# A New Gall-Forming Species of Anguina Scopoli, 1777 (Nemata: Anguinidae) on Bluegrass, Poa annua L., from the Coast of California 

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#### Abstract

Anguina pacificae n. sp. is described and illustrated from stem galls on bluegrass, Poa annua L., from golf courses along coastal California. The females are characterized by constrictions in the anterior and posterior connections of the isthmus with the respective parts of the esophagus, the long multicellular columella, and the sharply pointed tail tip. Males are dorsally curved after death; body width is increased markedly after 13 annuli in both sexes, and the tail is conical and with an acute terminus.


Key words: taxonomy, columella, galls, scanning electron microscopy.

During September 1978, Larry Costello, Farm Advisor, Extension Service, San Mateo County, collected Poa annua L. (bluegrass) with green stem galls containing nematodes from several golf courses along the Coast of Central California (San Francisco, San Mateo, and Monterey counties). This is the first record of this disease. Galls were located at the bases of stems. The inflorescence was not attacked. Dissected galls contained a central cavity with a variable number of adult nematodes ( $2-8$ ) inside, along with hundreds of larval stages and eggs. Detailed morphological and anatomical studies of this nematode con-

[^0]vinced us that it represents a new species. Mature galls were filled with bacteria appearing as a white cream.

## Materials and Methods

Galls from the type locality, Pebble Beach, San Mateo County, California, were dissected in tap water with the aid of a dissecting microscope; nematodes in bac-teria-filled galls were removed from the creamy material (unidentified bacteria) and washed in water. These nematodes and nematodes from galls without bacteria were killed and fixed with Seinhorst's (5) fixative, processed into glycerin, and mounted in anhydrous glycerin (3). For scanning electron microscope (SEM) observations, nematodes were prepared as previously reported (2). Nematodes were subsequently processed through critical point drying and mounted on stubs. Nematodes were at-


Fig. 1. Anguina pacificae n. sp. Adult female. A) Esophageal region. B) Cephaic region. C) Posterior body. D) Tail. E) Tail tip. F-J) Reproductive system. F-G) Ovary. H) Oviduct. I) Section of columella. J) Uterus and vulva.
tached to the aluminum foil with a small amount of glue prepared from transparent (Scotch) tape in benzene. Specimens were sputter-coated with $400-500 \AA$ of gold and examined with an ETEC electron microscope at 10 kV .

