The Sting nematode in California:
Research and extension at UC Riverside's Nematology Department

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Fig. 1 Sting nematode damage caused severe stress and nutrient deficiency in Bermudagrass.

Fig. 2 Sting nematode feeding caused root stunting (right) compared to healthy carrots (left).

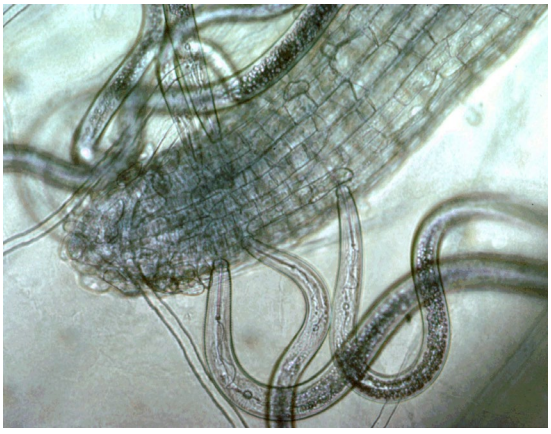

Fig. 3 Sting nematodes feeding on root tips.

In 1992, the University of California Riverside Nematologists discovered an invasive roundworm species in several golf courses in Rancho Mirage, CA. In prior years, superintendents of those properties had noticed areas of yellowing and dying turfgrasses as soon as the early spring sun warmed up the soil to about 70˚F (Fig. 1).

The news of the presence of the Sting nematode caused considerable concern among the horticultural and agricultural communities and regulatory agencies. The nematode was known as a severe pest with a vast host range in the sandy soils of its native southeastern US. The risk potential of the invasive nematode in the inland desert regions of Southern California was considered high for turfgrasses and many other economically important plants (Fig. 2). Fortunately, the dire predictions have not become a reality. In the following, we will briefly summarize our research findings that supported the outreach efforts.

The Sting nematode (*Belonolaimus longicaudatus*) is a microscopic roundworm that feeds by puncturing root cells with its mouth stylet and withdrawing nutritional content. The nematodes feed on the outside near the root's tip (Fig. 3). Their relatively long mouth stylet damages the growing tip region responsible for the root growth and may lead to cell death and stunting of the roots. The parasitism often predisposes the damaged tissues to secondary infections by soil fungi and bacteria. The injuries interfere with normal root functions such as uptake of water and nutrients while the microbes accelerate root decay.

The California Department of Food and Agriculture (CDFA) initially classified the Sting nematode as a class "Q" rated pest suspected of economic importance but uncertain because of inadequate

Fig. 4 The UC Riverside Nematology Quarantine and Containment Facility

Fig. 5 In vitro culture of corn roots with Sting nematodes

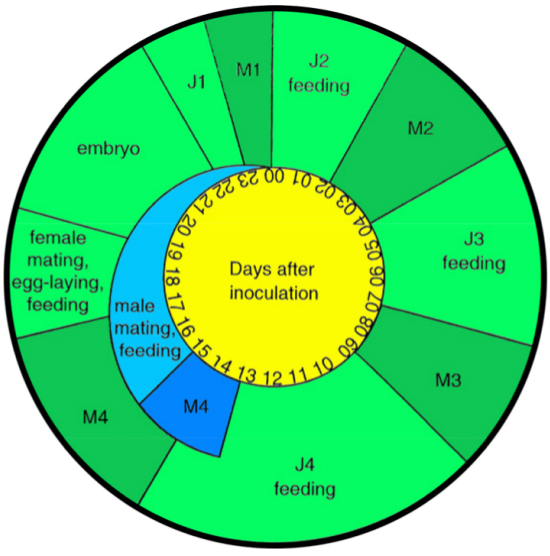

Fig. 6 The life cycle of the Sting nematode

information. A class "A" rating would have required state-enforced action such as quarantine regulation and eradication efforts. Instead, infested properties had to comply with state and county-enforced compliance agreements. Mowers and other turfgrass machinery had to be thoroughly cleaned before leaving the property. Soil, turfgrass, or other plant debris from infested properties had to be decontaminated by appropriate treatments, such as approved composting procedures. The UC Riverside Nematology Quarantine and Containment Facility was the only location in the state that A or Q-rated nematode pests could be cultured and investigated safely (Fig. 4).

Surprisingly, the life cycle of the Sting nematode was unknown, even though the pest was first described more than 40 years earlier. Rearing the nematode on corn roots in a Petri dish with a transparent tissue culture medium allowed us to observe its complete life cycle with dissecting scopes and an inverted microscope (Fig. 5). Males were attracted to females, which indicated the presence of pheromone production. The minimum time for one generation was 24 days at 82˚F (Fig. 6); the heat sum requirement was approximately 375-degree days (base temperature 12.8˚C).

Further year-long studies on Sting nematode-infested golf courses revealed the population dynamics and provided clues to mitigate the nematode's activities. Understanding the life cycle of Sting nematode and seasonal population dynamics helped pinpoint the most efficacious timing for using biological or chemical treatments.

To evaluate the potential threat to agriculture, we determined the host status of 60 different plant species and cultivars to the California population of the Sting nematode. Almost all tested plants allowed reproduction of the pest. Only tobacco, okra, and watermelon qualified as non-hosts.

