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Full Length Research Paper

Management of root-knot nematode *Meloidogyne incognita* (Kofoid and White) Chitwood in Tomato (*Lycopersicon esculentum* Mill. through poultry manure and rapeseed cake

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Study was conducted in Diredawa Haramaya University Research Station in naturally root-knot nematode infested soil to determine the effect of poultry manure and rapeseed cake against root-knot nematode infestation and yield of tomato var. Mraglobe. Five levels of poultry manure (0, 5, 10, 15 and 20 ton/ha) and three levels of rapeseed cake (0, 100 and 200 kg/ha) were applied in experimental plot three weeks before transplanting the seedlings in band placement manner to the depth of 20 cm. The experiment laid in RCBD factorial arrangement. The result revealed that there were significant interaction effect of poultry manure above 5-20 ton/ha with rapeseed cake at 200 kg/ha at P<0.05 on number of egg/egg mass, root galling, population of J2 and yield and growth of the crop. Infestation of root-knot nematode was significantly reduced as both poultry manure and rapeseed cake increased. Fruit yield increased when poultry manure raised 0 to 15 ton/ha with rapeseed cake 200 kg/ha. Applications of poultry manure at 5 ton/ha to 15 ton/ha in combination with rapeseed cake at 200 kg/ha gave highest yield and suppressed root knot nematode.

Key words: *Meloidogyne incognita,* poultry manure, rapeseed cake, root-knot nematode, tomato.

INTRODUCTION

Tomato (*Lycopersicon esculentum* Mill.) is one of the most important vegetable crops in Ethiopia (EARO, 2004). The national average yield under farmers' condition is 9 and 25 and 40 ton/ha at demonstration and experimental research plots, respectively (Abdissa et al., 2010). However, much higher tomato yield has been reported in other countries like 73.87 ton/ha in USA, 63.55 ton/ha in Spain, 88.91 to 146 ton/ha in the Netherlands (Hassan et al., 2010). The eastern part of Ethiopia is considered to be a high potential area for vegetable production and marketing (Bezabih and Hadera, 2007). Despite production and market potential of the area the occurrence of several factors contributed a lot in reducing yield/unit of production and quality.

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Several reports indicated that disease and insect pests are the major bottleneck in tomato growing areas (Esheteu et al., 2006). *Meloidogyne incognita* (Kofoid and White) Chitwood was identified as a pest causing root-knot in vegetables in the region by Tadele and Mengistu (2000), and Wondirad and Tesfamariyam (2002). Since then, little research has been done on management aspects of root-knot nematode. Wondirad et al. (2009) stated that most studies done in Ethiopia on root-knot nematode have yet focused mainly on generating baseline information rather than management.

Soil amendments with different types of organic manures are effective in reducing the population densities of many soil-borne plant pathogens including root-knot nematode (Hassan et al., 2010). Cruciferous residues as soil amendments are effective in reducing the population densities of many soil-borne plant pathogens (Ritu and Lodha, 2002). Studies in Nigeria, South East Asia and many other developing countries indicated that poultry manure, cruciferous residues and other organic west soil amendment increased the yield of tomato and suppressed root-knot nematode (Riegel and Noe, 2000; Orisajo et al., 2008; Pakeerathan et al., 2009; Ogulumba et al., 2010; Wachira et al., 2009). Rahman et al. (2002) also reported that application of poultry manure at 3 ton/ha with mustard cake at 300 kg/ha significantly reduced root knot-nematode population and increased yield of tomato. Therefore, current study was conducted to determine the effect of poultry manure, rapeseed cake and their combinations on root-knot nematode infestation, growth and yield of tomato.

MATERIALS AND METHODS

Experimental site description

The experiment was conducted from July to the end of December, 2010 at Haramaya University's Research Station located at 9°6′N latitude and 41°8′E longitude in Diredawa town at an altitude of 1197 m above sea level. The mean annual rainfall of the area is 520 mm, with mean minimum and maximum temperature of 14.5 and 34.6°C, respectively.

