into submedial lobes by a small bulging area (Fig. 14.17C). Lateral fields with six incisures throughout most of body length, gradually reducing to two towards extremities (Fig. 14.17G). Glandular pharyngeal bulb overlapping intestine on dorsal side for half to one body width. Vulva distinct, anterior ovary outstretched sometimes reaching pharyngeal region, oocytes in a double row in anterior part but a single row nearer uterus. Post-vulval sac extending about three quarters of distance to anus. Tail conoid with slight ventral curvature with a narrow rounded terminus.

Type host: Potato, *Solanum tuberosum* L.

Other hosts: Economically important crops are Ipomoea batatas, Iris spp., *Tulipa* spp., *Dahlia* spp., *Gladiolus* spp., *Rheum rhabarbarum*, *Trifolium* spp. and *Daucus carota*. The known host range comprises more than hundred species of plants from a wide variety of families. *Ditylenchus destructor* is also capable of reproducing on the mycelium of many fungi.

Symptoms and pathogenicity: The nematode attacks the subterranean parts of plants (tubers, stolons, bulbs, rhizomes, roots), but may occasionally invade above-ground parts, mainly the base of stem (Fig. 14.6). On heavily infected tubers there are typically slightly sunken areas with cracked and wrinkled skin which is detached in places from the underlying flesh. The flesh has a dry and mealy appearance, varying in color from light to dark brown or black.

Distribution: The species is recorded from many countries from Europe, North and South America and parts of Asia and Australia (presently considered as eradicated) mainly from temperate regions.

***Ditylenchus gigas* Vovlas, Troccoli, Palomares-Rius, De Luca, Liébanas, Landa, Subbotin & Castillo, 2011 – the giant stem nematode**
(Fig. 14.20)

The first description of a disease on broad beans caused by the stem nematode, whose adults were considerably larger than *D. dipsaci* adults, was published more than 100 years ago and was named as a ‘giant’ race of *D. dipsaci*. The giant race causes a severe disease of broad bean in several European and African countries and is generally more damaging than any of the other *D. dipsaci* races as it causes more severe symptoms to field broad beans and produces more infested seed. Several studies have shown that this race differs from the normal race in its biology.

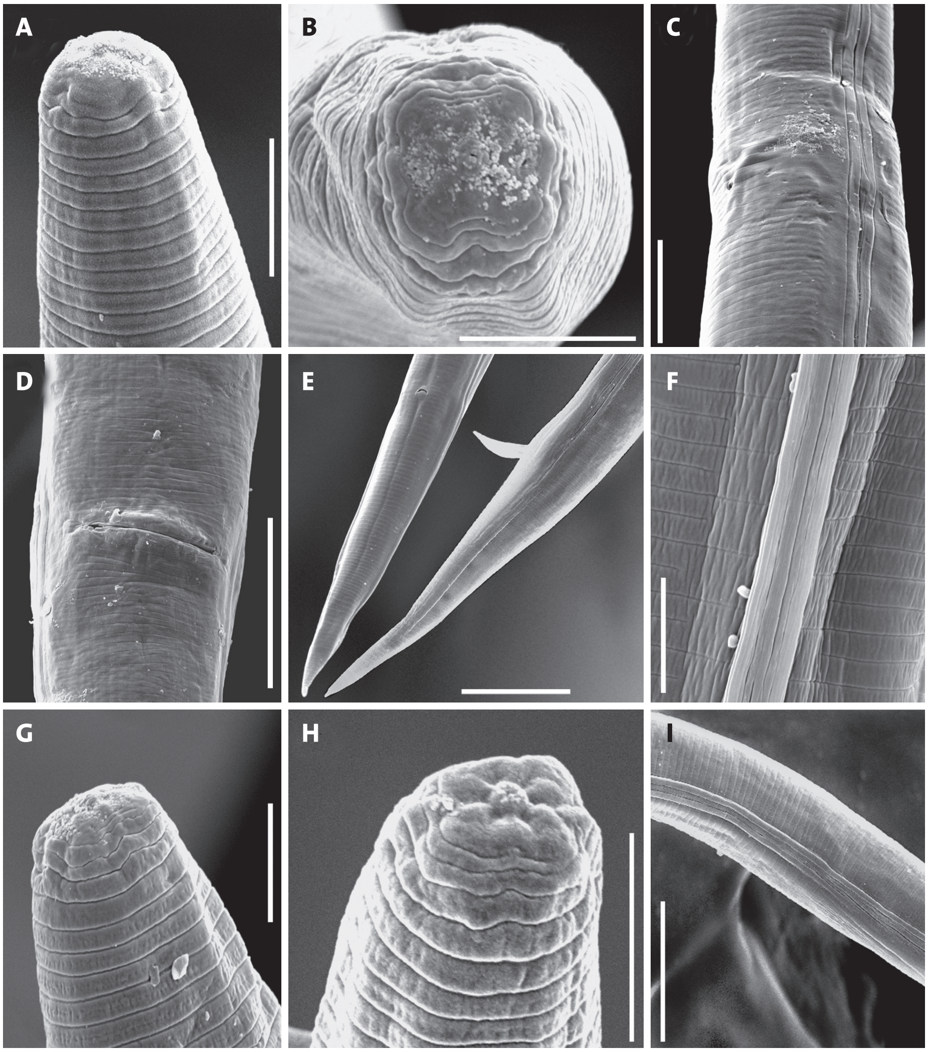

Fig. 14.20. Scanning electron microscope photographs of *Ditylenchus gigas*. A-F: Female; G-I: Male. A: Anterior end; B: En face view; C: Excretory pore; D: Vulva in ventral view; E: Female (left) and male (right) tail (anus opening arrowed); F, I: Lateral field, showing four incisures; G, H: Anterior end in different views. (Scale bars: A, B, F-H = 5 μm; C = 10 μm; D, E, I = 20 μm) After Vovlas *et al*. (2011).

