RESISTANCE LEVEL OF SOME CITRUS CULTIVARS TO THE CITRUS NEMATODE (*TYLENCHULUS SEMIPENETRANS* COBB) IN WEST JAVA, INDONESIA

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Abstract

This study aimed to determine resistance level of some citrus cultivars to the citrus nematode (*Tylenchulus semipenetrans* Cobb). Number of *T. semipenetrans* female penetrated per cm root not significantly different on all citrus cultivar. Number *T. semipenetrans* female per cm root on all citrus cultivar was < 1 nematode per cm root. This fact showed that *Citrus jambhiri*, *C. reticulate*, *C. aurantium*, *C. nobilis*, *C. medica*, *C. macrocarpa*, and *C. hystryx* ABC showed resistant reaction on *T. Semipenetrans*.
Citrus cultivars have effect on *T. semipenetrans* female development. Number *T. semipenetrans* female that penetrated root system on *C. medica*, *C. microcarpa*, *C. aurantium*, *C. hystrix* ABC, *C. nobilis*, and *C. reticulata* that were 1.25-4.25 nematodes. This fact showed that the sixth citrus cultivars retarded *T. semipenetrans* development.
Resistance level of citrus cultivar to*T. semipenetrans* could determined based on reproductive index (R).
Citrus cultivar have effect on number J2 *T. semipenetrans* in the soil. *C. jambhiri*, *C. reticulata*, *C. aurantium*, *C. nobilis*, and *C. medica* have reproductive index was low (< 1), mean that final population *T. semipenetrans* in the soil decrease. This fact showed that cultivars resistant on *T. semipenetrans*. On the contrary, *C. microcarpa* and *C. hystrix* ABC have reproductive index hight (>1), mean showed that cultivars susceptible on *T. semipenetrans*.
Some citrus cultivars have resistance level that different on *T. semipenetrans* attacking. *C. jambhiri*, *C. reticulata*, and *C. aurantium* were resistant to *T. Semipenetrans*.

Key words : citrus, resistance, slow decline, *Tylenchulus semipenetrans*

Citrus productivity in Indonesia in 2005 about 17-25 t ha^{-1} and their potention was 25-40 t ha^{-1} (Research and Development Agriculture Institute, 2007), while citrus yield in USA and Brazil reached 20-30 t ha^{-1} (Iqbal, 2003). Citrus yield in Indonesia was lower one caused by attacking fungal,bacterial, viral and nematodes that decreased quality and quantity of citrus. The presenced of plant parasitic nematode in citrus plant to become problem that very difficult was handled. In sub-tropic countries, the citrus nematode (*Tylenchulus semipenetrans*) caused citrus damage. Shurtleff and Averre (2000) reported that *T. semipenetrans* caused slow decline diseased, such as plant to become chlorotic, stunt growth, dieback branchs, leaves falled, decrease production, fruit not homogen and small.

The major characteristic of citrus industry used rough lemon (*Citrus jambhiri*) as rootstock. In fact this type very susceptible to*T. semipenetrans*, so that citrus production was low. Now all citrus rootstock was needed screening that could used as resistance resources (Iqbal, 2003).Resistance level of citrus cultivar to *T. semipenetrans* was not known in West Java, Indonesia. Rootstock resistant could used for controlling *T. semipenetrans*. The objective of this research was to determine resistance level from some citrus cultivar to *T. semipenetrans*.

MATERIAL AND METHOD

Some citrus cultivars (*C. jambhiri*, *C. reticulata*, *C. aurantium*, *C. nobilis*, *C. medica*, *C. microcarpa*, and *C. hystrix* ABC) were assayed to determine resistance to the citrus nematode (*Tylenchulussemipenetrans*).The soil was sterilized steam, and seed from seventh citrus cultivar were transplanted in container 150 cm x 60 cm. Citrus seeds were planted with plant distance 5 cm x 5 cm, and seeds were allowed grow until old-3 months. Seedling rough lemon (*C. jambhiri*) old-6 months was planted in the pot in green house. After one week transplanted, pots were inoculated by 2000 J2 *T.semipenetrans*. J2 *T.semipenetrans* were collected from soil in citrus plant around in Garut, Indonesia.

