

Description of *Tylenchorhynchus qasimii* sp. n. with a New Report of *T. kegasawai* Minagawa, 1995 from Pakistan

MUSARRAT RAMZAN,¹ ZAFAR A. HANDOO,² SHAHINA FAYYAZ¹

Abstract: A new stunt nematode, from soil around the roots of coconut (*Cocos nucifera* L.) and rice (*Oryza sativa* L.) from Karachi, Pakistan, is described and illustrated as *Tylenchorhynchus qasimii* n. sp. This new species is characterized by having females with 3–4 head annules, anteriorly directed stylet knobs, absence of post anal extension, presence of rounded sperm filled spermatheca and conoid to bluntly rounded hemispherical tail terminus. Males are common. Also included is the record of *T. kegasawai* from around the roots of rice (*O. sativa* L.), a new report from Sindh, Pakistan.

Key words: *Cocos nucifera* L., *Oryza sativa* L., taxonomy, *Tylenchorhynchus qasimii* n. sp., *Tylenchorhynchus kegasawai*.

Stunt nematodes (*Tylenchorhynchus* spp.) are economically important plant pathogens and contain 111 valid species that parasitize a wide variety of plants (Handoo, 2000). The history of the genus and the taxonomic changes to stunt nematodes were proposed by different workers and are summarized by Handoo (2000) and Handoo *et al.* (2007). In July 2007, during a survey of rice (*Oryza sativa* L.) fields and coconut (*Cocos nucifera* L.) plantations in the Sindh region of Pakistan, one new species and an occurrence of a rare, known species of the genus *Tylenchorhynchus* were found. The infected plants showed symptoms of nematode damage such as root and stem discoloration, bark damage and chlorosis of leaves.

The objectives of this study were to describe and illustrate these two *Tylenchorhynchus* species and to assess the diagnostic value of their morphological characters. The new species is herein described as *T. qasimii*, and a new record is *T. kegasawai* Minagawa, 1995.

MATERIALS AND METHODS

Specimens of new species were obtained from soil around roots of coconut (*C. nucifera*) and rice (*O. sativa*) from the Coastal Agricultural Research Station (CARS), Southern Agricultural Research Centre (SARC), Pakistan Agriculture Research Council (PARC), Jinnah Avenue, Malir Hault, Karachi and Nasirabad, Pakistan, and the specimens of a new record (*T. kegasawai*) from soil around the roots of rice (*O. sativa*) from Nasirabad, Pakistan. Females and males were isolated from soil by sieving and Baermann funnel extraction and then killed by gentle heat, fixed in triethanolamine formalin (TAF), transferred to 1.25% glycerin solution, dehydrated for 2–5 d at 50–55°C and mounted in glycerin on permanent slides for observation and measurements (Hooper, 1986). Line drawings were made with the help of a drawing tube attached to

a compound microscope, and measurements in micrometers, unless otherwise stated, were obtained with an ocular micrometer. The nematodes were examined with a compound microscope and identified using a recent taxonomic key (Handoo, 2000).

SYSTEMATICS

Tylenchorhynchus qasimii n. sp.
(Figs. 1A–J; 2A–J, Table 1)

Female. Body arcuate to open C shape after fixation, no longitudinal striae or ridges. Body annules distinct 1.6–2.4 µm wide around midbody, 0.8–2.4 µm at oesophageal end and 1.6–2.4 µm at tail regions. Lateral fields originating at the conus of the stylet and extending up to hyaline region of tail, with four incisures, outer two incisures crenate. Lip region low, rounded, almost continuous to body contour; with three to four annules. Stylet moderately strong, shaft and conus equally long 8–8.6 µm; knobs anteriorly directed. Dorsal gland orifice (DGO) about 2.4 µm long behind stylet base. Median oesophageal bulb oblong, basal bulb pyriform. Cardia well developed. Hemizonid usually just anterior to excretory pore, 1–1.5 annules wide. Vulva a transverse slit slightly posterior to the middle of the body and distinctly protruding. Epiptygma absent. Reproductive system amphidelphic, didelphic; anterior and posterior ovaries equally developed. Spermatheca rounded, filled with rounded spermatozoa. Tail conoid to bluntly rounded hemispherical, tail terminus smooth. Phasmids located slightly anterior to middle of the tail, 12–16 annules posterior to anus. Post anal extension of intestine absent.