Compared with the normal race, the giant race has a limited host range. The normal race of *D. dipsaci sensu stricto* is assumed to have diploid chromosome number 2n = 24, whereas the giant race parasitizing broad beans is tetraploid with 2n = 48-60. Crossing experiments with diploid races of *D. dipsaci sensu stricto* and the giant race showed that F1 hybrids were formed, but they were infertile. Several studies including PCR-RFLP, protein isoelectric focusing, RAPD and ITS-rRNA gene sequence analysis also confirmed that the giant race is different from normal races. These results led to conclusion that the giant race should be considered as a separate species – *Ditylenchus gigas*.

Measurements

After Vovlas *et al*. (2011).

Female: L = 1.5-1.9 mm; a = 43.0-56.4; b = 7.3-9.3; c = 16.8-27.6; V = 80-83; stylet = 11.5-13.0 μm.

Male: L = 1.3-1.7 mm; a = 34.3-63.0; b = 6.7-10.7; c = 15.7-20.0; stylet = 11.0-12.5 μm; spicules = 23.5-28 μm; gubernaculum = 8.0-11.0 μm.

Description: Labial region flattened, separated from rest of body by a slight constriction, appearing smooth in anterior two-thirds and with a basal annulus in posterior third. Stylet delicate, knobs distinctly sloping backwards. Median bulb oval. Lateral field with four incisures, inner two sometimes faint and indistinct. Post-vulval uterine sac well developed, occupying two vulval body widths. Tail elongate conoid, tapering posteriorly to a finely rounded terminus. Bursa leptoderan, extending anterior to cloaca for about 1.5 anal body widths and covering 72-76% of tail length. Spicules arcuate ventrad, slightly cephalated. Gubernaculum simple, slightly thickened in the central part.

Type host: broad beans, *Vicia faba* L.

Other hosts: *Lamium* spp., *Ranunculus arvensis*, *Convolvulus arvensis* and *Avena sterilis*.

Symptoms and pathogenicity: Stems infested by this nematode are often stunted, twisted and blistered and darker brown or black in color. Infestations in the pod-bearing region are more common and leaf petioles and pods may be distorted (Fig. 14.5). Extensive, brown necrotic zones were observed on the surface of stem toward the end of the crop season. Secondary rotting at the stem base sometimes causes the whole plant to collapse. The lowest pods on plants tend to become infested. Ripe infested pods contain many dried, resistant, fourth-stage juveniles attached to the seeds, especially in the slit in the hilum. Several thousand live nematodes can be recovered from individual seeds, even several years after harvest (Hooper, 1971; Vovlas *et al*., 2011).

Distribution: European and African countries, mainly bordering the Mediterranean Sea.

***Ditylenchus myceliophagus* Goodey, 1958 – the mushroom spawn nematode**
(Fig. 14.21)

Ditylenchus myceliophagus has been reported from most mushroom growing countries throughout the world, predominantly in temperate areas. It is considered the most voracious and damaging nematode for mushroom cultures.

Measurements

After Goodey (1958), Hesling (1974) and Brzeski (1991).

Considerable variation in measurements of juveniles and adults occurs and is dependent on their age, host, nutrition and environment.

Female: L = 0.54-1.4 mm, a = 23-50, b = 5.4-11.7, c = 9.8-20.5, c' = 3.1-6.7, V = 73-90, stylet = 6.5-8.5 μm.

Male: L = 0.50-0.88 mm, a = 21.4-46, b = 4.5-8.7, c = 8.3-15.3, c' = 3.9-6.2, stylet = 7-9 μm.

Description: Relaxed specimens straight or ventrally arcuate. Labial region flattened and cap-like, not offset, without visible striations, hexagonal in outline, medial lips large, each indented to form two submedial lobes (Fig. 14.17B). Lateral field with six equally spaced incisures, in some species becoming subdivided to a maximum of 12 lines (Fig. 14.17F). Stylet thin and delicate, cone about one third of stylet length, knobs rounded. Median bulb fusiform and muscular; basal, glandular bulb broadly spatulate; its basal portion slightly overlapping intestine, usually dorsally, but sometimes ventrally; extent of overlap variable. Post-vulval sac present, 28 to 50 μm long, extending to about halfway between vulva and anus. Bursa extending posteriad from just anterior to base of retracted spicules to about three-quarters of tail length.

Type host: cultivated mushroom *Agaricus bisporus*.

Other hosts: *Ditylenchus myceliophagus* is a mycophagous nematode and there appears to be no accepted evidence that it can feed on higher plants, although successful culturing on callus tissue has been reported. The nematode has been cultured on a wide range of other fungi, including saprophytic, plant pathogenic, predaceous, and animal pathogenic forms (Sturhan & Brzeski, 1991).