Experimental materials

A tomato seed variety named Marglobe was obtained from Melkasa Agricultural Research Center. The poultry manure was obtained from Haramaya University's poultry farm. Rapeseed cake was obtained from Hamaressa oil factory, Harar. The choice of soil amendments was made based on relative availability, accessibility, simplicity for application and familiarity with farmers' existing practices and their basic differences compared to other amendments options.

Raising seedlings

Nursery bed was prepared in a field at research farm, the width and length of the bed was 1 m x 2 m, respectively. Seed of the tomato variety Marglobe was manually drilled into the rows spaced 10 cm apart. Urea was applied at the rate of 100 g/5 m^2. The nursery beds were mulched and irrigated two times/day.

Preparation of experimental plots and seedling transplanting

There were 15 experimental plots which were prepared based on spacing (30 cm x 70 cm) for tomato and replicated three times. Each plot had 10.8 m^2. There were a total of 45 plots separated by 1 m and 1.5 m spacing between plots and between blocks respectively. There were four rows in each plot ten four week old tomato seedlings were transplanted in each rows finally giving 40 plants/plot.

Identification of *Meloidogyne* species

Species identification was performed based on perineal patterns for *M. incognita* as described by (Sasser and Carter, 1985; Jepson, 1987). Infested tomato roots were collected from the research farm. The roots were gently washed with clean water and soil and other debris were removed cleaned roots were allowed to dry. Galls from infested roots were manually dissected out using forceps and dissecting needles the adult females were separated after stained with acid fuchion the recovered females were collected and kept overnight in a Petri dish with lactopheno. Finally the perineal region of females were cut and was trimmed to square shape and placed to a drop of glycerine on a clean glass slide. The cover slip was gently placed on the glycerin drop and sealed with nail polish and the slide was labeled. Finally the perineal patterns of the sample were examined under compound microscope.

Extraction and counting of nematodes from soil and eggs from infested root

Extractions of second stage juvenile (J2) was carried out using a Baermann funnel method as described by Coyne et al. (2007). After tomato fruits were harvested ten 100 g soil samples from each plot were taken and thoroughly mixed and 100 g sub sample were prepared for J2 extraction. 100 g soil samples were placed in 250 ml beakers. Then the beakers were covered with a piece of muslin cloth and tightly tied with a rubber band and clamp then assembled upside down on the glass funnel filled with water and mounted on metal stand. After 24 h, the clamp was loosened slightly to allow 60 ml of suspension having nematodes to pass into a 100-ml beaker finally the suspensions were prepared for counted and expressed as J2/100 g. For extraction of egg mass form galled root ten egg mass/plant were dissected out from three plants/plot it was done for determination of average number of eggs/egg mass. The egg mass was transferred to a flask containing 60 ml of 5% sodium hypochlorite (common bleach) aqueous solution and vigorously shaken for 2 min. From the total suspension, 3 ml suspension was placed on counting dish and counted according to procedure described by Hussery and Barker (1973) and average number of egg/egg mass was calculated.

Field experiment

The experiment was carried out in completely randomized block design with three replications in factorial arrangements. Treatments included: Poultry manure in five level viz., 0, 5, 10, 15, and 20 ton/ha, rapeseed cake in three levels, viz., 0, 100, 200 kg/ha. Both soil amendments were applied in band placement manner by drilling to the depth of 20 cm in the experimental plot three weeks before transplanting the seedlings.

Data collection

Days to flowering for each treatment were taken when 75% of the plants flowered, and done by direct stand count. Other growth and yield data including days to maturity, plant height, fresh shoot weight, number of fruits/plant, average fruit weight and total yield/ha were taken from pre tagged ten plants from tow central rows.

Root galling assessment

Ten plants/plot were carefully uprooted washed with water to remove soil and other debris the roots then visually evaluated for root galling were scaled according to Taylor and Sasser (1978) (0= no galls, 2= 1–25% galling, 3=26–50% galling, 4=51–75% galling, 5=76–100% galling).