## Species Description

> Anguina pacificae n. sp.
> $($ Figs. $1-3)$

Females ( $n=26$ ): $\mathrm{L}=1.44-2.58 \mathrm{~mm}$ (mean $2.09 \mathrm{~mm}, 95 \%$ confidence interval $\pm 0.12) \mathrm{a}=19.4-33.2(26.5 \pm 1.3)$; $\mathrm{b}=8.1-13.1(10.9 \pm 0.59) ; \mathrm{c}=19.9-34.4$ (27.8 $\pm 1.7$ ); stylet $=8.9-12.4 \mu \mathrm{~m}$ (10.8 $\mu \mathrm{m} \pm 0.19$ ); dorsal gland orifice $=1.4-3.4$ $\mu \mathrm{m}(2.6 \mu \mathrm{~m} \pm 0.23)$; median bulb length $=$ $15.1-28.6 \mu \mathrm{~m}(19.8 \mu \mathrm{~m} \pm 1.01)$; median bulb width $=12.1-15.8 \mu \mathrm{~m}(13.9 \mu \mathrm{~m} \pm$ 0.39 ); esophagus length $=154.1-210.3 \mu \mathrm{~m}$ ( $188.3 \mu \mathrm{~m} \pm 6.2$ ); isthmus $=18.9-51.7 \mu \mathrm{~m}$ ( $56.5 \mu \mathrm{~m} \pm 2.97$ ); dorsal esophageal gland length $=39.7-71.7 \mu \mathrm{~m}(56.0 \mu \mathrm{~m} \pm 4.03)$; dorsal esophageal gland width $=12.4-33.4$ $\mu \mathrm{m}(21.7 \mu \mathrm{~m} \pm 2.3)$; excretory pore $=$ $115.5-148.3 \mu \mathrm{~m}(126.9 \mu \mathrm{~m} \pm 4.2$ ); postuterine sac length $=96.2-192.1 \mu \mathrm{~m}$ (139.8 $\mu \mathrm{m} \pm 10.36$ ); distance vulva to anus $=$ 155.3-316 $\mu \mathrm{m}$ ( $199 \mu \mathrm{~m} \pm 15.83$ ); tail $=$ $65.2-95.8 \mu \mathrm{~m}(74.9 \mu \mathrm{~m} \pm 2.68) ; \mathrm{V} \%=82-$ $89(86 \% \pm 1.8)$.
Holotype (female): $\mathrm{L}=2.08 \mathrm{~mm} ; \mathrm{a}=26.3$; $\mathrm{b}=10.0 ; \mathrm{c}=30.1$; stylet $=10.7 \mu \mathrm{~m}$; esophagus $=207 \mu \mathrm{~m}$; excretory pore $=$ $115.5 \mu \mathrm{~m}$; postuterine sac length $=152.8$ $\mu \mathrm{m}$; distance vulva to anus $=232 \mu \mathrm{~m}$; tail $=$ $69.0 \mu \mathrm{~m} ; \mathrm{V} \%=85.4$.
Description (female): Body cylindrical, assuming C shape after death; lip region without visible annuli by light microscope, not offset; after 13-18 anterior annuli, body increasing rapidly in diameter, and body annuli inconspicuous. Labial region (SEM) with two annuli and a few longitudinal lines; oral disc small, not prominent; oral opening circular; amphid aperture pore-like, located at lateral edges of oral disc. Lateral field with four main distinct lines, and many longitudinal fine lines between them. Cephalic framework weak. Stylet with fine cone and distinct shaft; stylet knobs oriented posteriorly without anterior projections. Procorpus variable in width, gradually widening to connect with median bulb (metacorpus), slight constric-
tion between procorpus and metacorpus. Lumen in procorpus displaced to ventral side by contents and duct of dorsal esophageal gland. Median bulb oblong, valve distinct (crescentic thickenings), distinct constriction at metacorpus-isthmus junction; isthmus of variable width, distinct constriction at junction with esophageal glands; lumen in isthmus also displaced to ventral side by contents of esophageal glands. Three esophageal glands present, two of them small and half the size of the large central gland; generally, one small gland dorsal and one ventral to large central gland, nuclei small but distinct. Central esophageal gland, a long, large lobe, nucleus conspicuous. Esophageal lumen passing through central gland to junction with intestine. Excretory pore located at level of posterior end of isthmus or anterior extreme of esophageal glands, hemizonid immediately anterior to excretory pore; nerve ring located in the middle of the isthmus or light posterior to it. Ovary with one or two flexures, sometimes straight and extending to esophagus. Ovary two or five cells in width, continuous with oviduct; lumen of oviduct clear, continuous with the columella; columella a long tube, five or six hexagonal cells wide, more than 14 cells long; continuous with short thick walled uterus, connected to vagina on one side, and postuterine sac on the other. Postuterine sac containing degenerate cells. Vulval lips distinct. Tail conical, sharply pointed tip.
Males ( $n=17$ ): $\mathrm{L}=1.22-1.84 \mathrm{~mm}(1.56$ $\mathrm{mm} \pm 0.075) ; \mathrm{a}=21.1-33.0(27.9 \pm 1.71) ;$ $\mathrm{b}=5.8-10.4(9.05 \pm 0.79) ; \mathrm{c}=19.8-29.8$ $(23.2 \pm 1.25)$; stylet $=9.3-12.7 \mu \mathrm{~m}(11.0$ $\mu \mathrm{m} \pm 0.46)$; dorsal gland orifice $=1.3-3.1$ $\mu \mathrm{m}(2.0 \mu \mathrm{~m} \pm 0.26)$; median bulb length $=$ $15.5-20.0 \mu \mathrm{~m}(18.2 \mu \mathrm{~m} \pm 0.84)$; median bulb width $=11.3-16.5 \mu \mathrm{~m}$ ( $13.3 \mu \mathrm{~m} \pm$ 0.75 ); esophagus length $=142.1-242.0 \mu \mathrm{~m}$ (179.5 $\mu \mathrm{m} \pm 17.3$ ); isthmus $=30.3-72.4$ $\mu \mathrm{m}(40.2 \mu \mathrm{~m} \pm 5.72)$; dorsal esophageal gland length $=24.1-68.9 \mu \mathrm{~m}(56.4 \mu \mathrm{~m} \pm$ 7.7); esophageal gland width $=10.0-32.7$ $\mu \mathrm{m}(19.7 \mu \mathrm{~m} \pm 3.7)$; excretory pore $=$ $98.9-137.9 \mu \mathrm{~m}(127.0 \mu \mathrm{~m} \pm 5.4) ; \mathrm{T}=$ $58.6-82.1 \%(68.7 \% \pm 4.7)$; caudal alae $=$ $60.3-84.5 \mu \mathrm{~m}(72.1 \mu \mathrm{~m} \pm 4.8)$; spicules $=$ $26.2-40.7 \mu \mathrm{~m}(34.2 \mu \mathrm{~m} \pm 2.7)$; gubernaculum $=10.6-15.5 \mu \mathrm{~m}(13.0 \mu \mathrm{~m} \pm 0.61)$; tail $=58.3-76.9 \mu \mathrm{~m}(68.7 \mu \mathrm{~m} \pm 2.7)$.