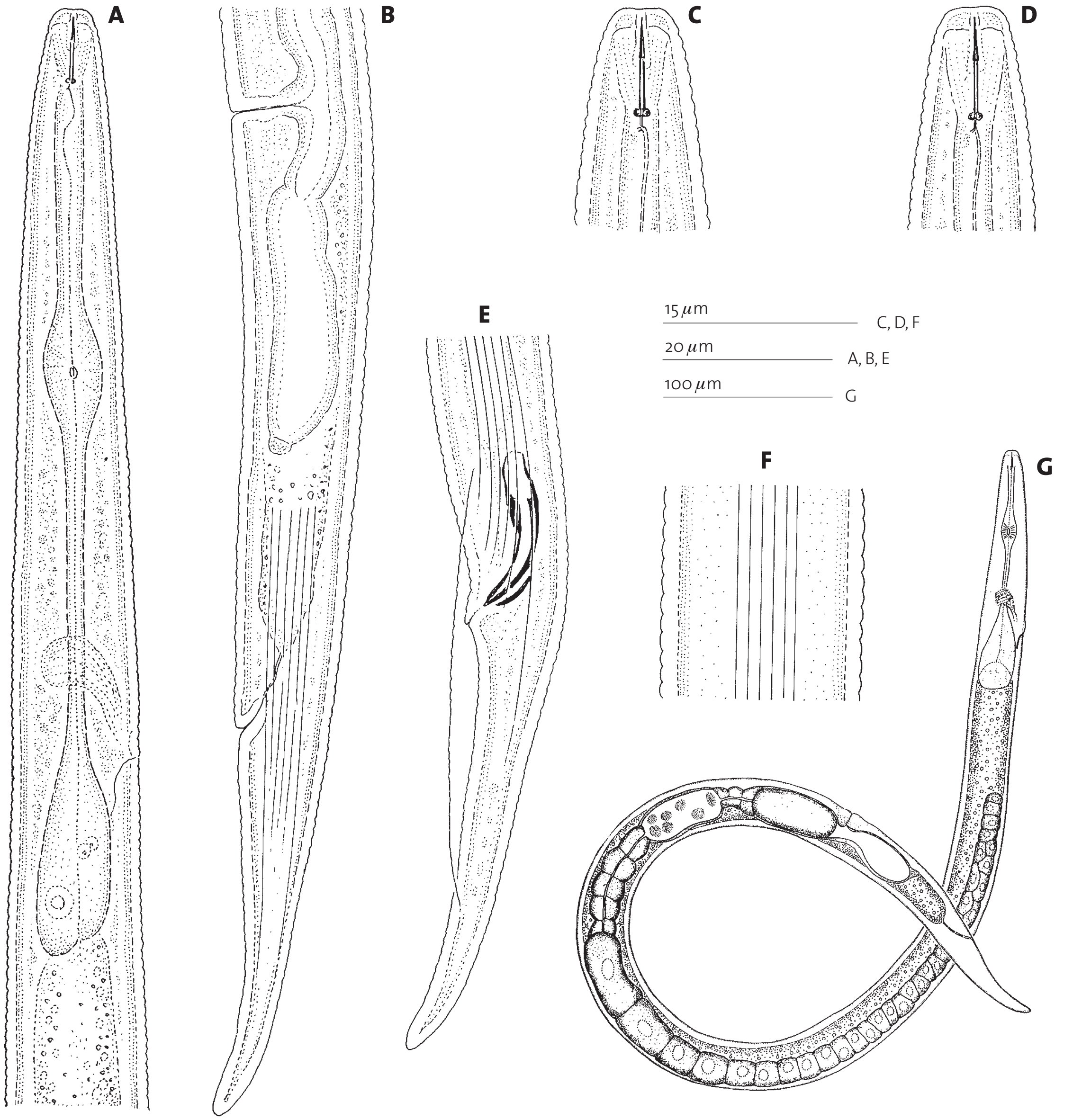

Fig. 14.21. *Ditylenchus myceliophagus*. A: Pharyngeal region of female; B: Posterior portion of female; C: Head end of female; D: Head end of male; E: Tail end of male; F: Lateral field: G. Female. A-F: Courtesy of M.R. Siddiqi; G: after Goodey (1958) and Hooper (1972).

Symptoms and pathogenicity: *Ditylenchus myceliophagus* pierces a fungal cell with its stylet and sucks out the contents. Mushroom or other fungus mycelium may appear unaffected until a critical population level is reached. The growth then stops and thereafter the mycelium degenerates rapidly. Most of the mycelium disappears and the compost in the affected area becomes sunken, soggy and foul-smelling (Hesling, 1974).

14.11.6. Genus *Orrina* Brzeski, 1981

Diagnosis: (after Siddiqi, 2000)

Body about 1 mm or less long, slender (a = 20-30), straight to ventrally arcuate when relaxed. Lateral fields narrow, with four incisures. Labial region flat, cap-like, slightly offset by narrowing of contour. Corpus cylindroid, muscular median bulb absent. Pharyngeal glands extending over intestine. Ovary outstretched, oocytes in one or two rows. Bursa subterminal. Fourth-stage juvenile is infective stage.

Type and only species: *Orrina phyllobia* (Thorne, 1934) Brzeski, 1981.