Male. Males abundant, morphologically similar to female except for sexual characters, and body in posterior region usually more curved than in female. Gubernaculum well developed, half of the spicule length or slightly longer. Tail terminus pointed.

Paratypes (Male and female): Slide T-5633p (2 female, 2 male) and T-5634p (3 female, 4 male) deposited in the USDA Nematode Collection (USDANC) at Beltsville, MD, USA, and holotype (female and male) on slide NNRC 118/734–740 deposited in the National Nema-

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¹ National Nematological Research Centre, University of Karachi, Karachi – 75270, Pakistan.

² Nematology Laboratory, USDA, ARS, Beltsville, MD 20705.

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E-mail: shahinaku@yahoo.com

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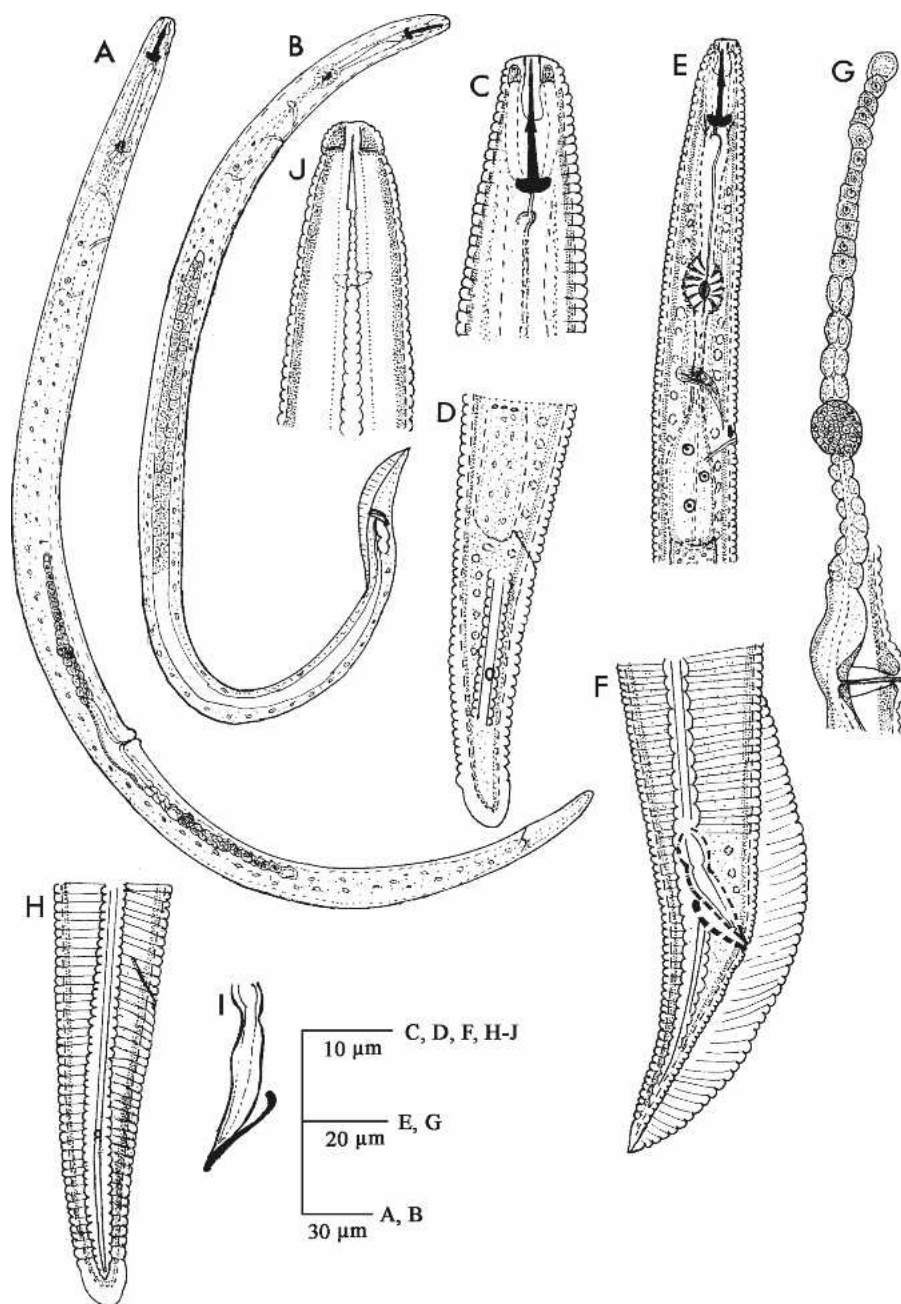