Eggs Isolation and extraction juvenille*T.semipenetrans.* Root infected by *T.seipenetrans* were taken from pots and were washed carefully with tap water. Roots were cutted to become 1-2 cm and were removed in beaker glass containing 200 mL sodium hypochlorite solution of 0.5 %, and then were stirred for 3-4

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minutes to solve egg sac (gelatin matrix) and release eggs from egg mass. Nematode suspension were sieved through sieving 75 µm to separate root fragments, and eggs nematode suspensi on sieving 50 µm, and 35 µm were washed and poured in beaker glass (Hussey and Barker, 1973). Eggs suspension were poured in petri dish and incubated for 3-4 days at room temperature for egg hatching. J2 *T. semipenetrans* that hatched were used for treatment inoculation.

T.semipenetrans Inoculation. Citrus seedling from each cultivar that old-3 months in seedling, then were planted in pot filled soil pasteurized steam. And then pots were inoculated with 200 J2 *T. semipenetrans* in the hold 5 cm deep at seedling around. After inoculation, the hold were filled again with soil, and watered daily.

Observation. The number of J2 *T.semipenetrans* in 100 mL soil were counted on 30 days after inoculation. J2 *T.semipenetrans* in the soil were extracted with Baermann funnel, and then number of J2 *T.semipenetrans* were counted under binocular microscope. After 30 days inoculation *T.semipenetrans*, citrus seedling were taken carefully with scope and were washed with tap water, and then were stained with lactophenol fuchsin solution. After 24 hours *T. semipenetrans* female that penetrate root were counted under microscope. Number of *T.semipenetrans* females per cm of root on each treatment were counted based on number *T.semipenetrans* female were devided with long of root system. Data were analysed by analysis of variance and to different of treatment means were used Duncan' multiple range test.

Resistance level of citrus plant on *T.semipenetrans* was determined based on number juvenilles per cm of root (Mani, 1989 *cit* Iqbal, 2003) was modifiefd as follow :

0 - ≤ 1 nematode per cm root was resistant

>1 - ≤ 2 nematode per cm root was moderately resistant

> 2 nematodes per cm root was susceptible.

Resistance level other of citrus cultivar on *T.semipenetrans*could determined based on reproductive index (R) that was comparison between J2 final population (Pf) with J2 initial population (Pi). If R < 1 means final population decrease, that showed resistant cultivar, while if R>1 means final population increased, that showed susceptible cultivar (Noe, 1985).

RESULTS AND DISCUSSIONS

Number of *T.semipenetrans* female penetrated per cm root not different on all citrus cultivar (table 1). Number *T.semipenetrans* female per cm root on all citrus cultivar was < 1 per cm root. This fact showed that *Citrus jambhiri*, *C. reticulate*, *C. aurantium*,*C. nobilis*, *C. medica*, on *T. semipenetrsndC. microcarpa*, and *C. hystryx* ABC showed resistant reaction on *T. semipenetrans.*

Table 1

Rates of Number *Tylenchulus semipenetrans* Female that Ppenetrated per cm of Root on Some Citrus Cultivars on 30 Days After Inoculation

Citrus cultivar	Number *T. semipenetrans* female per cm root	Cultivar reaction to *T. semipenetrans*
C. jambhiri	0.1050 a	Resistant
C. reticulata	0.0675 a	Resistant
C. aurantium	0.0550 a	Resistant
C. nobilis	0.0725 a	Resistant
C. medica	0.0525 a	Resistant
C. microcarpa	0.0600 a	Resistant
C. hystrix ABC	0.5400 a	Resistant

Table 2

Rates number of *T. semipenetrans* female per root system in some citrus cultivars on 30 days after inoculation

Citrus cultivar	Number of *T. semipenetrans* female per root system
C. jambhiri	10.50 a
C. reticulata	4.25 b
C. aurantium	3.25 b
C. nobilis	4.00 b
C. medica	1.25 b
C. microcarpa	1.75 b
C. hystrix ABC	3.75 b

Means in the same column followed by the same letter were not significantly different (P=0.05) according to Duncan's multiple range test. Rates of number T. semipenetrans female per root system in some of citrus (table 2).

Means in the same column followed by the same letter were not significantly different (*P*=0.05) according to Duncan's multiple range test.

Citrus cultivars have effect on *T. semipenetrans* female development. Number of *T.semipenetranas* female that penetrated root system on *C. jambhiri* highest that is 10.50 *T.semipenetrans* females, while number *T.semipenetrans* female that penetrated root system on *C.medica*, *C.microcarpa*, *C.aurantium*, *C.hystrix* ABC, *C.nobilis*, and *C.reticulata* that were 1.25-4.25 nematodes. This fact showed that the sixth citrus cultivars retarded*T.semipenetrans* development.Resistance level of citrus cultivar on *T.semipenetrans* could determined based on reproductive index (R) (table 3).