Orrina phyllobia (Thorne, 1934) Brzeski, 1981 – the nightshade gall nematode
(Fig. 14.22)

The nightshade gall nematode is of economic interest because of its potential as a biological control agent of the cosmopolitan silverleaf nightshade, which is an economically important perennial weed species and, in particular, troublesome in cotton production and difficult to control (Sturhan & Brzeski, 1991).

Measurements

After Thorne (1961) and Brzeski (1991).

Female: L = 0.59-0.84 mm, a = 20-32; b = 7.4-10.5; c = 11.4-17.6; c' = 2.9-4.5; V = 78-84; stylet = 9-11 μm.

Male: L = 0.67-0.85 mm; a = 27-34; b = 7.0-9.7; b' = 4.7-7.5; c = 16.3-19.2; c' = 2.5-3.1; stylet = 9-10 μm.

Description: Female cuticle finely striated. Lateral field with four incisures. Labial region set off. Stylet short, slightly longer than labial region width. Ovary a single line of oocytes except for a short section of multiplication. Post-vulval uterine branch extending two-thirds distance from vulva to anus. Tail of both sexes conical,

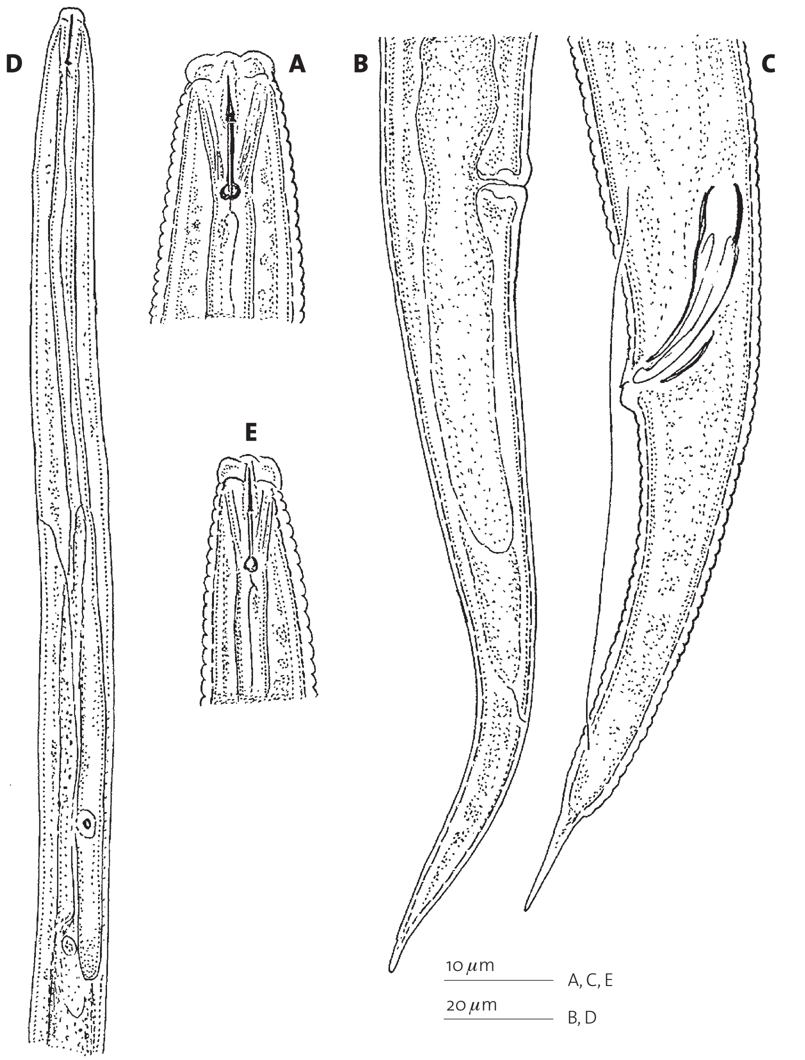

Fig. 14.22. *Orrina phyllobia*. A: Anterior end of female; B: Tail of female; C: Tail of male; D, E: Anterior end of J2. After Siddiqi (2000).

tip rounded. Spicules slightly arcuate, somewhat cephalated. Gubernaculum thin, tough-like. Bursa not extending to tail terminus.

Type host: silverleaf nightshade, *Solanum elaeagnifolium*.

Symptoms and pathogenicity: Visual indications of infection are usually evident after 10-14 days. Galls arise on the leaf surfaces, increase in size, and often engulf the entire leaf surface. Hundreds of thousands of nematodes can be found in a single gall. Heavily infected plants are severely stunted or even killed.

Distribution: Nematode is known from the southwest USA (Arizona and Texas) and southern India (Sturhan & Brzeski, 1991).

Biological control potential: Its extreme host specificity, the capacity to survive desiccation and the detrimental effects it causes on its host make *O. phyllobius* a promising agent for biological weed control (Robinson *et al*., 1978). In field conditions in the USA, and in heavily infested soil, 50% of the infected plants were killed each year whilst growth and reproduction of the surviving plants were reduced by 50% (Krall, 1991).