Assessment of root-knot nematode infestation

Five soils sample from each experimental plot were taken and roughly bulked to make 100 g soil for each plots. The soil samples taken for analysis and J2 were extracted through Baermann funnel method as described by Coyne et al. (2007). Finally, extracted J2 were counted and expressed as J2/100 g.

Data analysis

The data on root galling index, number of eggs/egg mass, final J2 population, and yield and growth parameters were subjected to Analysis of Variance (ANOVA) using GenStat version 7.2 computer packages (Gomez and Gomez, 1984). Treatment means were separated using Least Significant Difference (LSD) Test at 5% level of significance.

RESULTS AND DISCUSSION

Species identification

During species identification female perineal patterns form the experimental field corresponded with perineal patterns suggested by of Sasser and Carter (1985), Jepson (1987). Therefore based on the findings of this study and other previous reports species of root-knot nematode of the area was confirmed as *M. incognita* (Kofoid and White) Chitwood. Tadele and Mengistu (2000) recorded that the species of root-knot nematode prevalent in the area in general and in particular the research farm was *M. incognita.* Similar studies confirmed that the species of root-knot nematode prevailed in current study area as *M. incognita* (Metasebia et al., 2008).

Effects of poultry manure and rapeseed cake on root-knot nematode infestation

The main effect of poultry manure was highly significant (P<0.05) on root galling indices, final J2 and egg/eggmss. The main effect of rapeseed cake was also significant at (P<0.05) on the same the same parameters. Root galling index reduced when poultry manure applied at 5, 10, 15 and 20 ton/ha by 15, 30, 44.3 and 61.57%, respectively compared to control treatment. There was also significant interaction effect of both treatments on infestation of root-knot nematode. The highest number of eggs/egg mass was recorded from untreated control treatment (Table 1). There was reduction in the number of eggs/egg mass as the manure amendment was raised from 0 to 20 ton/ha by 14.8, 22.31, 54.88 and 86.14%, respectively, compared to unamended treatment. The lowest J2/100 g of soil was recorded at 20 ton/ha poultry manure and the second lowest number of J2/100 g was recorded at rapeseed cake at 200 kg/ha and poultry manure at 15 ton/ha (Table 1). Compared to untreated plot, combination of these amendments at these levels reduced the density of J2/100 g by 52.4 and 51.57% respectively. Thus high rate of poultry manure and lower rate of rapeseed cake have been required to bring down the population density of root knot nematode. The results of this study corresponds with Amarasinghe et al. (2007) who reported that the lowest nematode population and gall indices were recorded for rice amended with poultry manure. Similarly, Ogwulumba et al. (2010) reported that soil amended with organic materials (poultry droppings, grass ash and rice husk ash) at the range of 10 to 20 ton/ha significantly reduced the population of *Meleoidogye* spp in tomato in Nigeria. Riegel and Noe (2000) also indicated in cotton that with increase in rate of chicken litter amendment in soil infested with root-knot nematode, density of root knot nematode was significantly reduced. Similarly, Rahman et al. (2002) reported that plots incorporated with poultry manure at 3 ton/ha and mustard cake at 300 kg/ha resulted in significant reduction in population of root knot nematodes in tomato.

Effect of poultry manure and rapeseed cake on growth of tomato

Days to flowering and maturity

Poultry manure as main effect significantly (P<0.001) affected on days to 75% flowering and maturity (Table 2). The main effect rapeseed cake and the interaction effect of both amendments were non-significant on days to 75% flowering and fruit maturity (P<0.05). Days to 75% flowering and fruit maturity was significantly prolonged as poultry manure application rate increased beyond 15 ton/ ha. The shortest days to 75% flowering and days to fruit maturity was observed for plants treated with 15 ton/ha of poultry manure. Compared to the treatments amended with poultry manure at the rates of 5, 10, and 15 ton/ha, the treatment amended with poultry manure at the highest rate (20 ton/ha) significantly prolonged days to 75% flowering and fruit picking. The variation in flowering and maturity date revealed that application of poultry manure had an influence on the plant growth and

Table 1. The interaction effect of poultry manure and rapeseed cake against root-knot nematode infestation.