FIG. 1. *Tylenchorhynchus qasimii* n. sp. A. Entire female body; B. Entire male body; C. Anterior region; D. Female tail; E. Oesophageal region of female; F. Male tail; G. Anterior gonad of female; H. Female tail showing phasmid; I. Spicule; J. Anterior region showing origin of lateral field.

tode collection at NNRC, University of Karachi, Pakistan.

Type host and locality: Collected in June 2007 by one of us (Shahina Fayyaz) from soil around roots of coconut (*Cocos nucifera* L.) from field of CARS, SARC, PARC, Jinnah Avenue, Malir Hault, Karachi and rice (*Oryza sativa* L.) field of Nasirabad, Sindh, Pakistan.

Relationship and diagnosis: *Tylenchorhynchus qasimii* n. sp. is distinctive because of its slightly anteriorly directed stylet knobs, absence of the post anal extension, presence of sperm filled spermatheca, shape of tail and presence of males.

Specimens of *T. qasimii* n. sp., compared with the

existing species of the genus, share similarities to *T. varicaudatus* Singh, 1971, *T. haki* Fotedar and Mahajan, 1971, and *T. mashhoodi* Siddiqi and Basir, 1959 (comparative data in Table 2). Differences from *T. haki* include: the shape of stylet knobs, being slightly anteriorly pointed vs. rounded; presence of a sperm-filled spermatheca vs. spermatheca empty; position of phasmid slightly anterior to middle of the tail vs. just posterior to anus; absence of the post-anal extension vs. presence of the post-anal extension; longer tail ($c = 15.7\text{--}18.5$ vs. $20\text{--}27$); and males present vs. males not known. *T. qasimii* n. sp. differs from *T. varicaudatus* by number of lip annules ($3\text{--}4$ vs. 2); presence of sperm-filled sper-