14.11.7. Genus *Pterotylenchus* Siddiqi & Lenné, 1984

Diagnosis: (after Siddiqi and Lenné, 1984, and Krall, 1991)

Female straight to slightly arcuate. Lateral field with four incisures. Corpus nearly cylindrical, not differentiated into pre- and postcorpus; posterior region slightly swollen, lacking musculature and valve plates. Basal bulb elongate-saccate, with dorsal pharyngeal gland extending over intestine for about one body width. Vulva a long transverse slit flanked and partly covered by large prominent cuticular flaps. Post-vulval uterine sac present. Crustaformeria comprising 32-36 cells in four rows of eight to nine cells each. Spermatheca elongate, empty. Ovary outstretched, with oocytes in one or two rows. Tail elongate-conoid with a pointed tip.

Type and only species: *P. cecidogenus* Siddiqi & Lenné, 1984.

Pterotylenchus cecidogenus **Siddiqi & Lenné, 1984**
(Fig. 14.23)

The nematode was first detected in 1981 from galls on the stems of a tropical pasture legume, *Desmodium ovalifolium*. The stem galls are formed on nodes and stem divisions and were not easily detectable.

Female: L = 0.59-0.80 mm; a = 22-35; b = 4.4-5.8; c = 9.6-12.5; V = 80-84; stylet = 8-11 μm.

J2: L = 0.21-0.31 mm; a = 18-26; c = 6.1-7.5; stylet = 6-7.5 μm.

Male: not known.

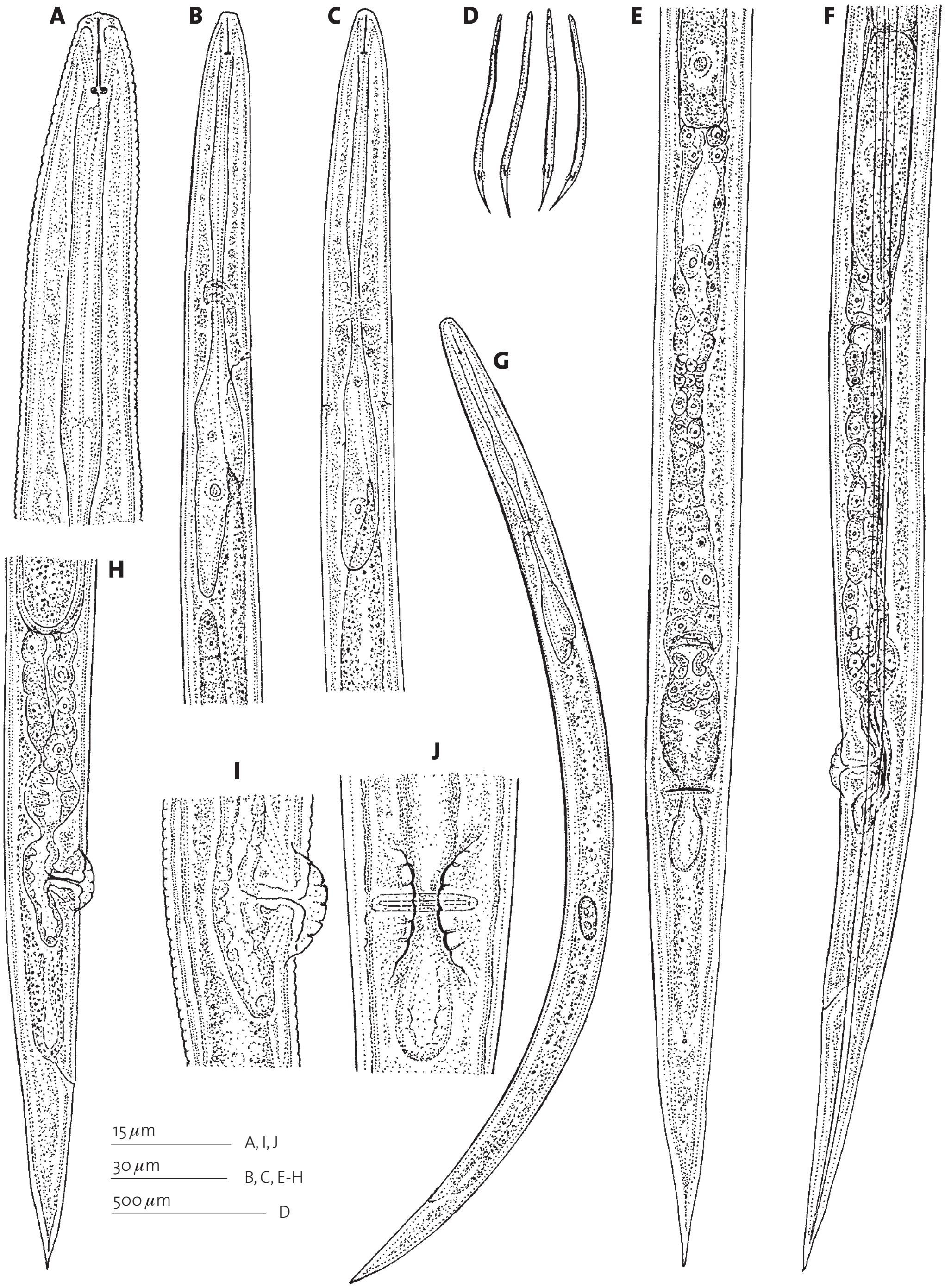

Fig. 14.23. *Pterotylenchus cecidogenus*. A-C: Head of female; D: Female; E, F, H: Tail of female; G: J2; I, J: Vulval region showing lateral vulval flaps. After Siddiqi and Lenné (1984).

