

Nematode fauna of Kurram Agency, Pakistan

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Abstract

The main aim of this study was to ascertain the diversity of plant parasitic nematodes at different locations of Kurram Agency, Pakistan. For this purpose, surveys were conducted and 150 samples of root and soil were collected from different locations of Kurram Agency. The detail morphological and taxonomical studies revealed a total of 26 species of plant parasitic nematodes belong to 17 genera, 13 families, 15 subfamilies and 3 orders while free-living soil nematodes revealed a total of 21 genera, 17 families and 8 orders. Overall percentage of plant parasitic and free-living nematodes was 40% and 60%, respectively. *Irantylenchus clavidorus* Kheiri, 1972 was encountered with highest occurrence (40%) followed by *Aphelenchus avenae* Bastian, 1865 (27.5%) and *Ditylenchus myceliophagus* Goodey, 1958 (10%). Morphological and detailed taxonomical studies of nematodes were carried out and six new records of plant parasitic nematodes viz., *Aphelenchoides richardsoni* Grewal, Siddiqi & Atkey, 1992, *Aphelenchoides unisexus* Jain & Singh, 1984, *Filenchus microdorus* Chawla, Prasad, Khan & Nand, 1969, *Helicotylenchus urobelus* Anderson, 1978, *Hemicriconemoides brachyurus* (Loos, 1949) Chitwood & Birchfield, 1957 and *Irantylenchus clavidorus* Kheiri, 1972 were reported. All these species were re-described with their measurements as well as illustrations. In addition, 17 new host records were reported viz., *Aphelenchoides besseyi* Christie, 1942, *Aphelenchoides goodeyi* Siddiqi & Franklin, 1967, *Aphelenchoides siddiqii* Fortuner, 1970, *Aphelenchus avenae* Bastian, 1865, *Basiria graminophila* Siddiqi, 1959, *Boleodorus pakistaniensis* Siddiqi, 1963, *Ditylenchus myceliophagus* Goodey, 1958, *Helicotylenchus willmottae* Siddiqi, 1972, *Heterodera schachtii* Schmidt, 1871, *Heterodera zaeae* Koshy, Swarup & Sethi, 1971, *Merlinius nagerensis* Sagir & Erum, 2017, *Pratylenchus flakkensis* Seinhorst, 1968, *Psilenchus hilarulus* de Man, 1921, *Quinisulcius capitatus* (Allen, 1955) Siddiqi, 1971, *Seinura oostenbrinki* Hussain & Khan, 1967, *Xiphinema brevicolle* Lordello & De Costa, 1961 and *X. index* Thorne & Allen, 1950. Population analysis of nematode fauna encountered from Kurram Agency was also determined.

Keywords: Survey, population analysis, taxonomy, new records, Kurram Agency

Agriculture is economically very important for every country. Pakistan is an agricultural country and about 70% people livelihood depends on agriculture directly or indirectly. Parachinar is the capital of Kurram Agency and the largest city of FATA (Federally Administrated Tribal Areas) of Pakistan. Geographically FATA is in the west of Pakistan. FATA have seven Agencies: Bajour, Muhamand, Khyber, Kurram, Orakzai, North Waziristan and South Waziristan; with six frontier regions: Bannu, Dera Ismail Khan,

Kohat, Lakki, Peshawar and Tank. The total area of Kurram Agency is 3, 38000 hectares with 1705 m altitude. Kurram Agency is mountainous and plain area and has moderate climate with much higher rainfall and the snowfall is common in winter season. The soil of Kurram Agency is clay form and fertile. The main water source is Kurram River with some small dams, tube wells, dug wells, springs and glaciers, water of which is used for irrigation. Kurram Agency is famous for walnuts, peanuts, fresh fruits and fresh vegetables. Main cereals of Kurram

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Agency are wheat, rice, maize and different beans (soya, mung and kidney beans). Due to lack of knowledge and data about pests, pathogens and nematodes, farmers of Kurram Agency lose the yield and quality of their farm produce. The crops are attacked by many major pathogens that cause different crop diseases. Plant parasitic nematodes are economically very important pests of cereals, fruits, vegetables, nuts and ornamentals in all over the world including Pakistan. The present research study comprised of survey, taxonomic and morphological studies and population analysis of nematode species: which provided the knowledge about biodiversity of nematodes species associated with important crops of Kurram Agency, Pakistan. Morphologically, the specimens correspond rather well with original description. However, it was deemed worthwhile to prepare complete illustration from this new material.

Material and Methods

Surveys were conducted during 2017-2018, to study the diverse nematode fauna associated with different crops of Kurram Agency. Collected soil and root samples were processed and nematodes were extracted by Cobb's wet sieving technique (Cobb, 1918) followed by a modified Baermann funnel method (Baermann, 1917). Encountered nematodes were heat killed, fixed in double-strength TAF (Tri-ethanol-amine Formaldehyde) containing 8% formalin and 2% tri-ethanol-amine in distilled water and mounted in dehydrated glycerin (Siddiqi, 2000). Illustrations were made by using a drawing tube attached to the compound microscope Nikon Eclipse E400 and photographed with a Nikon DS, Film camera, attached to the same microscope.

Results and Discussion

Sites of collection: During the present surveys a total of 150 soil and root samples were collected for the prevalence of plant parasitic and free-living soil nematodes from eleven different localities of Kurram Agency (Fig.1). Surveyed localities with their GPS were Agra, Alamsher, Borki, Dangila, Karman, KunjAlizai, Lalmai, Shublan, Shakh, Shingak and Zera (Table 1).



Fig. 1. Map of Kurram Agency, Pakistan.

Table 1. Surveyed sites of Kurram Agency and their G.P.S.

S.No	Localities	Latitude	Longitude
		N°	E°
1	Agra	33.81119	70.18214
2	Alamsher	33.84907	70.16849
3	Borki	33.85981	69.9374
4	Dangila	33.87098	70.22194
5	Karman	33.89319	70.12128
6	KunjAlizai	33.81801	70.02356
7	Lalmai	33.8634	69.96371
8	Shublan	33.81877	70.17032
9	Shakh	33.8362	70.08048
10	Shingak	33.83659	69.98328
11	Zeran	33.91959	70.12621

Prevalence and distribution of nematodes

The results of these samples showed prevalence of 47 nematode species belonging to 37 genera, 29 families and 10 orders *viz.*, Tylenchida, Aphelenchida, Rhabditida, Dorylaimida, Enoplida, Triplonchida, Mononchida, Monhysterida, Areolaimida and Chromadorida. A complete list of nematode species found during the study is given here.

List of nematodes species discovered during the present study

Achromadora spp.
Alaimus spp.
Amphidorylaimus spp.
Aphelenchoides besseyi Christie, 1942**
Aphelenchoides goodeyi Siddiqi & Franklin, 1967**
Aphelenchoides richardsoni Grewal, Siddiqi & Atkey, 1992*
Aphelenchoides siddiqii (Fortuner, 1970) Siddiqi, 1980**
Aphelenchoides unisexus Jain & Singh, 1984*
Aphelenchus avenae Bastian, 1865**
Basiria graminophila Siddiqi, 1959**
Boleodorus pakistanensis Siddiqi, 1963**
Diploscapter spp.
Diptherophora spp.
Discolaimoides spp.
Ditylenchus myceliophagus Goodey, 1958**
Dorylaimoides spp.
Ecumenicus spp.
Eudorylaimus spp.
Filenchus microdorus Chawla, Prasad, Khan & Nand, 1969*
Geomonhystera spp.
Helicotylenchus abuharazi Zeidan & Geraert, 1990
Helicotylenchus urobelus Anderson, 1978*
Helicotylenchus willmottae Siddiqi, 1972**
Hemicriconemoides brachyurus (Loos, 1949) Chitwood & Birchfield, 1957*
Heterodera schachtii Schmidt, 1871**
Heterodera zaeae Koshy, Swarup & Sethi, 1971**
Irantylenchus clavidorus Kheiri, 1972*
Ironus spp.
Laimydorus spp.
Merlinius nagerensis Sagir & Erum, 2017**
Mesorhabditis spp.
Mononchus spp.
Mylonchulus spp.
Paradorylaimus spp.
Pratylenchus allenii Ferris, 1961
Pratylenchus flakkensis Seinhorst, 1968**
Prismatolaimus spp.
Psilenchus hilarulus de Man, 1921**

Psilenchus vinciguerrae Brzeski, 1991
Quinisulcius capitatus (Allen, 1955) Siddiqi, 1971**
Rhabdolaimus spp.
Seinura oostenbrinki Hussain & Khan, 1967**
Tripyla spp.
Tylencholaimus spp.
Xiphinema brevicolle Lordello & Da Costa, 1961**
Xiphinema index Thorne & Allen, 1950**
Zeldia spp.