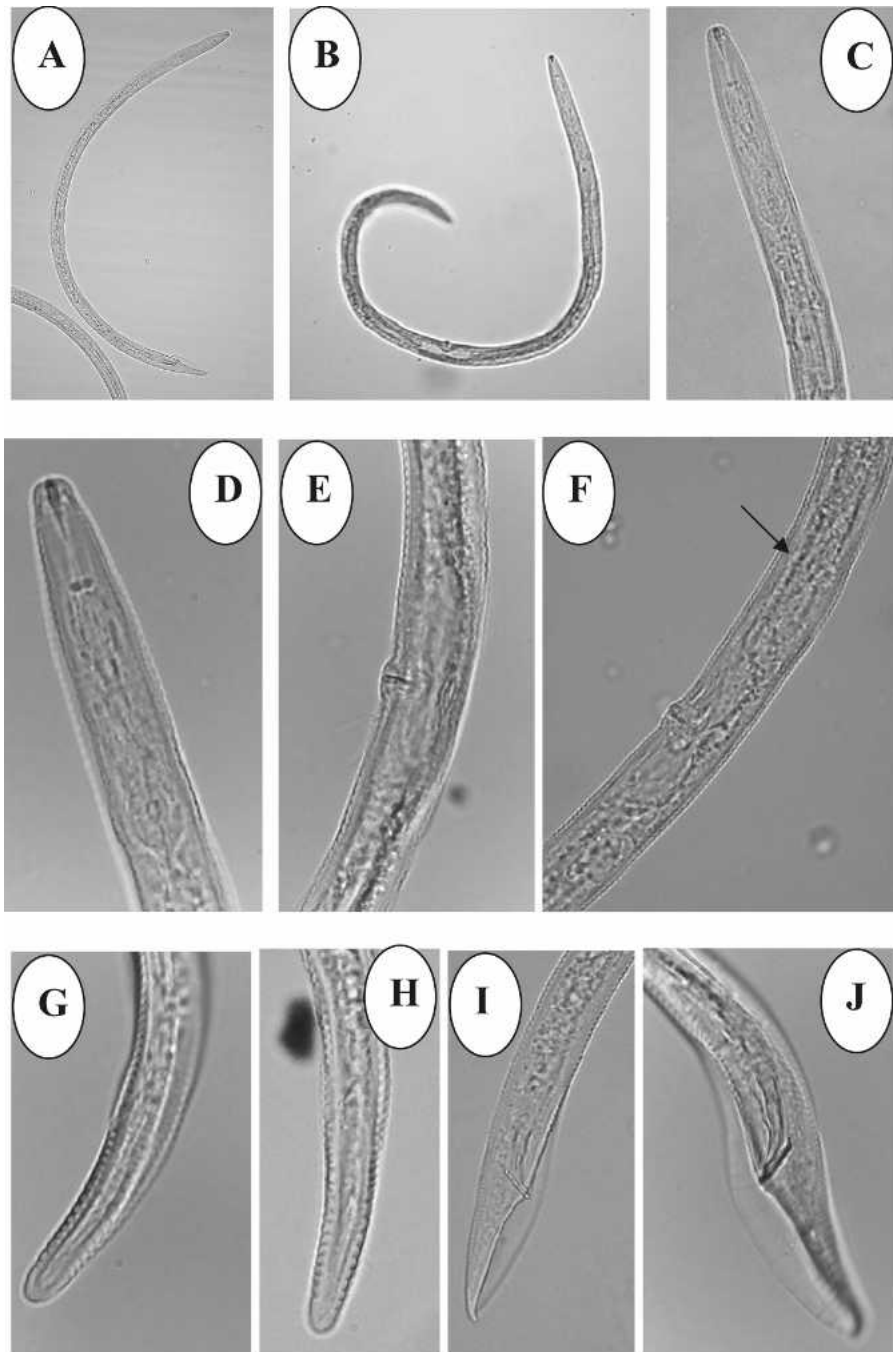


FIG. 2. Light micrographs of male and female *Tylenchorhynchus qasimii* n. sp. A. Total body of male; B. Total body of female; C. Anterior portion of female; D. Anterior region showing shape of stylet knob; E. Vulva region; F. Vulva region showing spermatheca (arrow); G, H. Female tails; I, J. Male tails.

matheca vs. absence of spermatheca; greater \acute{c} ratio (3–3.4 vs. 2.3–2.8); presence of males vs. absence; and shape of tail terminus (hemispherical vs. bluntly conical). Differences from *T. mashhoodi* include a slightly shorter stylet (15–17.6 vs. 16–20); the shape of stylet knobs are slightly anteriorly pointed vs. generally large rounded to posteriorly directed; the lateral field originates from conus of stylet vs. from base of stylet; presence of a distinctly protruding vulva vs. not protruding; and rectum length more than half of anal body width vs. half or less than anal body width and prominent phasmids (distinct but small in *T. mashhoodi*).

Etymology: The species name is given in honor of the Vice Chancellor of Karachi University, Pakistan, Prof. Dr. Pirzada Qasim Raza Siddiqui.

Tylenchorhynchus kegasawai Minagawa, 1995
(Table 3)

Female: Body slender, slightly ventrally curved after treatment by gentle heat; no longitudinal striae. Cuticular annulation distinct 2.4–3.2 μ m near mid body. Lateral field areolated, occupying about 1/3 of body width,

TABLE 1. Measurements (μm) of the holotype female, allotype male and female and male paratypes of *Tylenchorhynchus qasimii* n. sp.