Treatments interaction		J2/100 g	Egg/egg mass	RGI
Poultry manure (ton/ha)	Rapeseed cake (kg/ha)			
	0	887.0^{a}	202.50^{a}	4.33^{a}
0	100	858.7^{ab}	190.67^{ab}	4.33^{a}
	200	848.7^{ab}	166.08^{ab}	4.33^{a}
	0	851.7^{ab}	165.32^{ab}	4.33^{a}
5	100	828.3^{abc}	158.67^{ab}	3.81^{ab}
	200	808.3^{bcd}	150.77^{bc}	3.67^{b}
	0	777.3^{cd}	148.31^{bc}	3.53^{b}
10	100	776.7^{cd}	154.34^{bc}	3.17^{c}
	200	743.7^{d}	152.33^{bc}	3.00^{c}
	0	475.3^{e}	122.70^{bcd}	3.23^{c}
15	100	454.3^{f}	129.02^{bcd}	2.61^{d}
	200	429.3^{f}	107.33^{cd}	2.33^{d}
	0	456.8^{f}	111.31^{cd}	2.68^{de}
20	100	455.3^{f}	99.39^{cd}	2.73^{de}
	200	356.7^{g}	86.09^{d}	2.11^{f}
CV		10.82	12.07	8.08
LSD (0.05) for (pm x rc)		63.34	34.95	0.87

RGI=root galling index, pm=poultry manure, rc=rapeseed cake. Means within a column followed by the same letter are not significantly different (P<0.05).

development. This finding is in agreement with that of Wachira et al. (2009) who reported that tomato flowering was earliest and more pronounced in plants grown in soil amended with optimum poultry manure. The finding of the authors is consistent with the one found in this experiment at the higher levels of poultry manure application except at the highest (20 ton/ha) level of the treatment. The delay in flowering at 20 ton/ha poultry manure application might be have been due to the excessive rate of poultry manure, which might have led to development of vegetative growth rather than reproductive growth. Christo et al. (2010) also reported that application of poultry manure at higher rate resulted in prolonged days to 50% flowering in mungbean.

Plant height

Both poultry manure and rapeseed cake had significant (P<0.05) interaction effects on plant height tomato (Table 2).

The results revealed that tallest plants were recorded at the combined application of poultry manure at 20 ton/ha and rapeseed cake at 200 kg/ha. At this level, the plant height increased by 130% compared to the heights of plants grown in untreated control plot. Ogulumba et al. (2009) reported that poultry manure amendments in the range of 10 to 20 ton/ha produced a significant effect on plant height compared to untreated treatments in tomato infested with root-knot nematodes.

Fresh shoot, root and dry root weight

The main effect of poultry manure was highly significant (P<0.001) on fresh shoot, root and dry root weight yield. Rapeseed cake as main effect and the interaction effect of the two amendments were non-significant on this parameter of the plant (Table 2). Increasing poultry manure amendments from 5, 10, 15, and 20 ton/ha increased the fresh shoot weight by 23.3, 33.0, 58.2, and 62.9%, respectively, compared to tomato grown in the untreated control plot.

The fresh root weight obtained from the control treatment was relatively higher than other treatments amended with poultry manure. The presence of female root knot nematodes inside the root system evidently increased the volume of the root system compared to that of normal uninfected plants. This may have contributed to the increase in the fresh root weight of control treatment.

Table 2. The interaction effect of poultry manure and rapeseed cake on growth of tomato.