New record species*

New host records species**

Occurrence percentage of nematode taxa:

Among the nematodes species, the maximum nematodes species found were belonging to the order Tylenchida followed by Dorylaimida, Aphelenchida, Enoplida, Rhabditida, Mononchida, Chromadorida, Monhysterida, Areolaimida and Triplonchida, respectively. The frequency of the species of order Tylenchida was highest (32%) followed by order Dorylaimida (24%), Aphelenchida (14%) and the least frequency was of the orders Monhysterida, Chromadorida, Areolaimida and Triplonchida (2%) (Fig. 2).

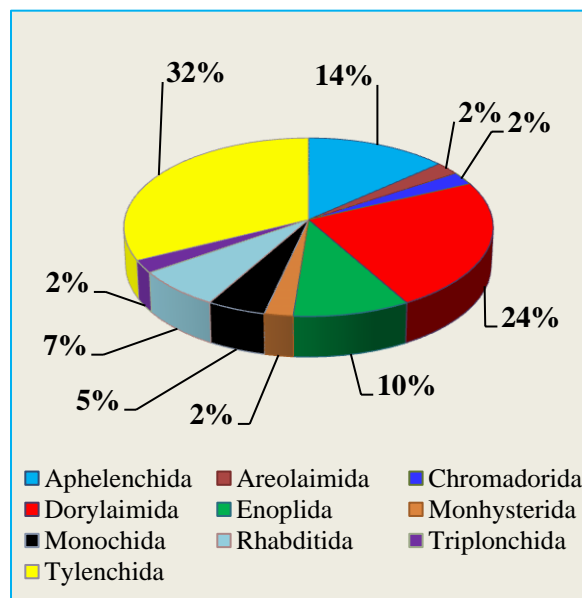


Fig. 2. Frequency (%) of nematode orders regarding to nematodes taxa from Kurram Agency.

Taxonomic studies

Aphelenchoides richardsoni Grewal, Siddiqi & Atkey, 1992

(Fig. 3 A-J; Table 2)

Description

Female: Body cylindrical and ventrally arcuate when relaxed by gentle heat. Cuticle finely annulated, lateral field with four incisures, head offset from body. Stylet about 10µm long, conus slightly shorter than shaft, basal knobs of stylet well developed. Median oesophageal bulb prominent, oval in shape with prominent valve plate situated slightly posterior from the center of median bulb. Nerve ring present just behind the median oesophageal bulb. Oesophageal glands extend dorsally over intestine. Excretory pore situated 60-63µm from anterior end at the level of nerve ring. Vulva with slightly raised lips. ovary outstretched, spermatheca elongated oval in shape, oocytes in a single row, post vulval uterine sac short (23-32µm long), vulva to anus distance 85-112µm, rectum 13-15µm long. Tail straight or slightly curved 22-32µm long bearing a single mucro towards ventral side.

Male: Male smaller than female, 385-499µm long. Body finely annulated except head region. Lateral field with four incisures. Head offset like female head, stylet 10µm long. Testes single outstretched, spicules 17-21µm long, thorn shape, arcuate sharply. Dorsal limb of spicules longer than ventral. Tail terminus pointed bearing single mucro. Three pairs of caudal papille present, first pair near to cloaca, second pair present in the mid of the tail and third pair near the tail terminus.

Remarks: *Aphelenchoides richardsoni* was first described from mushroom (*Agaricus bisporus*) from England by Grewal *et al.*, 1992. During the present survey specimens of *Aphelenchoides richardsoni* were collected from soil around

pumpkin (*Cucurbita moschata*) from Alizai Village of Kurram Agency, Pakistan. This species is reported for the first time in Pakistan, whereas male of *Aphelenchoides richardsoni* is not described earlier. The measurements of the specimens are within the range of *Aphelenchoides richardsoni* (Grewal, Siddiqi & Atkey, 1992).

Aphelenchoides unisexus Jain & Singh, 1984

(Fig. 4 A-I; Table 3)

Description

Female: Body slender, slightly arcuate ventrally when relaxed by gentle heat. Cuticle finely annulated except in head region. Lateral field with two incisures, head offset. Stylet conus a little shorter than shaft, with slight thickening of basal knobs. First region of oesophagus is cylindrical followed by a prominent oval median bulb with prominent valve plates. Elongate oesophageal glands lying dorsally along the intestine extending about 4-5 times by the body width. Nerve ring placed immediately behind median bulb. Excretory pore situated 73-83µm from anterior end, at the level of nerve ring or slightly behind. Vulval lips slightly protrude, ovary outstretched, not extending to oesophageal glands, spermatheca tubular, oocytes in 2-3 rows. Postvulval uterine sac well developed 50-64µm long. Vulva to anus distance 137-160µm, rectum distinct 14µm long. Tail 35-38µm long, arcuate, conoid bearing a star shaped mucro with 3-4 processes.

Male: Male smaller than female in size 420-507µm long. Body finely annulated except head region. Lateral field with two incisures. Head offset like female head, stylet 10µm long. Testes single outstretched, spicules 19-21µm long, thorn shape, sharply arcuate. Dorsal limb of spicules longer than ventral limb. Tail terminus bearing star shaped mucro. Three pairs of caudal papillae present, first pair near to cloaca, second pair

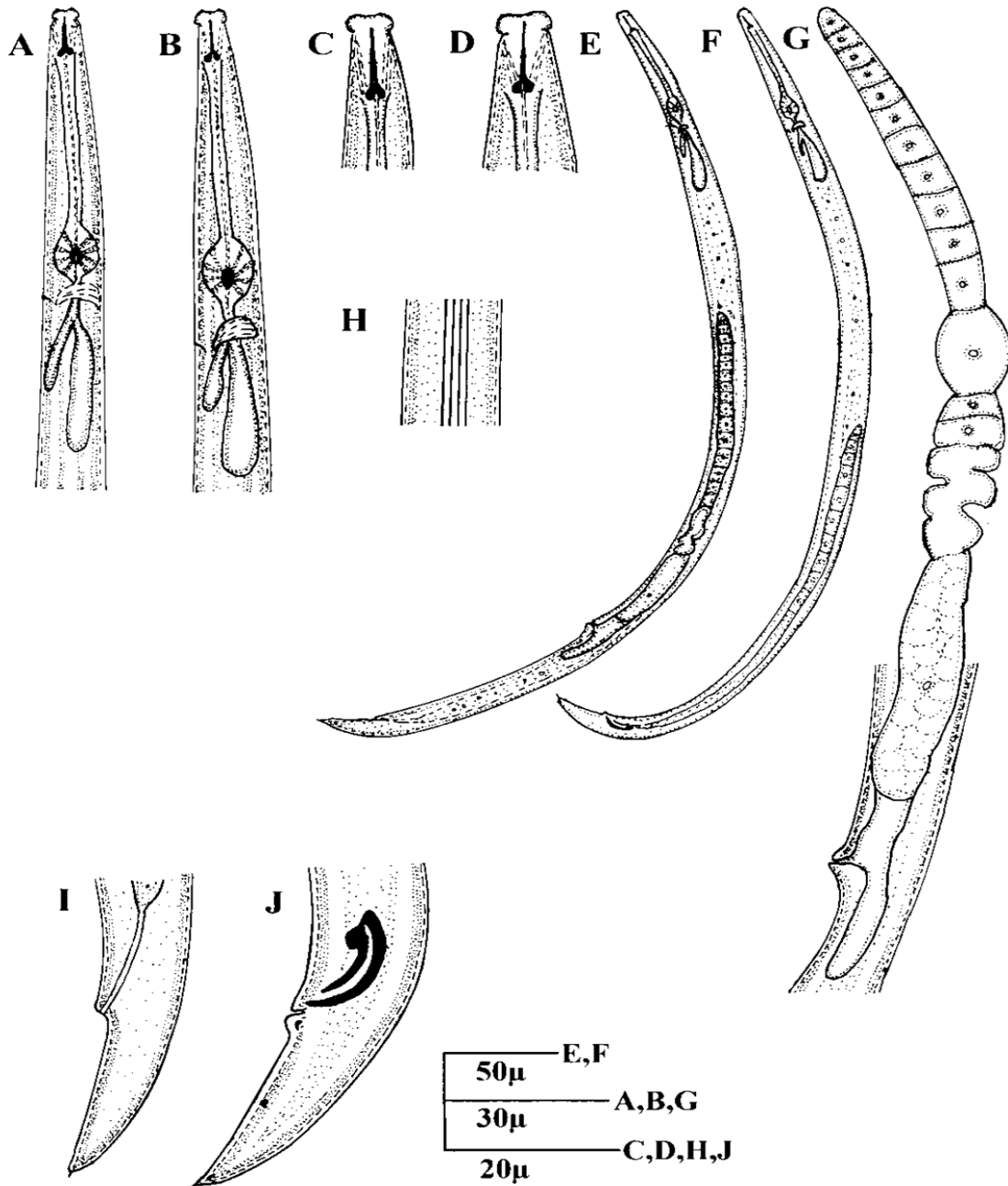


Fig. 3 (A-J). *Aphelenchoides richardsoni* Grewal, Siddiqi & Atkey (1992). Female: A. Oesophageal region; C. Anterior region; E. Entire body; G. Female gonad; H. Lateral fields; I. Tail region. Male: B. Oesophageal region; D. Anterior region; F. Entire body; J. Tail region.