Fig. 2. Anguina pacificae n. sp. A-C) Second-stage larvae. A) Posterior body. B) Tail tip. C) Esophageal region. D-H) Male. D) Esophageal region. E) Cephalic region. F) Tail. G) Spicule and gubernaculum. H) Tail tip. I) Gall on plant crown.


Fig. 3. Scanning electron micrographs of Anguina pacificae n. sp. A) Female cephalic region. B) Female posterior body. C) Female anal region. D) Second-stage larva tail. E) Female lateral field. F) Male tail.

Allotype (male): $\mathrm{L}=1.84 \mathrm{~mm} ; \mathrm{a}=31.7$; $\mathrm{b}=10.4 ; \mathrm{c}=25.2$; stylet $=12.7$; esophagus $=175.2 \mu \mathrm{~m}$; excretory pore $=141.0$ $\mu \mathrm{m}$; tail $=73.1 \mu \mathrm{~m} ; \mathrm{T}=67.9 \%$; spicules $=$ $41.4 \mu \mathrm{~m}$; gubernaculum $=11.4 \mu \mathrm{~m}$.

Description (male): Body cylindrical, arcuate with ventral surface outermost after death. Labial region continuous with body contour, not offset, with two annuli and irregular longitudinal lines when viewed
by SEM. In face view labial disc small and rhomboidal; oral opening circular; amphids pore-like and located laterally on outer margin of first annulus. Approximately 13 distinct body annuli visible posterior to labial disc, posteriorly from this point, body width increasing gradually, and body annuli become fine or obscure. Stylet short with distinct shaft, knobs rounded without anterior projections. Dorsal gland orifice distinct, gland contents displacing esophageal lumen to ventral side. Procorpus variable in width, slightly swollen near connection with median bulb, a slight constriction separating these two structures. Median bulb oblong with distinct valve (crescentic thickenings), connected with isthmus by distinct constriction; isthmus increasing gradually in width until constriction at esophageal glands. Two subventral glands almost one-half the size of dorsal gland, in most cases one subventral gland ventral and one dorsolateral. Dorsal gland forming major lobe. Esophageal lumen connecting with intestine at posterior extreme of dorsal gland; little or no gland overlap of intestine. Nerve ring located around posterior or middle region of isthmus. Excretory pore at level of connection of isthmus and esophageal glands or slightly posterior to it. Testis often with flexures, sometimes reaching median esophageal bulb; spermatozoa globular with granular appearance. Spicules slightly arcuate, distally conical. Gubernaculum short with slight anterior projection and acute terminus. Caudal alae leptoderan, beginning anterior to spicules and ending before tail tip. Tail with acute terminus.

Second-stage larvae $(n=20): \quad \mathrm{L}=0.63-$ $0.97 \mathrm{~mm}(0.71 \mathrm{~mm} \pm 0.043) ; \mathrm{a}=37.0-$ $56.4(43.8 \pm 2.1) ; \mathrm{b}=3.5-7.3(4.5 \pm 0.41)$; $\mathrm{c}=8.8-11.4(9.9 \pm 0.35) ;$ stylet $=10.0-$ $12.7 \mu \mathrm{~m}(11.0 \mu \mathrm{~m} \pm 0.35)$; dorsal gland opening $=1.7-3.4 \mu \mathrm{~m}$ (2.4 $\mu \mathrm{m} \pm 0.23$ ); median bulb length $=12.4-15.9 \mu \mathrm{~m}$ (13.9 $\mu \mathrm{m} \pm 0.44)$; median bulb width $=7.9-10.7$ $\mu \mathrm{m}(9.0 \mu \mathrm{~m} \pm 0.34)$; esophagus length $=$ 124.4-196.5 $\mu \mathrm{m}$ ( $159.2 \mu \mathrm{~m} \pm 8.3$ ); isthmus $(\mathrm{n}=9)=24.1-41.4 \mu \mathrm{~m}(28.8 \mu \mathrm{~m} \pm 4.0)$; dorsal esophageal gland length $=46.5-$ $68.9 \mu \mathrm{~m}(61.4 \mu \mathrm{~m} \pm 8.6)$; dorsal esophageal gland width $=6.9-11.4 \mu \mathrm{~m}(9.3 \mu \mathrm{~m} \pm 1.3)$; excretory pore $=92.8-124.5 \mu \mathrm{~m}$ (103.8 $\mu \mathrm{m} \pm 4.9)$; tail $=58.6-82.4 \mu \mathrm{~m}(70.4 \mu \mathrm{~m} \pm$ 3.2); genital primordium to anterior end
$(\mathrm{n}=7)=0.31-0.45 \mathrm{~mm}(0.37 \mathrm{~mm} \pm 0.04)$. Body slender, outstretched, annulation fine; head flattened without annulation or constriction from rest of body. Stylet thin, knobs round, projecting slightly posteriorly. Cephalic framework weak. Procorpus irregular in width and continuous with median bulb, no constriction present; isthmus elongate, posterior extreme slightly wider than anterior, no constriction present at junction with esophageal glands. Nerve ring between level of metacorpus and excretory pore. Excretory pore obscure, located at level of nerve ring. Hemizonid immediately anterior to the excretory pore. Subventral glands less than one-half size of dorsal gland. Dorsal gland large with conspicuous nucleus in posterior extreme. Genital primordium oval, $14.5-19.3 \mu \mathrm{~m}$ by $5.1-8.9 \mu \mathrm{~m}$, composed of three cells, central cell largest, nucleus conspicuous, external cells small and triangular. Tail long and tapering, with acute tip.