Description: Female body straight or slightly ventrally arcuate, often angular at vulva. Vulva about half body-width long; partially covered by large flaps composed of the cuticle of about six enlarged annuli. Juvenile similar to female in most details including dorsal glandular lobe and tail shape.

Symptoms and pathogenicity: Galls 5-20 mm in diameter induced on stems of *D. ovalifolium* and *D. barbatum*. Nematode infection leads to chlorosis, suppression of stem and root growth. The damage is most severe to young seedlings. Nematodes colonize more rapidly the wounded stems resulting from grazing or animal trampling.

Distribution: Colombia.

14.12. References

Bert, W., Leliaert, F., Vierstraete, A.R., Vanfleteren, J.R. & Borgonie, G. (2008). Molecular phylogeny of the Tylenchina and evolution of the female gonoduct (Nematoda: Rhabditida). *Molecular Phylogenetics and Evolution* 48, 728-744.

Bertozzi, T. & Davies, K.A. (2009). *Anguina paludicola* sp. n. (Tylenchida: Anguinidae): The nematode associated with *Rathayibacter toxicus* infection in *Polypogon monspeliensis* and *Lachnagrostis filiformis* in Australia. *Zootaxa* 2060, 33-46.

Bird, A.F. & Stynes, B.A. (1981). The life cycle of *Anguina agrostis*: post-embryonic growth of the second stage larva. *International Journal of Parasitology* 11, 243-250.

Brzeski, M.W. (1981). The genera of Anguinidae (Nematoda, Tylenchida). *Revue de Nématologie* 4, 23-34.

Brzeski, M.W. (1991). Review of the genus *Ditylenchus* Filipjev, 1936 (Nematoda: Anguinidae). *Revue de Nématologie* 14, 9-59.

Chizhov, V.N. (1980). [On the taxonomic status of several species of the genus Anguina Scopoli, 1777.] *Byulleten Vsesoyznogo Instituta Gel'mintologii* K. *I. Skryabina* 26:83-92.

Chizhov, V.N. & Berezina, N.V. (1986). [*Poa annua* L. – a new host of *Paranguina agropyri* (Nematoda: Tylenchida).] *Byulleten Vsesoyznogo Instituta Gel'mintologii K.I. Skryabina* 45, 68-73.

Chizhov, V.N. & Marjenko, A.Yu. (1984). [Morphology and biology of Subanguina radicicola]. *Zoologichesky Zhurnal* 63, 767-769.

Chizhov, V.N. & Subbotin, S.A. (1985). [Revision of the nematode from the subfamily Anguininae (Nematoda, Tylenchida) on the basis of their biological characteristics.] *Zoologichesky Zhurnal* 64, 1476-1486.

Chizhov, V.N. & Subbotin, S.A. (1990). [Phytoparasitic nematodes of subfamily Anguininae (Nematoda, Tylenchida). Morphology, trophic specialization, taxonomic.] *Zoologichesky Zhurnal* 69, 15-26.

Cid del Prado Vera, I. & Maggenti, A.R. (1984). A new gall-forming species of *Anguina* Scopoli, 1777 (Nemata: Anguinidae) on bluegrass, *Poa annua* L., from the coast of California. *Journal of Nematology* 16, 386-392.

Corbett, D.C.M. (1966). Central African Nematodes. III. *Anguina hyparrheniae* n. sp. associated with 'witches' broom of *Hyparrhenia* spp. *Nematology* 12, 280-286.

De Waele, D., Jones, B.L., Bolton, C. & Van den Berg, E. (1989). *Ditylenchus destructor* in hulls and seeds of peanut. *Journal of Nematology* 21, 10-15.

De Waele, D., Wilken, R. & Lindique, J.M. (1991). Response of potato cultivars to *Ditylenchus destructor* isolated from groundnut. *Revue de Nématologie* 14, 123-127.

De Waele, D., Venter, C. & Mc Donald, A.H. (1997). The peanut pod nematode, *Ditylenchus africanus*. *Nematology Circular* 218, 5 pp.

Esser, R.P. (1985). Characterization of potato rot nematode, *Ditylenchus destructor* Thorne, 1945 (Tylenchidae) for regulatory purposes. *Nematology Circular* 124, 3 pp.

Evtushenko, L.I., Dorofeeva, L.V., Dobrovolskaya, T.G. & Subbotin, S.A. (1994). Coryneform bacteria from plant galls induced by nematodes of the subfamily Anguininae. *Russian Journal of Nematology* 2, 99-104.

Evtushenko, L.I., Dorofeeva, L.V., Dobrovolskaya, T.G., Streshinskaya, G.M., Subbotin, S.A. & Tiedje, J.M. (2001). *Agreia bicolorata* gen. nov., sp. nov., to accommodate actinobacteria isolated from narrow reed grass infected by the nematode *Heteroanguina graminophila*. *International Journal of Systematic and Evolutionary Microbiology* 51, 2073-2079.

Fortuner, R. (1982). On the genus *Dilylenchus* Filipjev, 1936 (Nematoda: Tylenchida). *Revue de Nématologie* 5, 17-38.

Fortuner, R. & Maggenti, A.R. (1987). A reappraisal of Tylenchina (Nemata). 4. The family Anguinidae Nicoll, 1935 (1926). *Revue de Nématologie* 10, 163-176.