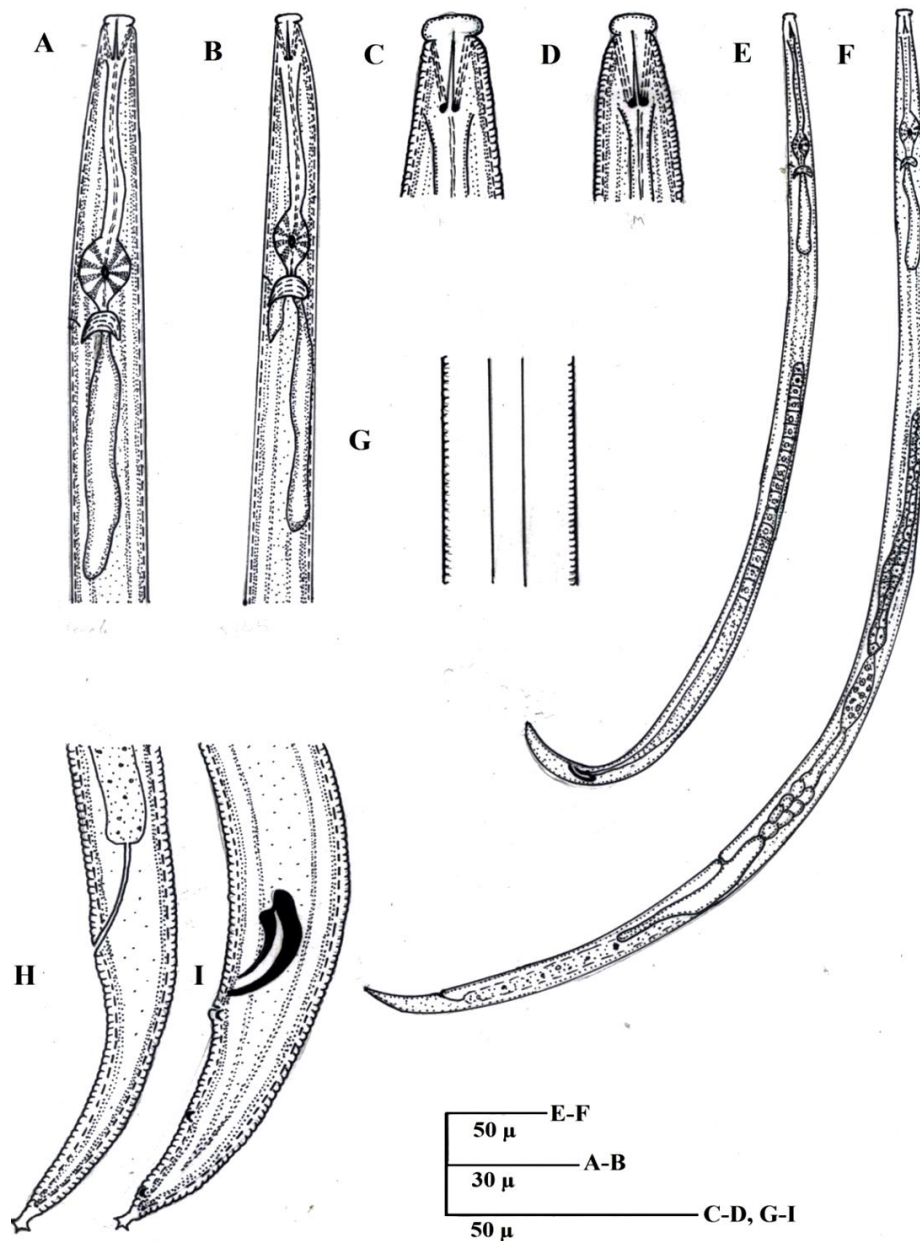


Fig. 4 (A-I). *Aphelenchoides unisexus* Jain & Singh (1984). Female: A. Oesophageal region; C. Anterior region; F. Entire body; G. Lateral field; H. Tail region. Male: B. Oesophageal region; D. Anterior region; E. Entire body; I. Tail region.

Table 2. Measurements of *Aphelenchoides richardsoni* (measurements are in μm except L).

Characters	Female(n=6)	Male (n=8)
	Mean \pm SD (Range)	Mean \pm SD (Range)
L	0.455 \pm 0.32 (0.39-0.49)	0.44 \pm 0.36 (0.38-0.49)
a	28.8 \pm 2.7 (25.3-32)	32.1 \pm 1.5 (30-33.6)
b	7.0 \pm 0.61 (6.5-8.1)	6.7 \pm 0.3 (6.4-7.1)
b'	4.52 \pm 0.23 (4.3-4.8)	4.12 \pm 0.50 (3.4-5.0)
c	18 \pm 1.9 (13.8-19.6)	17.9 \pm 1.5 (16.8-19.9)
c'	2.8 \pm 0.37 (2.4-3.5)	2.2 \pm 0.205 (2.1-2.6)
V%	72.5 \pm 1.17 (70.9-74.2)	-
Stylet	10 \pm 0.5 (10-11)	10.2 \pm 0.01 (10-10.5)
Excretory pore	61.8 \pm 1.3 (60-63)	65.2 \pm 4.6 (56-72)
Junction of DGO	65.6 \pm 5.0 (60-74)	65.5 \pm 3.2 (60-68)
DGO	100.2 \pm 5.6 (90-106)	107.7 \pm 11.4 (96-120)
Tail length	25.5 \pm 3.0 (22-32)	24.8 \pm 1.96 (20-26)
Anal body width	9 \pm 0.81 (8-10)	10.8 \pm 1.36 (8-12)
Body width	16.1 \pm 1.21 (12-18)	13.8 \pm 1.16 (12-16)
Vulva-anus distance	99.1 \pm 8.3 (85-112)	-
Post uterine sac	23.8 \pm 6.4 (11-32)	-
Spicules	-	19.3 \pm 1.21 (17-21)

Table 3. Measurements of *Aphelenchoides unisexus* (measurements are in μm except L).

Characters	Female (n=4)	Male (n=7)
	Mean \pm SD (Range)	Mean \pm SD (Range)
L	0.65 \pm 0.39 (0.60-0.71)	0.46 \pm 0.28 (0.42-0.50)
a	33.9 \pm 2.4 (30-36.1)	30.4 \pm 1.9 (28.4-33.8)
b	8.1 \pm 0.51 (7.5-8.9)	6.2 \pm 0.44 (5.6-6.8)
b'	4.9 \pm 0.77 (4.4-6.3)	4.1 \pm 0.23 (3.8-4.4)
c	17.5 \pm 0.78 (16.5-18.7)	15.9 \pm 1.6 (13.7-18.7)
c'	3.3 \pm 0.25 (3.1-3.7)	2.3 \pm 0.24 (2.2-2.8)
V %	70.7 \pm 0.68 (69.7-71.6)	-
Stylet	11	10
Excretory pore	77.7 \pm 3.9 (73-83)	63.6 \pm 1.71 (60-65)
Junction of DGO	79.5 \pm 0.5 (79-80)	74.2 \pm 2.1 (71-78)
DGO	130.2 \pm 9.98 (113-137)	111.6 \pm 7.9 (99-126.4)
Tail length	37 \pm 1.2 (35-38)	29.4 \pm 1.76 (27-32)
Anal body width	11 \pm 1 (10-12)	12.2 \pm 0.67 (11-13)
Body width	19.2 \pm 1.78 (17-21)	15.4 \pm 0.85 (14-16.8)
Vulva to anus distance	152.7 \pm 9.4 (137-160)	-
Post uterine sac	57.5 \pm 5.4 (50-61)	-
Spicules	-	19.7 \pm 0.69 (19-21)

present in the mid of the tail and third pair near the tail terminus.

Remarks: *Aphelenchoides unisexus* was first described from soil around the roots of chrysanthemum (*Chrysanthemum* sp.) by Jain & Singh, 1984. Recently the specimens of *Aphelenchoides unisexus* were collected from soil around tomato (*Lycopersicon esculentum*) from Shingak Village of Kurram Agency, Pakistan. The species *Aphelenchoides unisexus* is reported as a new record from Pakistan. Moreover, the male of *Aphelenchoides unisexus* is not described earlier. The measurements of the specimens are within the range of the original description (Jain & Singh, 1984), except slightly lower value of b' (3.5-4.5 vs. 4.4-6.3) and more anteriorly located vulva (V% =62.3-67.2 vs.69.7-71.6).