Linear measurements	Holotype female	Allotype male	Female paratypes Mean \pm SD (Range)	Male paratypes Mean \pm SD (Range)
n	1	1	15	10
L	617.6	595.2	595 \pm 24.6 (566–622)	586 \pm 21 (520–609)
Body width	20	15.2	18.0 \pm 1.75 (15.2–20)	16 \pm 1.2 (14.4–17.6)
Esophagus length	128	115.2	121.6 \pm 4.48 (116–128)	117.1 \pm 4.02 (111.2–121.6)
Stylet length	17.6	16	16.2 \pm 0.65 (15–17.6)	16.1 \pm 0.66 (15.2–16.8)
Stylet knob width	4	4	4.16 \pm 0.35 (4.0–4.8)	4.0 \pm 0 (4.0)
Median bulb from anterior end	68	67.2	65.2 \pm 2.4 (62.4–68)	66.4 \pm 2.03 (63.2–68)
Median bulb width	8	8	8.64 \pm 0.87 (8–9.6)	(8.16 \pm 0.35 (8.0–8.8))
Excretory pore from anterior end	102.4	97.6	99.6 \pm 3.4 (94–103)	96 \pm 1.69 (94–98)
Tail length	38.4	36	35.2 \pm 2.93 (31.2–38.4)	38.4 \pm 2.7 (35.2–41.6)
Anal body width	11.2	9.6	10.9 \pm 0.35 (10.4–11.2)	11.52 \pm 1.65 (9.6–13.6)
Spicule length	—	22.4	—	19.36 \pm 1.82 (17.6–22.4)
Gubernaculum length	—	11.2	—	11.0 \pm 0.66 (10.4–12)
Lip width	6.4	6.4	6.5 \pm 0.35 (6.4–7.2)	6.24 \pm 0.35 (5.6–6.4)
Lip height	4	3.2	3.36 \pm 0.35 (3.2–4.0)	3.2 \pm 0 (3.2)
Lip annules	4	3	3–4	3
Tail annules	16	—	16.6 \pm 2.5 (13–29)	—
Ratios				
a	30.8	39.1	33.0 \pm 2.58 (30.8–37.2)	36.8 \pm 3.71 (35.6–41.5)
b	4.8	5.1	4.84 \pm 0.11 (4.7–5.0)	4.96 \pm 0.20 (4.6–5.1)
c	4.8	16.5	16.9 \pm 1.15 (15.7–18.5)	15.26 \pm 1.03 (14.2–16.5)
\hat{c}	16.0	3.7	3.18 \pm 0.17 (3.0–3.4)	3.32 \pm 0.28 (3.0–3.7)
Percentage				
V%	57.6	—	57.9 \pm 0.63 (57.7–58.8)	—

formed by 4 incisures, outer ones distinctly crenate. Lip region low, flattened to rounded, almost continuous to body contour, with 3 annules, labial framework moderately sclerotized. Stylet sclerotized, knobs well developed, rounded, extending laterally or slightly posteriorly. Median oesophageal bulb oblong, 56–68 μm from anterior end, basal bulb pyriform, not overlapping intestine. Excretory pore at about the middle of isthmus. Hemizonid usually just anterior to excretory pore, one annule wide. Vulva slit-like, epiptygma absent, ovaries outstretched; spermatheca rounded to elongate oval,

filled with sperm; tail clavate; terminus rounded and smooth, phasmid in anterior half of tail. Post-rectal intestinal sac absent.

Male: Similar to female except for sexual characters and shorter body. Testis outstretched, bursa tylenchoid, finely crenate, enveloping entire tail. Spicule robust, terminus minutely bifurcate. Gubernaculum thin, proximal part directed anteriorly or occasionally bent around middle portion. Tail terminus pointed, phasmids in anterior half of tail.

Remarks: *Tylenchorhynchus kegasawai* Minagawa, 1995

TABLE 2. Comparative morphometric data (in μm) of closely related species of *Tylenchorhynchus qasimii* n. sp.

Species	<i>T. varicaudatus</i> ^a	<i>T. haki</i> ^b	<i>T. mashhoodi</i> ^c	<i>T. qasimii</i> n. sp
L	0.50–0.56	0.50–0.63	0.61–0.76	0.56–0.62
Stylet length	17–18	16–18	17–18	15–17.6
Stylet knob shape	Anteriorly directed knobs	Rounded	Posteriorly directed	Slightly anteriorly pointed
Spicule length	—	—	22–24	17–22.4
Gubernaculum length	—	—	12–13	10–12
Tail shape	Cylindrical	Subcylindrical	Ventrally arcuate	Lozenge shape
Post anal extension	—	Present	Absent	Absent
Lip annule	2	3	3	3–4
Tail annule	14–15	14–20	13–16	13–29
Ratios				
a	28–33	26–33	25.8–30	30.8–37.2
b	4.3–5.2	5.0–5.9	4.9–5.5	4.7–5.0
c	16–18.6	20–27	16–19.4	15.7–18.5
\hat{c}	2.3–2.8	—	—	3–3.4
Percentage				
V%	57–59	56–57	55–58	57–58

^a Singh, 1971.