Holotype (female): Deposited in University of California Davis Nematode Collection (UCDNC), UCNC No. 2123.

Paratypes: 11 females, UCNC No. 2124; 10 males, UCNC No. 2125; 3 second-stage larvae, UCNC No. 2126; and

1) 4 females, 2 males, and 2 second-stage larvae at Colegio de Postgraduados, Centro de Fitopatologia, Chapingo, Mexico;
2) 2 females, 2 males, and 1 second-stage larvae at Instituto de Biologia, Lab. Helmintologia, UNAM, Mexico;
3) 3 females, 1 male at Rothamsted Experimental Station, Nematology Department, Harpenden, Herts, England;
4) 2 females, 3 males at Laboratorium voor Nematologie, Landbouwhogeschool, Wageningen, Netherlands;
5) 3 females, 2 males at USDA Nematode Collection, Beltsville, Maryland.

Type locality: Pebble Beach Golf Course, San Mateo County, California.

Type habitat: Galls in the base of the stem of bluegrass (Poa annua L.).

## Differential Diagnosis

Anguina pacificae n. sp. is assigned to Anguina because of the esophageal constructions, the long multicellular columella, and the ovary with many oogonia in lateral view.
A. pacificae $\mathrm{n} . \mathrm{sp}$. is closely related to $A$.
agropyri (Kirjanova, 1955) Brezeski, 1981 and causes similar plant galls $(1,4)$. However, it differs from $A$. agropyri by the esophageal glands being well separated from the median bulb in the females of $A$. pacificae n. sp. They differ in the position of the excretory pore; in A. agropyri it is situated a little above the esophageal-intestinal valve, and in A. pacificae the excretory pore is situated at the level of posterior end of the isthmus. A. pacificae n. sp. has a sharply pointed tail tip, while A. agropyri has a conical tail tip. Males of A. pacificae n . sp. differ from $A$. agropyri by the type of bursa, in A. pacificae n. sp. the bursa ends before the tail tip. The spicules in A. agropyri have two reinforced longitudinal rays; A. pacificae n . sp. does not have these longitudinal rays. The gubernaculum in $A$. agropyri has a bifurcate anterior end and rounded terminus; in A. pacificae n . sp. the gubernaculum has slight anterior projection and ends in acute terminus. A. pacificae n. sp. is also similar to A. graminis (Steinbuch, 1799) Filipjev, 1936 in that females and males after fixation assume a $C$ shape. In this curved position the ventral surface is outermost in the males. These two species are similar in the size of females and males, and both have a low and flattened lip region. A. pacificae n. sp. differs in that the
procorpus is inflated; in A. graminis it is not markedly swollen. The tail of females and males in A. pacificae n. sp. is conical with acute tip while in A. graminis the tail is conoid with finely rounded tip. The males differ by the size of the spicules; in $A . p a$ cificae n . sp. they are $26-41 \mu \mathrm{~m}$ long and in A. graminis $40-42 \mu \mathrm{~m}$. The gubernaculum in A. pacificae n. sp. has a slight anterior projection and acute terminus; in A. graminis it is simple and concave. Finally, $A$. pacificae n. sp. causes green stem galls while A. graminis causes purple pigmented galls on the leaves of the host.

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