***Filenchus microdorus* Chawla, Prasad, Khan & Nand, 1969**

(Fig. 5 A-E; Table 4)

Description

Female: Body slender, gradually tapering towards both ends, ventrally arcuate when killed by gentle heat. Finely striated body cuticle. Lateral field with four incisures. Head rounded and continuous with body contour. Stylet 6-7 μ m long with small basal knobs, conus shorter than shaft. Excretory pore located near the anterior part of the basal bulb, located 68-74 μ m from anterior end. Opening of dorsal oesophageal gland located 2.5 μ m posterior to basal knobs of stylet. Procorpus shorter than isthmus, cylindrical, median bulb oval, located anterior to center of oesophagus, isthmus narrow, nerve ring located in the middle of isthmus, basal oesophageal bulb pyriform, oesophagus 78-86 μ m long. Cardia prominent. Vulva located at 71.8-74.2% of total body length. Vulval lips not raised. Ovary single, outstretched anteriorly, oocytes arrange in a single row. Post uterine sac small, 4-5.6 μ m long.

Distance from vulva to anus 42-52 μ m. Rectum and anus prominent. Tail 85-94 μ m long, gradually tapering to an acute terminus.

Male: Not found.

Remarks: *Filenchus microdorus* was first described from soil around the roots of rye (*Brassica juneca*) from Khanpur, U.P. India. Recently the specimens of *Filenchus microdorus* were collected from soil around the roots of okra (*Abelmoschus esculentus*) from Borki, Kurram Agency, Pakistan. The measurements of the specimens are within the range of the *Filenchus microdorus* (Chawla, Prasad, Khan & Nand, 1969).

***Helicotylenchus urobelus* Anderson, 1978**

(Fig. 6 A-F; Table 5)

Description

Female: Body spiral when killed the specimens on gentle heat. Lip region hemispherical bearing four fine transverse annules. Cephalic frame work prominent. Lateral field with four incisures. Stylet 23-27 μ m long, knobs anterior surface concave. Orifice of dorsal oesophageal gland 7-10 μ m from stylet base. Excretory pore 85-110 μ m posterior to head apex.

Oesophagus 126-145 μ m long and oesophagus junction 105-120 μ m long from anterior end of head. Vulva 61.18-64.03% of total body length. Ovaries two outstretched, oocytes in a single row except for a short region of multiplication, spermatheca present. Phasmids located 3-7 annules anterior from anus level. Tail with a short to pronounced set-off ventral projection, tapering to a smooth rounded ventral projection and in some specimens prominent pointed ventral projection present.

Male: Not found.

Remarks: *Helicotylenchus urobelus* was described by Anderson in 1978. During the

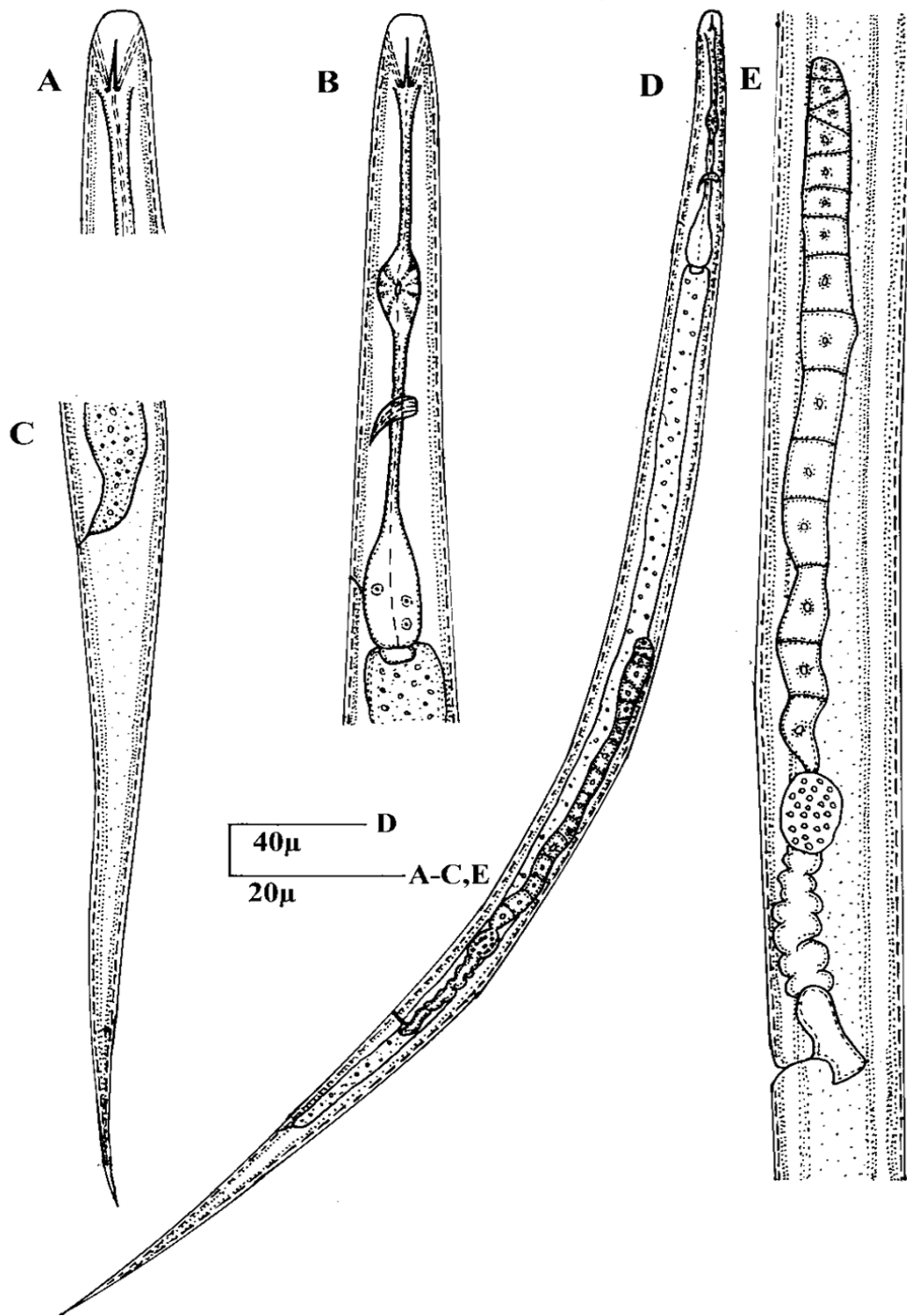


Fig. 5 (A-E). *Filenchus microdorus* Chawla, Prasad, Khan & Nand (1969). Female: A. Anterior region; B. Oesophageal region; C. Tail region; D. Entire body; E. Female gonad.

