

Observations on a *Xiphinema insigne* Population with Several Males from Hangzhou, China¹

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Abstract: A population of *Xiphinema insigne* with several males was found on the campus of Zhejiang University, Hangzhou, China, in November 1998 in the rhizosphere of Yulan Magnolia. Morphometrics of nine males and 25 females of this population are given herein. No sperm were found in the genital tracts of 118 females that were examined. This agrees with all other observations reported for this species except for the synonym of *X. insigne*, *X. neodimorphicaudatum*, in which males are as common as females and the females genital tracts contain numerous sperm. In the males of this study population, sperm were observed only in the seminal vesicles at the distal end of the testes before their juncture with the vas deferens. The population did not fit within either of the two forms of the species. This and other populations have filled in the gaps between the two forms, making the morphometrics for all reported populations a continuum.

Key words: China, males, morphometrics, nematode, sperm, taxonomy, ultrastructure, *Xiphinema insigne*.

Xiphinema insigne was first described as *X. insignis* Loos, 1949 from Ceylon (Sri Lanka). Lordello (1953) modified the name to its present form in accordance with the rules of Zoological Nomenclature. Tarjan and Luc (1963) designated *X. indicum* Siddiqi, 1959 as a junior synonym of *X. insigne*, and Luc et al. (1984) considered *X. neodimorphicaudatum* Khan, 1982 and *X. tugewai* Darekar & Khan, 1983 as junior synonyms of *X. insigne*. The species is distributed widely with reports from Sri Lanka, Japan, United States (North Carolina, California), Philippines, Mauritius, Malawi, Thailand, India, Pakistan, Israel (Luc and Southey, 1980), Malaysia (Razak and Loof, 1998), Korea (Choi et al., 1992), Egypt (Oteifa and Tarjan, 1965), South Africa (Heyns, 1991), and China (Xu et al., 1995). Lal et al. (1982) reported *X. insigne* reproduced well and was pathogenic to grape, causing chlorosis, swollen root tips, blackened root tips, and significant reductions in shoot height, shoot weight, root weight, leaf number, and leaf area in pot

experiments in India. This species has not been implicated as a virus vector.

In the synonymy of *X. indicum* with *X. insigne* by Tarjan and Luc (1963), *X. indicum* had a slightly longer, but overlapping, total stylet length (158 to 167 μm vs. 146 to 159 μm) and on average a shorter tail (80 μm vs. 97 μm) than *X. insigne*. They stated, "We believe there are no valid differences between *X. insigne* and *X. indicum* and that these two species may be regarded as conspecific." Cohn and Sher (1972) accepted Tarjan and Luc's synonymy. In a table, Southey (1973) designated, with no text as to why, two forms of *X. insigne*—indicum type and long-tail type. Bajaj and Jairajpuri (1977) accepted this view in a study of 23 *X. insigne* populations from India in which they indicated that 18 of the populations were of the indicum-form and five were of the insigne-form. They gave ranges for the indicum-form odontostyle length (97 to 110 μm), odontophore length (57 to 64 μm), and tail length (66 to 90 μm) and for the insigne-form odontostyle length (80 to 95 μm), odontophore length (55 to 59 μm), and tail length (88 to 156 μm). Phukan and Sanwal (1982) reported two populations of each form in Assam, India. Luc and Southey (1980) and Razak and Loof (1998) reported populations with intermediate measurements.

Males of *X. insigne*, while rare, have been described from populations from Japan, United States (California) (Luc and

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Southey, 1980), India (Phukan and Sanwal, 1982; Bajaj and Jairajpuri, 1977), Israel (Cohn, 1969), Trinidad (Hunt and Singh, 1984), and Malaysia (Razak and Loof, 1998). The structure of the male genital branch was described, and it was noted that sperm were never observed in the female genital tract (Jairajpuri and Bajaj, 1978). Conversely, *X. neodimorphicaudatum* Khan, 1982, considered a junior synonym of *X. insigne* by Luc et al. (1984), has more males than females in the type collection, and in females the "anterior part of the uterus is filled with a large number of minute rounded sperms" (Khan, 1982).

The first reports of *Xiphinema insigne* from China were by Fang et al. (1994) from Guanzhou and Guangdong provinces and Xu et al. (1995) from Zhejiang, Jiangsu, Guansi, and Fujian provinces. Since then, the species has been reported from Sichuan (Wang et al., 1996), Inner Mongolia (Han et al., 1997), and Hebei (Peng, pers. comm.) provinces.