Treatments interaction		Days to flowering	Maturity	Plant height	Fresh shoot weight	Fresh root weight	Dry root weight
Poultry manure (ton/ha)	Rapeseed cake (kg/ha)						
	0	52.33^{ab}	81.67^{a}	54.42^{a}	166.5^{a}	49.85^{a}	11.61^{a}
0	100	52.67^{ab}	81.33^{a}	54.71^{a}	170.5^{a}	49.40^{a}	11.58^{a}
	200	50.00^{abc}	81.33^{a}	54.97^{a}	173.2^{a}	47.99^{ab}	10.62^{ab}
	0	48.33^{def}	80.33^{a}	63.14^{a}	213.6^{b}	47.77^{ab}	10.56^{ab}
5	100	49.67^{ef}	79.67^{ab}	64.81^{a}	219.8^{b}	47.35^{ab}	10.85^{ab}
	200	48.33^{ghi}	79.00^{ab}	64.67^{ab}	232.2^{bc}	46.83^{ab}	9.03^{b}
	0	47.00^{efg}	77.67^{c}	70.81^{ab}	249.4^{c}	48.66^{ab}	8.33^{c}
10	100	45.67^{efgh}	77.33^{c}	74.76^{ab}	251.8^{c}	48.15^{ab}	8.71^{c}
	200	47.33^{ef}	77.33^{c}	76.07^{ab}	260.0^{c}	46.21^{b}	8.33^{c}
	0	43.00^{ghi}	67.67^{d}	109.88^{c}	396.0^{d}	46.49^{b}	7.98^{d}
15	100	42.67^{hi}	67.67^{d}	113.60^{c}	411.3^{e}	46.04^{b}	8.82^{bc}
	200	57.00^{i}	68.00^{d}	102.85^{c}	414.3^{e}	41.07c	9.38^{b}
	0	56.67^{ab}	93.67^{e}	101.57^{c}	450.5^{e}	59.64^{d}	11.11^{a}
20	100	57.00^{bc}	93.00 ^{e}	122.63^{d}	461.2^{e}	61.17^{d}	11.19^{a}
	200	59.67^{a}	93.00^{e}	127.40^{d}	463.8^{e}	63.78^{d}	11.31^{a}
CV		4.81	1.87	19.02	3.23	6.69	8.87
LSD (P < 0.05) for (pm x rc)		3.52	2.67	4.583	17.28	5.75	1.545

pm=poultry manure, rc=rapeseed cake. Means within a column followed by the same letter are not significantly different at P<0.05.

Similar findings also indicted the same result as the root infection increases so does the root weight Riegel and Noe, 2000; Tadele and Mengistu, 2000. Increasing application of poultry manures from 0 to 5, 10, and 15 ton/ha decreased the dry root weight by 7.5, 33.2 and 34.4%. The lowest dry root weight was observed in treatments amended with 10 and 15 ton/ha of poultry manure. However the highest was recorded from the control treatment and tomato plant which amended with 20 ton/ha of poultry manure. The severely infected roots showed increment in dry root weight in response to increased application of poultry manure. Tadele and Mengistu (2000) reported increased dry weight of infected plant compared to the healthy ones. Similarly, Zareen et al. (2001) also indicated that the dry root weight of control treatment inoculated with root-knot nematodes was significantly greater than the dry root weight of all other treatments for tomato.

Effect of poultry manure and rapeseed cake on fruit yield of tomato

The result indicated that there were significant interaction effect of treatments on the fruit yield of the crop at (P<0.001). Poultry manure as main effect significantly affected yield parameters. The main effect of rapeseed cake was non-significant on average fruit weight at (P<0.05) on the yield parameters (Table 3). The result revealed that the lowest number of fruit/plant was recorded from the combination of poultry manure applied at 20 ton/ha and rapeseed cake applied at 200 kg/ha. Average fruit weight increased when the application of poultry manure was raised from 0 to 20 ton/ha. The highest average fruit weight was obtained from application of poultry manure at 20 ton/ha. This increase amounted to 245.87% compared to the average fruit weight of plants grown without the application of poultry manure. Raising the application level of poultry manure form 0 to 15 ton/ha increased the average fruit weight by 15, 35.5 and 111.2%, respectively, compared to the control treatment. This result is in agreement with the findings of Oglumba et al. (2009) who reported that tomato plants treated with organic amendments including poultry manure in the range of 10 to 20 ton/ha produced higher number of fruit/plant as well as weight compared to tomato plants not supplied with organic fertilizer grown in root knot nematode infested soil. Similarly Rhaman et al. (2002) indicated that plots treated with poultry manure at 3 ton/ha and mustard cake at 300 kg/ha produced