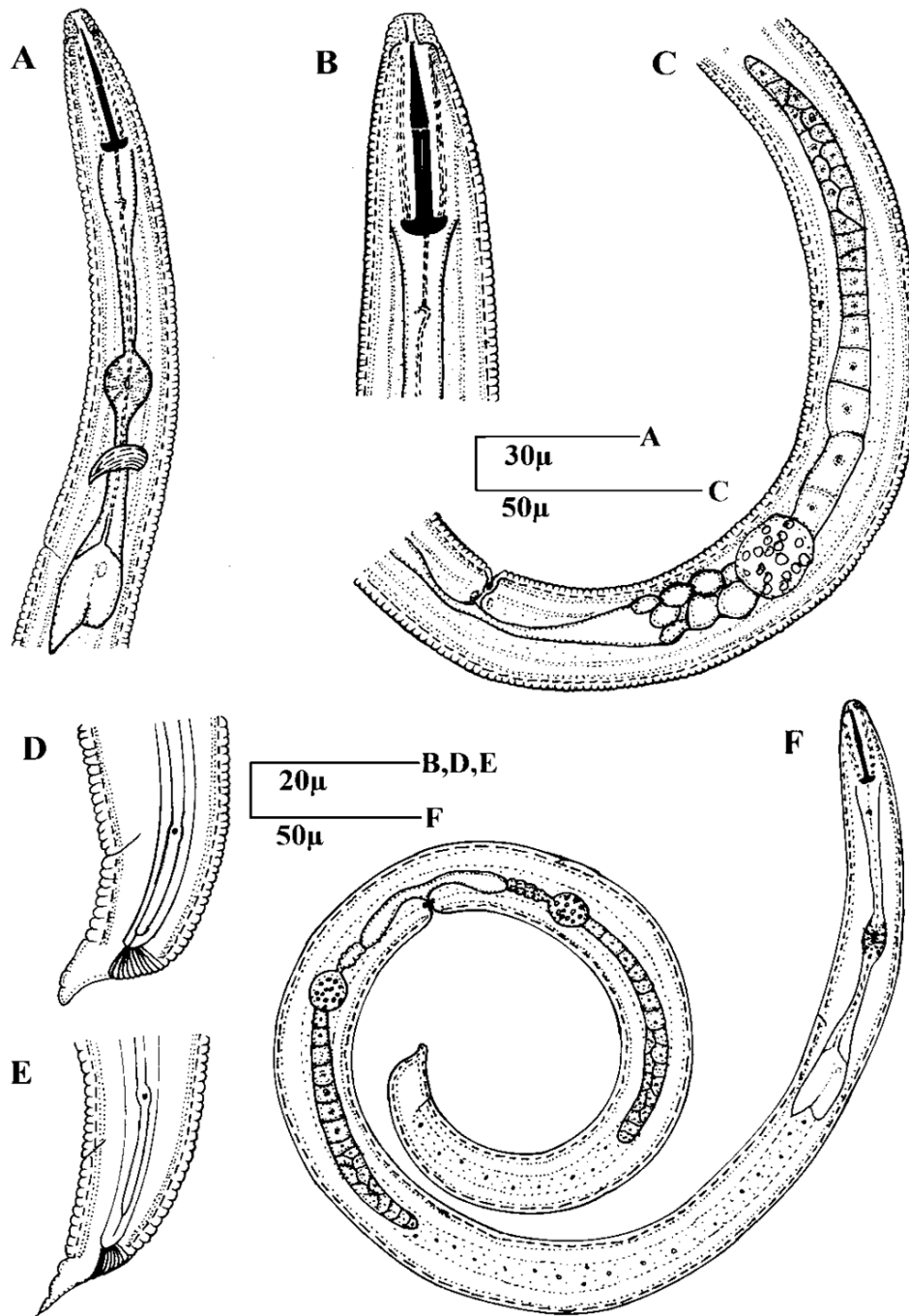


Fig. 6 (A-F). *Helicotylenchus urobelus* Anderson (1978). Female: A. Oesophageal region; B. Anterior region; C. Female gonad; D, E. Tail region; F. Entire body.

Table 4. Measurements of *Filenchus microdorus* (measurements are in μm except L).

Characters	Female (n=7) Mean \pm SD	Range
L	0.49 \pm 0.02	0.46-0.52
a	30.0 \pm 2.08	27.5-32.6
b	5.8 \pm 0.18	5.7-6.2
c	5.5 \pm 0.14	5.3-5.7
c'	7.8 \pm 0.53	7-8.5
V%	72.4 \pm 0.7	71.8-74.2
Stylet	6.8 \pm 0.34	6-7
Excretory pore	71.5 \pm 1.7	68-74
Oesophageal length	83.8 \pm 2.5	78-86
Tail length	88.7 \pm 3.2	85-94
Anal body width	11.2 \pm 0.69	10-12
Maximum body width	16.5 \pm 1.17	15-19
Vulva to anus distance	48.5 \pm 4.06	42-52
Post uterine sac	4.5 \pm 0.62	4-5.6

Table 5. Measurements of *Helicotylenchus urobelus* (measurements are in μm except L).

Characters	Female (n=8) Mean \pm SD	Range
L	0.65 \pm 0.02	0.59-0.68
a	27.2 \pm 1.7	24.70-29.56
b	5.7 \pm 0.14	5.61-6
b'	4.8 \pm 0.20	4.53-5.13
c	34.1 \pm 2.5	31.3-38.64
c'	1.2 \pm 0.09	1.13-1.42
V %	62.0 \pm 0.91	61.0-64.0
Stylet	25.5 \pm 1.3	23-27
Excretory pore	105.2 \pm 5.3	94-110
Junction of dorsal oesophageal gland	113.6 \pm 5.45	105-120
Dorsal oesophageal gland	135.2 \pm 5.9	126-145
Tail length	19.2 \pm 1.19	17-21
Anal body width	14.6 \pm 0.85	13-16
Body width	24.1 \pm 1.69	21-27

present studies the specimens of *H. urobelus* were collected from soil around the roots of okra (*Abelmoschus esculentus*), kidney bean (*Phaseolus vulgaris*), mint (*Mentha*), cucumber (*Cucumis sativus*), peach (*Prunus persica*), wheat (*Triticum aestivum*), chinara tree (*Platanus orientalis*) from Borki, chili (*Capsicum frutescens*) from Alamsher, maize (*Zea mays*), mung bean (*Vigna radiata*), kidney bean (*Phaseolus vulgaris*) from Zeran, pumpkin (*Cucurbita moschata*) from Alizai as a new record from Pakistan. The measurements of the specimens are under the range of *H. urobelus* Anderson, 1978 except their body posture lose spiral vs spiral.

***Hemicriconemoides brachyurus* (Loos, 1949)
Chitwood & Birchfield, 1957**

(Fig. 7 A-D; Table 6)

Description

Female: Body elongated, cylindrical when killed on gentle heat, enclosed in a double cuticle sheath attached at anterior end and separate on tail. Two rounded lip annules present, posterior annules larger than anterior. Total body annules (R) 102-112. Head slightly offset. Stylet 56-59.2µm long, stylet knobs anteriorly cup shaped. 18-19 annules present from anterior end to excretory pore (Rex). Vulva 92.7-94% of body length. Vulval sheath present in cuticle. Anus 2-3 annules posterior to vulva (Rvan). Body diameter narrow immediately posterior to vulva. Anus to tail terminus (Ran) have 6 annules. Tail terminus rounded or hemispherical.

Male: Not found.

Remarks: Specimens of *Hemicriconemoides brachyurus* were collected from soil around the roots of walnut (*Juglans* spp.) from Borki Village of Kurram Agency, as a new record of Pakistan. The measurements of the specimens are within the range of the *H. brachyurus* (Loos, 1949) Chitwood & Birchfield, 1957.

***Irantylenchus clavidorus* Kheiri, 1972**

(Fig. 8 A-H; Table 7)

Description

Female: Body straight to slightly arcuate upon relaxation. Cuticle finely annulated, lateral field with four incisures, cephalic region smooth, elevated, rounded slightly truncated anteriorly, continuous with body contour, marked with fine transverse striae. Amphids with longitudinal slit-like apertures. Stylet well developed 11-12µm long, conus small about one-third of total stylet length, the posterior portion becomes gradually wider. Usually three basal knobs absent, ends in a clavate base swelling more developed on dorsal side, the lumen of oesophagus joint to the lumen of the stylet, clavate base on the ventral side. Dorsal oesophageal gland orifice opening present about one half of the stylet length behind the stylet base. Procorpus shorter than isthmus, Median bulb muscular, ovate to round, isthmus cylindrical, encircled by nerve ring at its middle. Basal bulb pyriform having three gland nuclei. Excretory pore usually near the base of isthmus 79-126µm from anterior end. Hemizonid just anterior to excretory pore.

Vulva a transverse slit 58.6-66% of total body length. Vagina at right angle to body axis, postvulval uterine sac about one-half of vulva body width long. Ovary single, outstretched anteriorly, oocytes arrange in a single row, spermatheca well developed. Rectum and anus distinct. Rectum shorter than anal body width. Tail elongated filiform with fine terminus.

Male: Male are similar to female except that the body is much slenderer than female. Male with single testis, outstretched, bursa adanal, gubernaculum 4-6µm, simple trough-shaped, Spicules well developed 18-28µm, pointed, ventrally curved. Anterior cloacal lip pointed, posterior one rounded.

Remarks: *Irantylenchus clavidorus* was described from soil around roots of alfalfa in Isfahan, Iran by Kheiri, 1972. Recently the