During a workshop on Virus Vectoring Nematodes at the Institute of Biotechnology, Zhejiang University, Hangzhou, China, in November 1998, a population of *Xiphinema insigne* with a ratio of 1 male: 13.1 females was discovered. The presence of males in the population provided an excellent opportunity to look for sperm in the genital tract of females as evidence of insemination, which, along with reporting the morphometrics of the males and females, is the main objective of this study.

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MATERIALS AND METHODS

Nematodes were extracted from the soil collected from the rhizosphere of Yulan Magnolia using the decanting and sieving method. *Xiphinema insigne* specimens were handpicked from the samples, heat-killed, fixed in 2% formalin, processed to anhydrous glycerine by a modification of Seinhorst's rapid method, mounted on glass slides, and examined with a high-resolution light microscope with Nomarski differential interference contrast. Morphometrics of males and females were measured with the aid of an ocular micrometer.

RESULTS

A total of nine male and 118 female *Xiphinema insigne* specimens were examined. Morphometrics of all nine males and 25 of the females are given in Table 1. Measurements (μm) of all 118 females were made for length of odontostyle (102 ± 1.9 (97 to

TABLE 1. Morphometrics of characters of nine males and 25 females of *Xiphinema insigne* from Yulan Magnolia on the campus of Zhejiang University, Hangzhou, China.

Character	Males	Females
<i>n</i>	9	25
Length	2,583 \pm 148 (2,318–2,797)	2,617 \pm 149 (2,241–2,875)
Distance from lip to guide ring	93 \pm 4.2 (85–97)	96 \pm 2.2 (90–99)
Odontostyle length	102 \pm 4.1 (93–107)	102 \pm 2.2 (97–106)
Odontophore length	63 \pm 1.3 (61–65)	63 \pm 2.4 (58–67)
Total stylet length	165 \pm 5 (156–171)	165 \pm 3.5 (158–171)
Tail length	69 \pm 10 (57–83)	128 \pm 13.8 (83–146)
Hyaline length of tail	16 \pm 4.4 (8–20)	17 \pm 4.1 (9–23)
Spicule length (arc)	49 \pm 1.6 (47–52)	—
Lip width	11 \pm 0.6 (10–12)	11.5 \pm 0.7 (10.2–12.2)
Mid-body width	36 \pm 2.7 (31–39)	39 \pm 2.9 (33–43)
Anal-body width	29 \pm 1.1 (27–31)	23 \pm 1.2 (20–24)
T/V (% of body length)	59 \pm 4 (52–63)	33 \pm 1.2 (31–36)
Ratios:		
a	71 \pm 5.3 (63–81)	68 \pm 3.7 (61–77)
b	6.7 \pm 0.5 (6.1–7.7)	6.7 \pm 0.6 (5.9–8.5)
c	38 \pm 4.8 (30–49)	21 \pm 2.9 (17–30)
c'	2.4 \pm 0.4 (1.9–3.0)	5.6 \pm 0.6 (4.1–6.5)

All measurements are in μm ; data given is mean; standard deviation; and range is in parentheses.

106)), odontophore (63 ± 2.2 (57 to 68)), total stylet (165 ± 2.8 (158 to 171)), tail (127 ± 11 (83 to 148)), anal body diameter (23 ± 1.1 (20 to 24)), and ratio c' (tail length/anal body width) (5.6 ± 0.5 (3.7 to 6.8)). Of the 118 females studied, in no instance were sperm found in the female genital tract. A typical female anterior region showing the entire stylet is shown (Fig. 1A). A typical female tail is shown (Fig. 1B) as is an abnormally short female tail, 58 μm long and bluntly rounded (Fig. 1C). In males, the number of ventromedian supplements num-

bered 5 (4 specimens), 6 (4 specimens), or 7 (1 specimen) with an average of 5.7 per male. In males, sperm were found only in the seminal vesicle (distal) area of the testes; no sperm were found in the vas deferens connecting the cloaca to the juncture of the testes. Male tails varied in shape from almost digitate to conical (Fig. 1D-G).