^b Fotedar and Mahajan, 1971.

^c Siddiqi and Basir, 1959; Siddiqi, 1961.

TABLE 3. Measurements (μm), ratios and percentages of *T. kegasawai* Minagawa, 1995

Linear measurements	Female n = 3 Mean \pm SD (Range)	Male n = 2
L	679.8 \pm 64.8 (637–754)	573.6, 649.6
Body width	21.8 \pm 1.84 (20.8–24.0)	16.8, 17.6
Esophagus length	141 \pm 6.1 (136–148)	125.6, 128
Stylet length	21.8 \pm 1.66 (20–23.2)	16.8, 20.8
Stylet knob width	4.26 \pm 0.46 (4.0–4.8)	3.2, 4
Median bulb from anterior end	63.4 \pm 4.10 (60–68)	66
Median bulb width	10.1 \pm 0.46 (9.6–10.4)	9.6
Excretory pore from anterior end	105 \pm 8.18 (96–112)	—
Tail length	55.2 \pm 2.26 (53.6–56.8)	45.6
Anal body width	14.0 \pm 0.56 (13.6–14.4)	13.6, 14.4
Spicule length	—	23.2, 24.8
Gubernaculum length	—	11.2
Lip width	6.13 \pm 0.46 (5.6–6.4)	5.6, 6.4
Lip height	3.7 \pm 0.46 (3.2–4.0)	3.2, 4
Lip annules	3	3
Tail annules	19–22	—
Ratios		
a	31.0 \pm 0.40 (30.6–31.4)	34.1, 36.9
b	4.7 \pm 0.20 (4.6–5.0)	4.5, 5.0
c	12.5 \pm 0.7 (11.8–13.2)	12.5, 14.2
c̄	3.9 \pm 0 (3.9)	3.1, 3.3
Percentage		
V%	51.6 \pm 2.26 (50–53.2)	—

represents the first record of this species from Pakistan. Morphological and morphometric characters fit the original description of *T. kegasawai* except for few differences which may be due to ecological variation. The Pakistan specimens have longer body length (573–754 vs. 523–631 μm), in male greater “a” ratio (a = 34.1–36.9 vs. 25.5–29.6) and basal bulb not overlapping intestine as compared to basal bulb slightly overlapping to intestine in population described by Minagawa (1995).

Since to the original description, several reports of variations within *T. mashhoodi* Siddiqi and Basir, 1959 have been published (Baqri and Jairajpuri, 1970, Samsoen and Geraert, 1975, Ray and Das, 1983, Kleynhans and Heyns, 1984, Khan and Khan, 1997, Pathak and Siddiqui, 1997), leading to significant expansion of the range of morphometric data for this species. However, we do not agree here with Baqri and Jairajpuri (1970) on some of the detailed intra-specific variations in tail and stylet shapes of some of the nine populations they reported of *T. mashhoodi*. From a careful review of that study (page 63, Figs. 6, 11 and 17), it is quite evident that the authors were dealing with a mixed population of *T. mashhoodi* and *T. annulatus* and accordingly had synonymized *T. martini* with *T. mashhoodi*. However, *T. martini* is now a synonym of *T. annulatus*. Anderson and Potter (1991) reported that *T. mashhoodi* has been confused with *T. elegans* or other similar species occurring in the tropics or subtropics and that subsequent redefi-

nitions of *T. mashhoodi* by Singh and Khera (1978), Ray and Das (1983), Maqbool et al. (1983), Kleynhans and Heyns (1984) and Gupta and Uma (1985) similarly are more representative of *T. elegans* or other close species and have suggested that records of distribution and pathogenecity reported for *T. mashhoodi* may be attributed to other species.

Keeping in view our observations and above remarks related to variations reported for *T. mashhoodi*, it is concluded that *T. qasmii* sp. n. clearly differentiates from *T. mashhoodi*, and accordingly we describe here this new species from Pakistan and also present extensive morphometric data on population of *Tylenchorhynchus kegasawai* Minagawa, 1995.

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