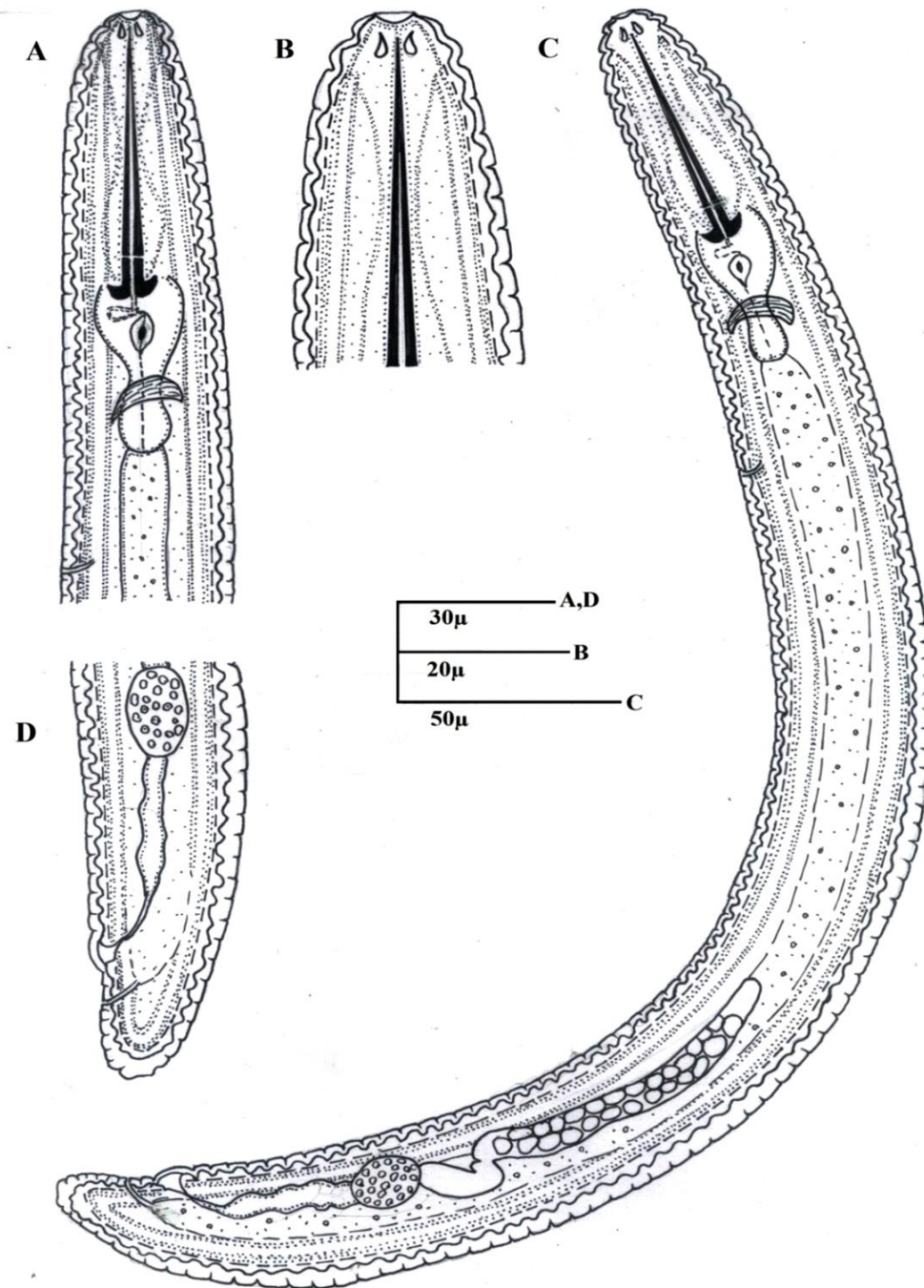


Fig. 7 (A-D). *Hemicriconemoides brachyurus* (Loos, 1949) Chitwood & Birchfield (1957). Female: A. Oesophageal region; B. Anterior region; C. Entire body; D. Tail region.

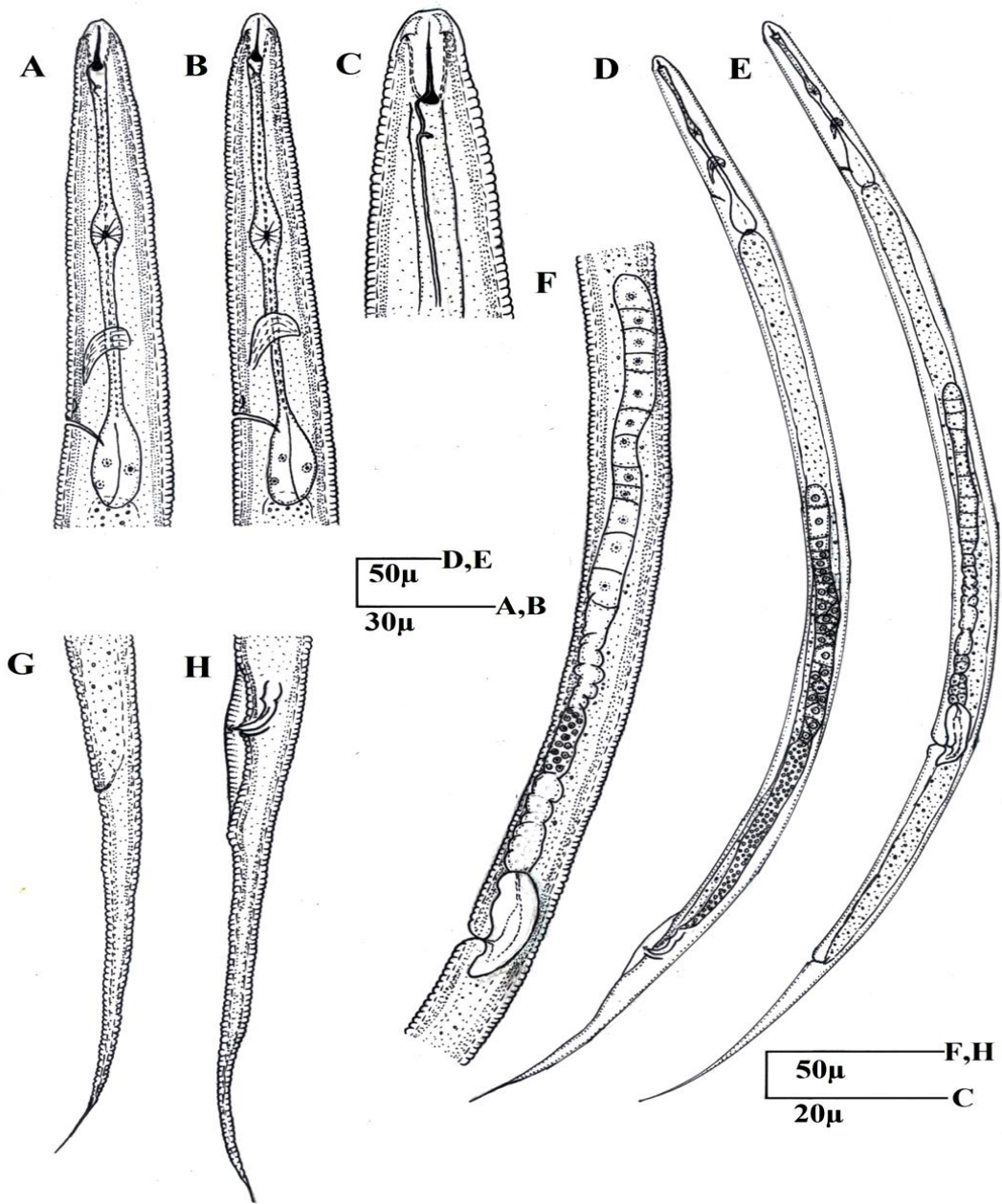


Fig. 8 (A-H). *Irantylenchus clavidorus* (Kheiri, 1972). Female: A. Oesophageal region C. Anterior region; E. Entire body; F. Female gonad, G. Tail region. Male: B. Oesophageal region; D. Entire body; H. Tail region.

Table 6. Measurements of *Hemicriconemoides brachyurus* (measurements are in μm except L).

Characters	Female (n=4)	Range
	Mean \pm SD	
L	0.41 \pm 0.03	0.35-0.46
a	12.27 \pm 0.76	11.5-13.3
b	4.5 \pm 0.12	4.3-4.6
c	23.8 \pm 2.06	20.8-26.6
c'	0.88 \pm 0.04	0.83-0.96
V%	93.35 \pm 0.46	92.7-94
Stylet	56.5 \pm 2.41	53-59.2
Excretory pore	112.5 \pm 5.5	103-116
Oesophageal length	90.5 \pm 6.1	83-100
Tail length	17.3 \pm 1.50	15-19.2
Anal body width	19.5 \pm 0.86	18-20
Max. body width	33.7 \pm 1.71	31-35
R	106.5 \pm 3.6	102-112
Rv	8.2 \pm 0.43	8-9
R van	2.2 \pm 0.43	2-3
Ran	6.0 \pm 0.00	6-6
Rex	28.7 \pm 0.43	28-29

Table 7. Measurements of *Irantylenchus clavidorus* (Kheiri, 1972) (measurements are in μm).