The Sting nematode was likely introduced with sod or ornamental plants from the southeastern US to Rancho Mirage, CA when billionaire Walter Annenberg and his wife Leonore turned the 200-acre estate Sunnylands into an exclusive private golf course. The property was frequently visited in the

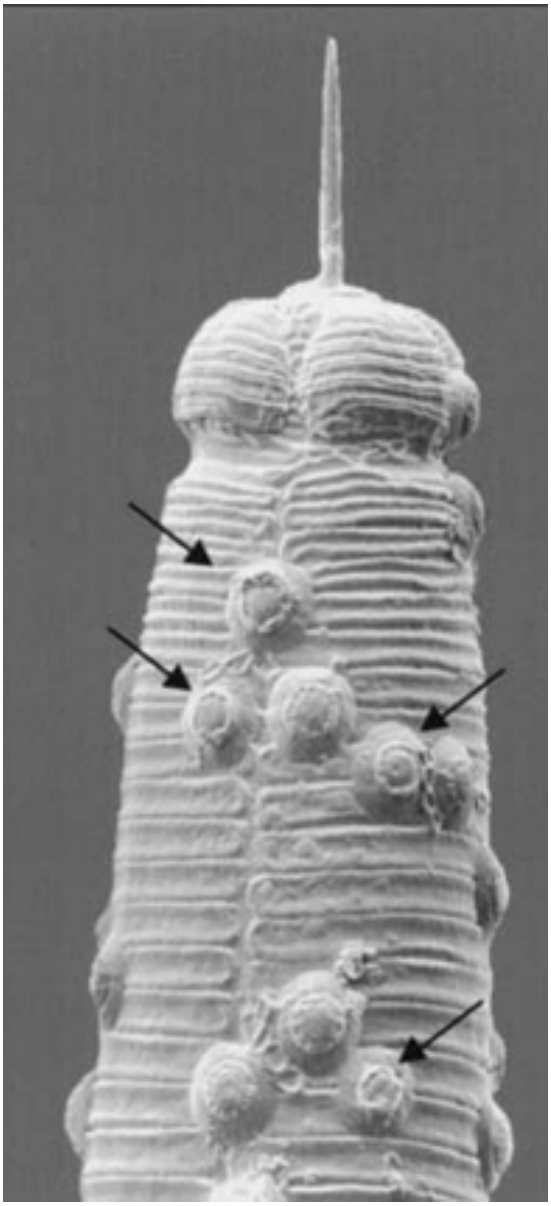

Fig. 7 Head region of a Sting nematode with spores of *Pasteuria usga* attached (© R. Giblin-Davis, University of Florida).

Fig. 8 Presentation at UCR Turf Day

1980s by US President Ronald Reagan and his wife, Nancy. An anecdotal story suggests that before one of their visits, sandy soil was excavated from the lawn for some security feature and dumped outside the property's pink wall. It was up for grabs, and several adjacent golf clubs used it to build mounts where years later, "spring decline" symptoms appeared, and Sting nematodes were subsequently found. Our laboratory obtained several Sting nematode populations from various geographical locations (North Carolina, Georgia, Arkansas, and Florida) to compare specific DNA sequences with those from populations collected from 5 different golf courses at or near Rancho Mirage. Preliminary results indicated a high genetic variation among the southeastern populations, while all Coachella Valley populations appeared closely related. The results strongly suggested a single introduction event, followed by dissemination to neighboring golf courses.

In collaboration with Nematologists at the University of Florida who discovered a new bacterial parasite of the Sting nematode in the late 1990s, we modified our Petri dish culture method to include the novel hyperparasite. The technique allowed key observations during parasitism and the life cycle of the bacteria for which the name *Pasteuria usga* was suggested. The bacteria spores attached to the nematode's skin (Fig. 7) and penetrated its host via an infection peg. *Pasteuria* grew throughout the body, eventually killing the nematode while producing new spores. Thousands of spores were released during the decomposition of the dead nematode, ready to attack the remaining Sting nematodes. The biotech company Pasteuria Bioscience developed a method to rear the previously unculturable bacteria in liquid media in large fermenters. In 2010, a commercial formulation of the biocontrol agent was sold as Econem™ for Sting nematode control in turfgrasses. In 2012, the agrochemical company Syngenta acquired Pasteuria BioScience and replaced Econem™ with a different *Pasteuria* product (Clariva®) for soybean cyst nematode control. The production of Econem™ was discontinued.

Another significant aspect of our Nematology program has been the outreach with science-based

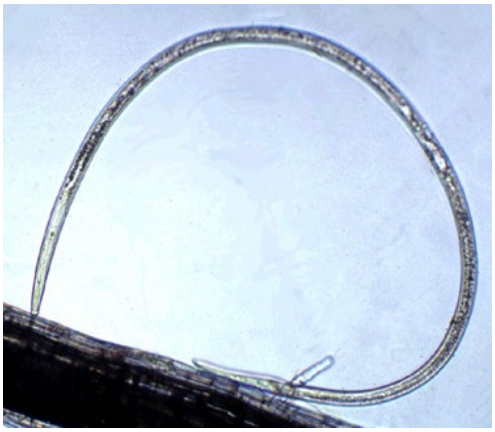

Fig. 9 Sting nematode feeding on a plant root.

information through UC Cooperative Extension to golf course superintendents, pest control advisers, industry personnel, and homeowners to mitigate the risk of further spread of this nematode in California. Over the past three decades, we have provided information through talks and poster presentations, newsletters, websites, and discussions during the annual UCR Turf Day (Fig. 8, 9).

In conclusion, we look back at a successful research and extension program targeting the Sting nematode in California. Since its discovery 30 years ago, no report of additional infestations in any California crop has been reported.