Table 3. The interaction effect of poultry manure and rapeseed cake fruit yield**.**

Treatments interaction		Number of fruit/plant	Average fruit weight	Fruit yield (ton/ha)
Poultry manure (ton/ha)	Rapeseed cake (kg/ha)			
	0	19.62[a]	59.07[a]	7.34[a]
0	100	19.70[a]	64.34[ab]	7.85[a]
	200	20.01[a]	66.70[ab]	8.08[a]
	0	20.03[ab]	74.35[ab]	13.79[b]
5	100	21.36[ab]	74.86[ab]	14.02[b]
	200	21.80[ab]	83.32[b]	16.59[b]
	0	22.050[b]	98.44[bc]	16.85[b]
10	100	23.11[b]	100.77[c]	17.41[b]
	200	23.13[b]	103.10[c]	17.95[b]
	0	25.07[c]	183.40[d]	22.18[c]
15	100	25.95[cd]	205.17[e]	22.26[c]
	200	26.94[d]	207.81[e]	23.46[c]
	0	17.04[e]	215.63[f]	12.587[b]
20	100	15.84[e]	215.72[f]	14.067[b]
	200	13.46[f]	218.34[f]	14.543[b]
CV		10.82	12.07	8.08
LSD (0.05) for (pm x rc)		63.34	34.95	0.87

pm=poultry manure, rc=rapeseed cake. Means within a column followed by the same letter are not significantly different at P<0.05.

better results in terms of the number of fruit/plant and fruit weight in tomato. However, in this study, plots amended with 20 ton/ha of poultry manure resulted in significant reduction in the number of fruit/plant.

In general, the result indicated that overall fruit yield was significantly influenced by poultry manure application. Yield increased when application of poultry manure was raised from 0 to 15 ton/ha by 87.37, 119.6 and 199.5%, respectively compared to untreated control. The highest total yield was recorded from a treatment which was amended with 15 ton/ha of poultry manure. The total fruit yield declined significantly when the rate of application of poultry manure was raised to 20 ton/ha. Compared to the total fruit yield obtained from plants that received no poultry manure, plants treated with 20 ton/ha poultry manure had increased total fruit yield by about 46.5%. However, the total fruit yield of plants treated with poultry manure at the rate 15 ton/ha exceeded the total fruit yield of plants treated with 20 ton/ha poultry manure by about 55%. The decline in fruit yield at the higher rate of poultry manure might be due to excessive availability of nutrient promoted the plant vegetative growth in addition to that there was less number of fruit per plant was recorded for the highest poultry manure treatment.

Current finding was in consistent with that of Hassan et al. (2010) who reported that amending soil infested with *Melodoigyne* spp. with organic substances in the range of 15 to 40 ton/ha gave the highest tomato fruit yields. Similarly Oglumba et al. (2009) found the similar result in which tomato treated with optimum (10-15 ton/ha) organic manures gave better fruit and suppressed root-knot nematode.