Characters	Female (n=30)	Male (n=13)
	Mean \pm SD (Range)	Mean \pm SD (Range)
L	0.80 \pm 0.07 (0.62-0.96)	0.76 \pm 0.05 (0.64-0.86)
a	33.6 \pm 3.64 (25.5-40)	37.9 \pm 6.28 (28.9-49.8)
b	6.2 \pm 0.43 (5.1-7.6)	5.9 \pm 0.42 (5-6.6)
c	5.6 \pm 0.5 (4-6.9)	4.9 \pm 0.32 (4.2-5.4)
c'	10.3 \pm 2.09 (5.8-14.5)	11.1 \pm 1.43 (8.5-13.3)
V%	62 \pm 1.37 (58.6- 66)	-
Stylet	11.9 \pm 0.26 (11-12)	11.8 \pm 0.36 (11-12)
Excretory pore	102.2 \pm 9.6 (79 -126)	98.7 \pm 4.22 (93-107)
Oesophageal length	129.5 \pm 10.1 (105-150)	127.7 \pm 5.4 (119-139)
Tail length	141.8 \pm 11.6 (112-165)	154.7 \pm 13.7 (136-174)
Anal body width	14.07 \pm 2.5 (10 -19)	14 \pm 1.46 (12-17)
Body width	24.3 \pm 3.9 (17-32)	20.5 \pm 2.95 (16-24)
Spicules	-	21.4 \pm 2.43 (18 - 28)
Gubernaculum	-	5.4 \pm 0.63 (4-6)

specimens of *Irantylenchus clavidorus* (Kheiri, 1972) were collected from soil around tomato (*Lycopersion esculentum*), chilli (*Capsicum frutescens*), radish (*Raphanus sativus*), okra (*Abelmoschus esculentus*), bottle gourd (*Lagenaria siceraria*), eggplant (*Solanum melongena*), pumpkin (*Cucurbita moschata*), rice (*Oryza sativa*), maize (*Zea mays*), mung bean (*Vigna radiata*), kidney bean (*Phaseolus vulgaris*), fresh green bean, soya bean (*Glycine max*), walnut (*Juglans* spp.), peanut (*Arachis hypogaea*), mint (*Menthas achalinensis*) from Kurram Agency, Pakistan. The measurements of the specimens are within the range of the *Irantylenchus clavidorus* (Kheiri, 1972). This is the new record from Pakistan, identified during the present study.

New host records species: During the survey, seventeen new host records were reported viz., *Aphelenchoides besseyi* Christie, 1942 from Shingak, *Aphelenchoides goodeyi* Siddiqi & Franklin, 1967 from Kunj Alizai, *Aphelenchoides siddiqii* Fortuner, 1970 from Shingak, *Aphelenchus avenae* Bastian, 1865 from Shingak, *Basiria graminophila* Siddiqi, 1959 from Shakh, *Boleodorus pakistanensis* Siddiqi, 1963 from Shingak and Zeran, *Ditylenchus myceliophagus* Goodey, 1958 from Laimai and Zeran, *Helicotylenchus willmottae* Siddiqi, 1972 from Borki, *Heterodera schachtii* Schmidt, 1871 from Borki, *Heterodera zae* Koshy, Swarup & Sethi, 1971 from Borki, *Merlinius nagerensis* Sagir & Erum, 2017 from Shingak and Borki, *Pratylenchus flakkensis* Seinhorst, 1968 from Borki, *Psilenchus hilarulus* de Man, 1921 from Kunj Alizai, Borki, Dangila, Lalmai, Karman, Shakh, Shingak and Zeran, *Quinisulcius capitatus* (Allen, 1955) Siddiqi, 1971 from Shingak, *Seinura oostenbrinki* Hussain & Khan, 1967 from Borki, *Xiphinema brevicolle* Lordello & De Costa, 1961 from Borki and *X. index* Thorne and Allen, 1950 from Borki (Table 8).

Cluster analysis and similarity indices: The dendrogram of cluster analysis among the localities of surveyed areas was shown in Fig.9. Nematode population in localities was differentiated by two main clades. In clade I Agra, Alamsher and Dangila form a subclade with Lalmai and Shublan. In clade II Karman and Kunj Alizai exist in subclade with Borki and Shakh. Shingak and Zeran have similar nematode population and found in clade II. Cluster analysis of forty seven nematode species population shown in two main clades, with two subclades each (Fig. 10).

In clade I *Aphelenchoides* unisexus, *Aphelenchoides siddiqii*, *Amphidorylaimus* spp. and *Diploscapter* spp. formed a subclade I with *Geomonhystera* spp., *Eudorylaimus* spp., *Dorylaimoides* spp., *Achromadora* spp. and *Discolaimoides* spp., *Zeldia* spp., *Xiphinema brevicolle*, *Psilenchus vinciguerrae*, *Laimydorus* spp., *Hemicriconemoides brachyurus*, *Helicotylenchus urobelus*, *Diptherophora* spp., *Aphelenchoides besseyi* and *Ironus* were in subclade II along with *Paradorylaimus* spp., *Mononchus* spp., *Aphelenchoides goodeyi*, *Alaimus* spp. and *Aphelenchoides richardsoni*. Clade II has two subclades, *Tripyla* spp. *Pratylenchus flakkensis*, *Filenchus microdorus*, *Ecumenicus* spp. and *Basiria graminophila* formed a subclade I along with *Xiphinema index*, *Seinura oostenbrinki*, *Rhabdolaimus* spp., *Prismatolaimus* spp., *Mylonchulus* spp., *Mesorhabditis* spp., *Irantylenchus clavidorus*, *Heterodera zae*, *Helicotylenchus willmottae* and *Helicotylenchus abuharazi*.

Tylencholaimus spp. formed subclade II with *Quinisulcius capitatus*, *Psilenchus hilarulus*, *Pratylenchus alleni*, *Merlinius nagerensis*, *Heterodera schachtii*, *Ditylenchus myceliophagus*, *Boleodorus pakistanensis* and *Aphelenchus avenae*. Jacords Coefficient of similarity among species is shown in Table 9 (Past version 2.17).

Table 8. New host record nematodes species according to host and localities encountered during present studies.

Nematodes	Hosts	Locations
<i>Aphelenchoides besseyi</i> Christie, 1942	<i>Abelmoschus esculentus</i>	Shingak
<i>A. goodeyi</i> Siddiqi and Franklin, 1967	<i>Solanum melongena</i>	Kunj Alizai
<i>A. siddiqii</i> Fortuner, 1970	<i>Lycopersicon esculentum</i>	Shingak
<i>Aphelenchus avenae</i> Bastian, 1865	<i>Lycopersicon esculentum</i>	Shingak
<i>Basiria graminophila</i> Siddiqi, 1959	<i>Arachis hypogaea</i> <i>Capsicum frutescens</i> <i>Solanum melongena</i>	Shakh
<i>Boleodorus pakistanensis</i> Siddiqi, 1963	<i>Lycopersicon esculentum</i>	Shingak, Zeran
<i>Ditylenchus myceliophagus</i> Goodey, 1958	<i>Prunus armeniaca</i>	Laimai, Zeran
	<i>Solanum melongena</i>	
	<i>Vigna radiata</i>	
	<i>Zea mays</i>	
	<i>Zea mays</i>	
<i>Helicotylenchus willmottae</i> Siddiqi, 1972	<i>Phaseolus vulgaris</i>	Borki
	<i>Juglans</i> spp.	
	<i>Juglans</i> spp.	
<i>Heterodera schachtii</i> Schmidt, 1871	<i>Prunus armeniaca</i>	Borki
	<i>Prunus persica</i>	Borki
<i>H. zaeae</i> Koshy, Swarup and Sethi, 1971	<i>Pinus gerardiana</i>	Borki
<i>Merlinius nagerensis</i> Sagir and Erum, 2017	<i>Arachis hypogaea</i>	Shingak,
	<i>Prunus persica</i>	Borki
	<i>Prunus avium</i>	
<i>Pratylenchus flakkensis</i> Seinhorst, 1968	<i>Juglans</i> spp.	Borki
	<i>Abelmoschus esculentus</i>	Alamsher
<i>Psilenchus hilarulus</i> de Man, 1921	<i>Arachis hypogaea</i>	Kunj Alizai
	<i>Capsicum frutescens</i>	Borki,
	<i>Juglans</i> spp.	Dangila
	<i>Lycopersicon esculentum</i>	Lalmai
	<i>Phaseolus vulgaris</i>	Karman
	<i>Solanum melongena</i>	Shakh
	<i>Vigna radiata</i>	Shingak
		Zeran
		Shingak
<i>Quinisulcius capitatus</i> (Allen, 1955) Siddiqi, 1971	<i>Capsicum frutescens</i>	Shingak
<i>Seinura oostenbrinki</i> Hussain and Khan, 1967	<i>Lagenaria siceraria</i>	Borki
<i>Xiphinema brevicolle</i> Lordello and Da Costa, 1961	<i>Prunus persica</i>	Borki
	<i>Platanus orientalis</i>	
<i>X. index</i> Thorne and Allen, 1950	<i>Prunus persica</i>	Borki
	<i>Salix tetrasperma</i> Roxb	