Giblin-Davis, R.M., Erteld, C., Kanzaki, N., Ye W, Zeng, Y. & Center, B.J. (2010). *Ditylenchus halictus* n. sp. (Nematoda: Anguinidae), an associate of the sweat bee, *Halictus sexcinctus* (Halictidae), from Germany. *Nematology* 12, 891-904.

Goodey, J.B. (1958). *Ditylenchus myceliophagus* n. sp. (Nematoda: Tylenchidae). *Nematologica* 3, 91-96.

Goodey, T. (1930). On *Tylenchus agrostis* (Steinbuch, 1799). *Journal of Helminthology* 8, 197-210.

Goodey, T. (1932). The genus *Anguillulina* Gerv. & v. Ben., 1859, vel *Tylenchus* Bastian, 1865. *Journal of Helminthology* 10, 75-180.

Hesling, J.J. (1974). *Ditylenchus myceliophagus*. *CIH Descriptions of plant-parasitic nematodes. Set* 3, No. 36. Farnham Royal, UK, Commonwealth Agricultural Bureaux, 4 pp.

Hooper, D.J. (1971). Stem eelworm (*Ditylenchus dipsaci*), a seed and soil-borne pathogen of field beans (*Vicia faba*). *Plant Pathology* 20, 25-27.

Hooper, D.J. (1972). *Ditylenchus dipsaci*. *CIH Descriptions of plant-parasitic nematodes. Set* 1, No. 14. Farnham Royal, UK, Commonwealth Agricultural Bureaux, 4 pp.

Hooper, D.J. (1973). *Ditylenchus destructor*. *CIH Descriptions of plant-parasitic nematodes. Set* 2, No. 21. Farnham Royal, UK, Commonwealth Agricultural Bureaux, 3 pp.

Hooper, D.J. & Southey, J.F. (1978). *Ditylenchus*, *Anguina* and related genera. In: Southey, J.F. (Ed.). *Plant nematology*. London, UK, HMSO, pp. 78-97.

Ibrahim, S.K. & Perry, R.N. (1993). Desiccation survival of the rice stem nematode *Ditylenchus angustus*. *Fundamental and Applied Nematology* 16, 31-38.

Ivanova, T.S. (1966). [Biological control of mountain bluet (*Acroptilon picridis* C.A.M.).] *Izvestiya Academil Nauk Tadzhikoi* SSR 2, 51-63.

Kirjanova, E.S. & Ivanova, T.S. (1968). [New species of the genus *Paranguina* Kirjanova, 1955 (Nematoda: Tylenchidae) from Tadzhikistan.] In: [*The Gorge of Kondor*.] Danin, Dushambe, pp. 200-217.

Kirjanova, E.S. & Krall, E.L. (1971). [*Plant parasitic nematodes and their control*.] Vol. 2. Leningrad, USSR, Nauka.

Krall, E.L. (1991). Wheat and grass nematodes: *Anguina*, *Subanguina*, and related genera. In: Nickle, W.R. (Ed.). *Manual of agricultural nematology*. New York, NY, USA, Marcel Dekker, Inc., pp. 721-760.

McClure, M., Schmitt, M.E. & McCullough, M.D. (2008). Distribution, biology and pathology of *Anguina pacificae*. *Journal of Nematology* 40, 226-239.

McKay, A.C. (1993). Livestock deaths associated with *Clavibacter toxicus*/*Anguina* sp. infection in seedheads of *Agrostis avenacea* and *Polypogon monspeliensis*. *Plant Disease* 77, 635-641.

Nicoll, W. (1935). Vermes. *Zoological Record* 72, 105.

Ou, X. & Watson, A.K. (1992). *In vitro* culture of *Subanguina picridis* in Russian knapweed callus, excised roots and shoot tissues. *Journal of Nematology* 24, 199-204.

Paramonov, A.A. (1962). [*Fundamentals of plant helminthology*.] Vol. I. Moscow, USSR, Nauka, 480 pp.

Paramonov, A.A. (1967). A critical review of the suborder Tylenchina (Filipjev, 1934) (Nematoda: Secementea). *Trudy gel'mintologicheskoi laboratorii Akademii Nauk SSSR* 18, 78-101.

Paramonov, A.A. (1970). [*Fundamentals of plant helminthology. Taxonomy of nematodes of the superfamily Tylenchoidea*.] Vol. 3. Moscow, USSR, Nauka, 253 pp.

Powers, T.O., Szalanski, A.L., Mullin, P.G., Harris, T.S., Bertozzi, T. & Griesbach, J.A. (2001). Identification of seed gall nematodes of agronomic and regulatory concern with PCR-RFLP of ITS1. *Journal of Nematology* 33, 191-194.

Price, P.C., Fisher, J.M. & Kerr, A. (1979). On *Anguina funesta* n. sp. and its association with *Corynebacterium* sp., in infecting *Lolium rigidum*. *Nematologica* 25, 76-85.

Riley, I.T. & Barbattibarbetti, M.J. (2008). Australian anguinids: their agricultural impact and control. *Australasian Plant Pathology* 37, 289-297.

Riley, I.T., Reardon, T.B. & McKay, A.C. (1988). Electrophoretic resolution of species boundaries in seed-gall nematodes, *Anguina* spp. (Nematoda: Anguinidae), from some graminaceous hosts in Australia and New Zealand. *Nematologica* 34, 401-411.