DISCUSSION

The morphometrics of the 25 measured females of this population agree closely with

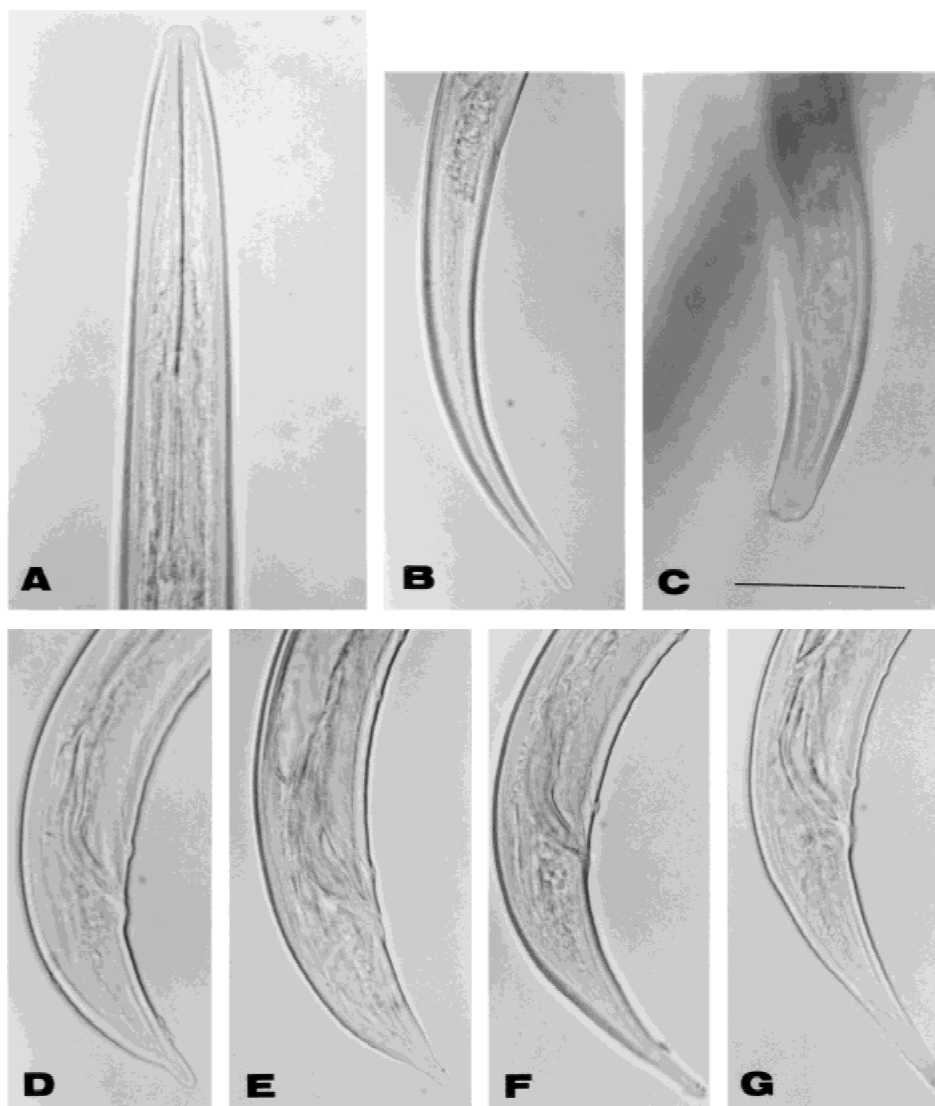


FIG. 1. *Xiphinema insigne* from China. A) Anterior of female. B) Tail of female. C) Abnormal female tail. D-G) Male Tails. Bar = 50 μm .

those of the Japanese and Fresno populations of Luc and Southey (1980). However, a few had tails that were much shorter (83 μm , 106 μm , 114 μm) than the average tail length (128 μm), with the remaining tail lengths from 120 to 144 μm . When compared to the 'forms' of Bajaj and Jairajpuri (1977), the stylet length was similar to the indicum-form and the tail length to the insigne-form, but the bodies were longer than either. The morphometric ranges of the nine males are most similar to those of the male from 'Fresno' (Luc and Southey, 1980).

The absence of sperm in the uteri of the 118 females of the study population agrees with observations on all other populations except the presumed amphimictic population described by Khan (1982) as *X. neodimorphicaudatum*, later placed in synonymy with *X. insigne* by Luc et al. (1984). This study shows that, while amphimixis is theoretically possible for *X. insigne* due to the presence of males, it is unlikely as no sperm were observed in the uteri of any of the females of this population in which males were relatively common.

Xiphinema insigne is widespread on the sub-continent of India and east Asia. Reported populations vary greatly in body length, component, and total stylet length, and in tail length, not only between populations but also often within populations. It is apparent that the population of this study is intermediate to the indicum-form and the insigne-form, based on its component and total stylet lengths being similar to the former and the tail length and c' ratio closer to the latter. This population, and several others reported, make the character gaps between the two forms a continuum.

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