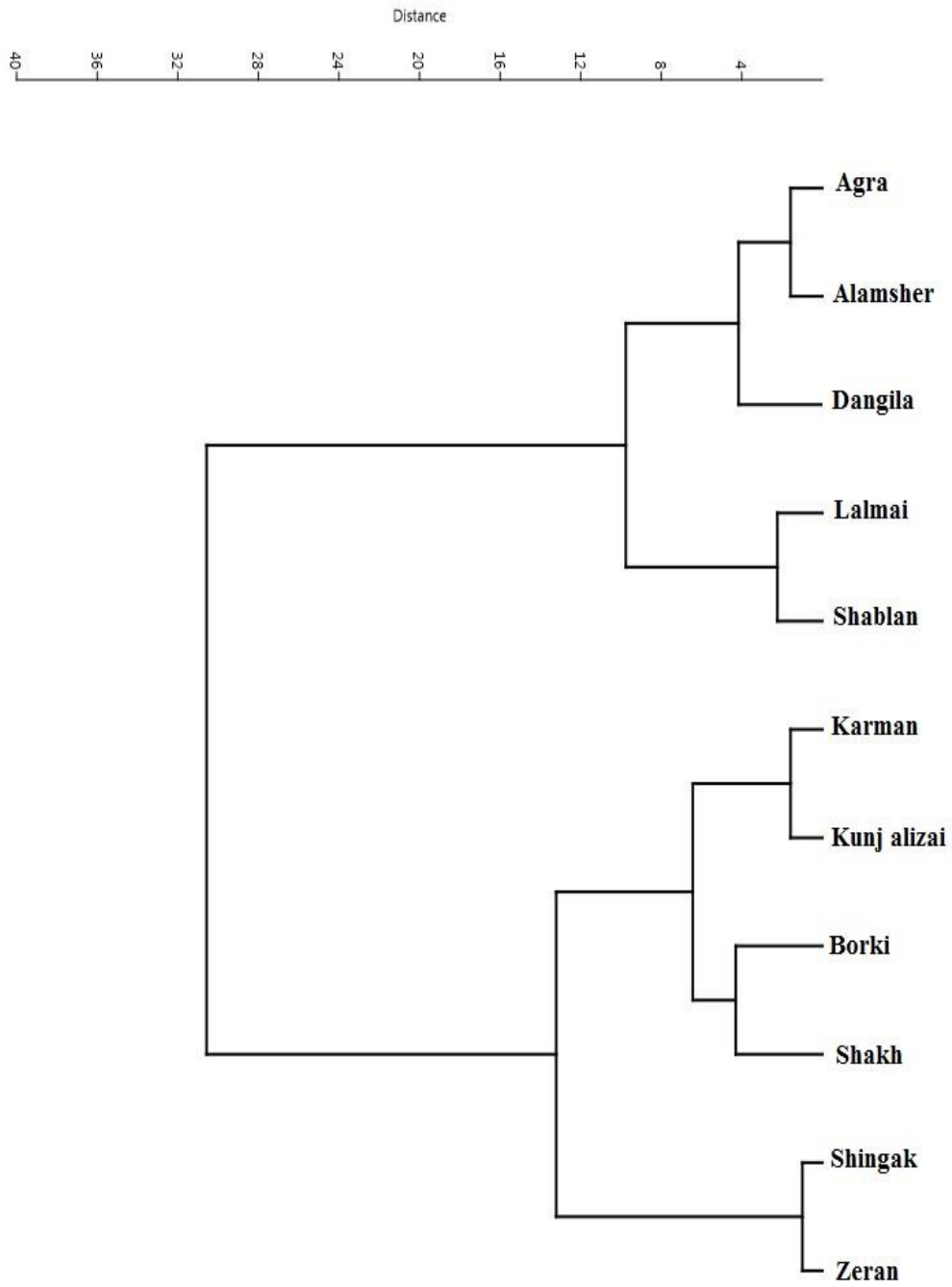


Fig. 9. Dendrogram resulting from the average linkage of 11 localities of Kurram Agency based on nematode population.

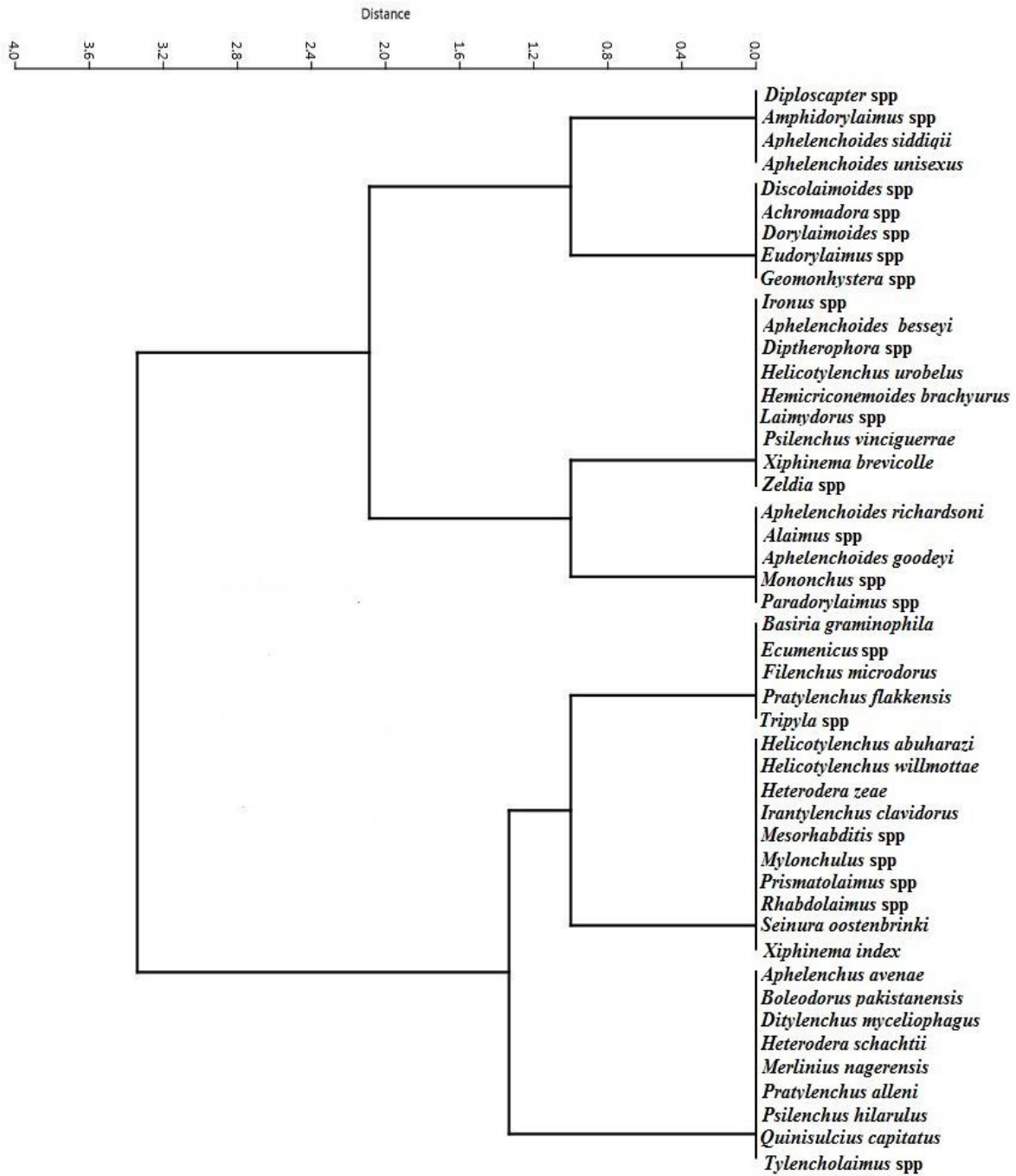


Fig. 10. Euclidean distance of nematode population of Kurram Agency, Pakistan.

Table 9. Similarity index of localities of Kurram Agency, Pakistan.

Localities	Agra	Alamsher	Borki	Dangila	Karman	Kunj alizai	Lalmai	Shablan	Shakh	Shingak	Zeran
Agra	0	2.23607	21.095	5.83095	15.5242	17.72	7.81025	7.28011	21.5407	13.4536	13.4536
Alamsher	2.236068	0	19.0263	3.60555	13.3417	15.5242	8.60233	7.2111	19.3132	11.3137	11.4018
Borki	21.095023	19.0263	0	16.0312	6.32456	5	26.3059	23.5372	6.08276	13.0384	14.4222
Dangila	5.8309519	3.60555	16.0312	0	10.0499	12.1655	10.4403	8.06226	15.8114	7.81025	8.06226
Karman	15.524175	13.3417	6.32456	10.0499	0	2.23607	20.0998	17.2627	6.40312	7.07107	8.48528
Kunj alizai	17.720045	15.5242	5	12.1655	2.23607	0	22.0227	19.105	4.24264	8.06226	9.43398
Lalmai	7.8102497	8.60233	26.3059	10.4403	20.0998	22.0227	0	3.16228	25.0799	15.2971	14.5602
Shablan	7.2801099	7.2111	23.5372	8.06226	17.2627	19.105	3.16228	0	22.0227	12.1655	11.4018
Shakh	21.540659	19.3132	6.08276	15.8114	6.40312	4.24264	25.0799	22.0227	0	10.0499	11.1803
Shingak	13.453624	11.3137	13.0384	7.81025	7.07107	8.06226	15.2971	12.1655	10.0499	0	1.41421
Zeran	13.453624	11.4018	14.4222	8.06226	8.48528	9.43398	14.5602	11.4018	11.1803	1.41421	0

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