Robinson, A.F., Orr, C.C. & Abernathy, J.R. (1978). Distribution of *Nothanguina phyllobia* and its potential as a biological control agent for silver-leaf nightshade. *Journal of Nematology* 10, 363-366.

Seshadri, A.R. & Dasgupta, D.R. (1975). *Ditylenchus angustus*. *CIH Descriptions of plant-parasitic nematodes. Set* 5, No. 64. Farnham Royal, UK, Commonwealth Agricultural Bureaux, 3 pp.

Siddiqi, M.R. (1971). Structure of the oesophagus in the classification of the superfamily Tylenchoidea (Nematoda). *Indian Journal of Nematology* 1, 25-43.

Siddiqi, M.R. (1980). The origin and phylogeny of the nematode orders Tylenchida Thorne, 1949 and Aphelenchida n. ord. *Helminthological Abstracs, Series B* 49, 143-170.

Siddiqi, M.R. (1986). *Tylenchida parasites of plants and insects*. Farnham Royal, UK, Commonwealth Agricultural Bureaux, 645 pp.

Siddiqi, M.R. (2000). *Tylenchida parasites of plants and insects*. Second edition. Wallingford, UK, CABI Publishing, 848 pp.

Siddiqi, M.R. & Lenné, J.M. (1984). *Pterotylenchus cecidogenus* n. gen., n. sp., a new stem-gall nematode parasitizing *Desmodium ovafifolium* in Colombia. *Journal of Nematology* 16, 62-65.

Southey, J.F. (1972). *Anguina tritici*. *CIH Descriptions of plant-parasitic nematodes. Set* 1, No. 13. Farnham Royal, UK, Commonwealth Agricultural Bureaux, 4 pp.

Southey, J.F. (1973). *Anguina agrostis*. *CIH Descriptions of plant-parasitic nematodes. Set* 2, No. 20. Farnham Royal, UK, Commonwealth Agricultural Bureaux, 3 pp.

Southey, J.F., Topham, P.B. & Brown, D.J.F. (1990). Taxonomy of some species of *Anguina* Scopoli, 1777 (*sensu* Brzeski, 1981) forming galls on Gramineae: value of diagnostic characters and present status of nominal species. *Revue de Nématologie* 13, 127-142.

Sturhan, D. & Brzeski, M.W. (1991). Stem and bulb nematodes, *Ditylenchus* spp. In: Nickle, W.R. (Ed.). *Manual of agricultural nematology*. New York, NY, USA, Marcel Dekker, Inc., pp. 423-464.

Stynes, B.A. & Bird, A.F. (1980). *Anguina agrostis*, the vector of annual rye grass toxicity in Australia. *Nematologica* 26, 475-490.

Subbotin, S.A., Krall, E.L., Riley, I.T., Chizhov, V.N., Staelens, A., De Loose, M. & Moens, M. (2004). Evolution of the gall-forming plant parasitic nematodes (Tylenchida: Anguinidae) and their relationships with hosts as inferred from Internal Transcribed Spacer sequences of nuclear ribosomal DNA. *Molecular Phylogenetics and Evolution* 30, 226-235.

Thorne, G. (1945). *Ditylenchus destructor* n. sp., the potato rot nematode, and *Ditylenchus dipsaci* (Kühn, 1857) Filipjev, 1936, the teasel nematode. *Proceedings of the Helminthological Society of Washington* 12, 27-33.

Thorne, G. (1961). *Principles of nematology*. New York, NY, USA, McGraw-Hill Book Co., Inc., 553 pp.

Triantaphyllou, A.C. & Hirschmann, H. (1980). Cytogenetics and morphology in relation to evolution and speciation of plant-parasitic nematodes. *Annual Review of Phytopathology* 18, 333-359.

Van den Berg, E. (1985). Notes on the genus *Afrina* Brzeski, 1981 (Anguinidae: Nematoda) with description of new and known species. *Phytophylactica* 17, 69-79.

Vovlas, N., Troccoli, A., Palomares-Rius, J.E., De Luca, F., Liébanas, G., Landa, B.B., Subbotin, S.A. & Castillo, P. (2011). *Ditylenchus gigas* n. sp. parasitizing broad bean: a new stem nematode singled out from the "*Ditylenchus dipsaci* species complex" based on polyphasic identification and molecular phylogeny. *Plant Pathology* 60, 762-775.

Watson, A.K. (1986a). Morphological and biological parameters of the knapweed nematode, *Subanguina picridis*. *Journal of Nematology* 18, 154-158.

Watson, A.K. (1986b). Biology of *Subanguina picridis*, a potential biological control agent of Russian knapweed. *Journal of Nematology* 18, 149-154.

Wendt, K.R., Swart, A., Vrain, T. & Webster, J.M. (1995). *Ditylenchus africanus* sp. n. from South Africa; a morphological and molecular characterization. *Fundamental and Applied Nematology* 18, 241-250.

Zhao, Z.Q., Davies, K., Alexander, B. & Riley, I.T. (2011). *Litylenchus coprosma* gen. n., sp. n. (Tylenchida: Anguinata), from leaves of *Coprosma repens* (Rubiaceae) in New Zealand. *Nematology* 13, 29-44.