Conclusion

Application of poultry manure the range of 5 to 15 ton/ha in combination with rapeseed cake at 200 kg/ha significantly reduced root-knot nematode J2 populations, number of egg/egg mass, root galling index and increased tomato fruit yield. Therefore, it could be conclude that incorporating poultry manure at 5 to 15 ton/ha and rapeseed cake at 200 kg/ha into the soil infested with root-knot nematode at least three weeks before transplanting tomato seedlings can significantly suppress root-knot nematode infestation thereby enhancing fruit yield of tomato.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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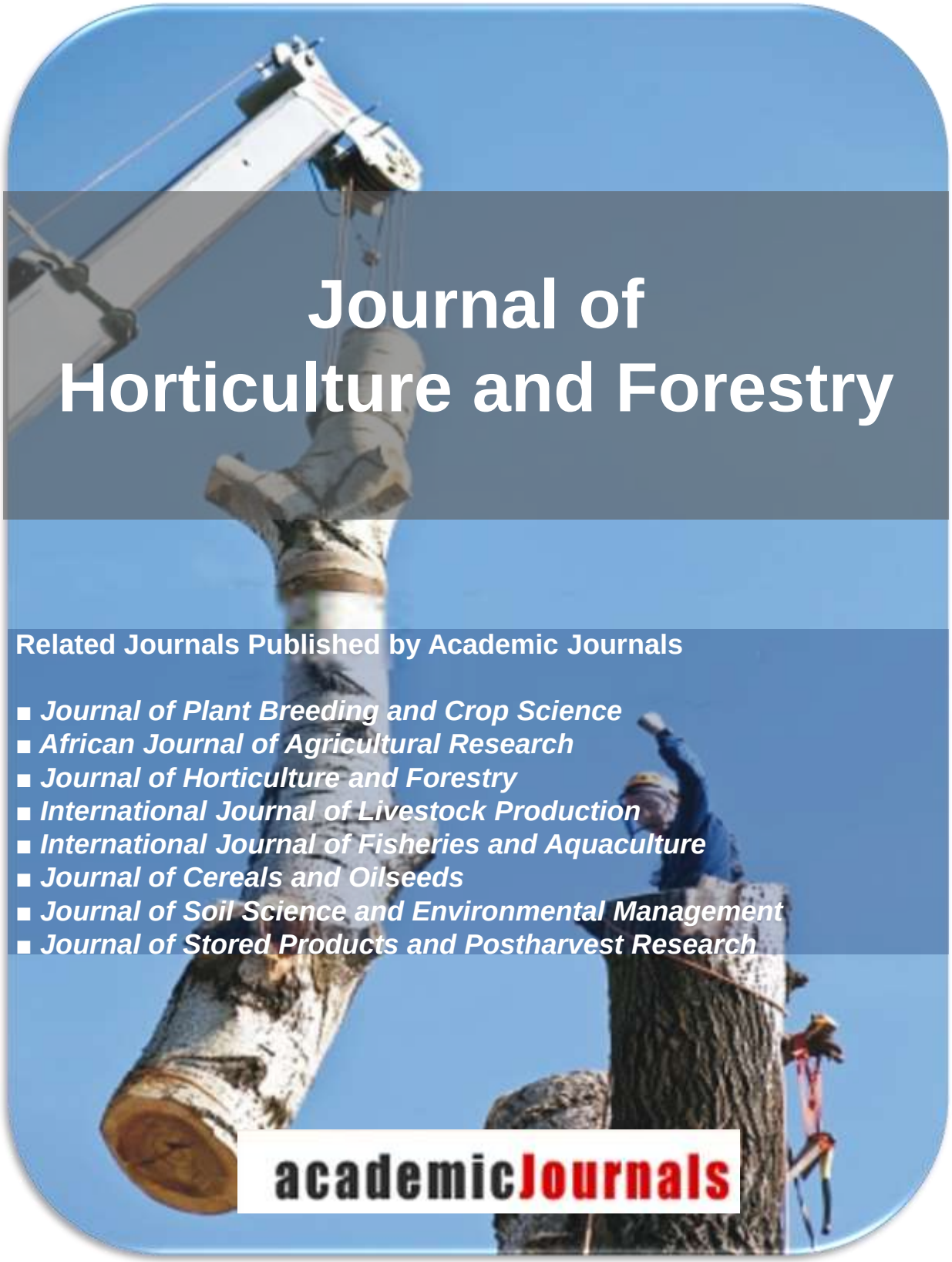

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