Table 3

Initial Population, Final Population, Reproductive Index, and reaction of citrus cultivars to*T.semipenetrans* on 30 days after inoculation

Citrus cultivar	Initial population number J2 per 100 mL of soil (Pi)	Final population number J2 per 100 mL of soil (Pf)	Reproductive index (R) = Pf/Pi	Reaction of citrus cultivar on *T. semipenetrans*
C. jambhiri	200	10.25	< 1	Resistant
C. reticulata	200	12.50	< 1	Resistant
C. aurantium	200	4.75	< 1	Resistant
C. nobilis	200	171.00	< 1	Resistant
C. medica	200	186.25	< 1	Resistant
C. microcarpa	200	525.50	> 1	Susceptible
C. hystrix ABC	200	281.25	>1	Susceptible

Citrus cultivar have effect on number J2 *T.semipenetrans* in the soil. Number of J2 *T.semipenetrans* in the soil was lowest was reached at *C.aurantium*, *C. jambhiri*, and *C.reticulata* cultivar that were 4.75, 10.25, and 12.5 J2 per 100 ml, respectively. Number J2 highest on *C.microcarpa*, and then *C.hystrix* ABC, *C.medica*, and *C.nobilis*.

C.jambhiri, *C. reticulata*, *C. aurantium*, *C.nobilis*, and C. medica have reproductive index was low (< 1), mean that final population *T.semipenetrans*in the soil decrease,. This fact showed that cultivars resistant on *T.semipenetrans*. Sebaliknya, *C. microcarpa* and *C. hystrix* ABC have reproductive index hight (>1), mean showed that cultivars susceptible on *T. semipenetrans*.

C. jambhiri, *C. reticulata*, *C. aurantium* cultivars were resistant on *T. semipenetrans* because can retard J2 population development, so that J2 final population very low, selain itu plant growth and root development lebih baik were compared with other cultivar. *C. nobilis* and *C. medica* although resistant but less retarded J2 population, so that number of J2 final population near number J2 initial population or reproductive index near 1, selain itu their root less was compared with *C. jambhiri*, *C.reticulata*, and *C. aurantium*.*C. microcarpa* and *C. hystrix* ABC were susceptible on *T. semipenetrans* because could tolerate *T. semipenetrans* reproduction their resulted final population J2 increased, root development and plant growth decreased.

Whitehead (1998) states that nematode attacking in root resulted drying increased because decrease in take soil hara, so that shoot growth was retarded. Population density of *T. semipenetrans* in the soil low on *C. aurantium*, *C. jambhiri*, and *C. reticulata*, so that root destruction that was caused low, their resulted shoot growth lebih baik. If root was attacked, the cortex separate very fast, and then root decay or damage resulted decreasing root volume so that root weight low.

Based on recognition number of J2 *T. semipenetrans* in trhe soil, reproductive index (R), and number*T. semipenetrans* female in root, maka could recommended that *C. Aurantium*, *C. Jambhiri*, and *C.reticulata* could used as alternative as rootstock development that resistant on *T. semipenetrans*. This fact because *C. jambhiri*, *C. reticulata*, and *C. aurantium* could resulted reproductive index < 1, number *T. semipenetrans* female per cm root < 1, and number J2 *T. semipenetrans* in the soil lowest were compared with other cultivars. Selain itu, that citrus cultivars have shoot weight and root well compared with other cultivars.

C. aurantium and *C. reticulata* could used as alternative for rootstock in citrus seedling development, that exception *C. jambhiri* (RL) and Japanesche Citrun (JC) that now still were used as rootstock. *C.aurantium*, *C. reticulata*, and *C.*

jambhiri showed rasistant reaction on *T. semipenetrans*. Number juvenilles decrease until to become adult female in citrus cultivar root that resistant showed there were some factors that not suitable for infection site and nematode development.

C. aurantium resistant on *T. semipenetrans*, this fact according with Jagdale *et al*. (1984) stated that *C. sinensis* or *C. aurantium* caused *T. semipenetrans* female and juvenilles population low on 17 citrus seedling in India and considered very resistant. Based on histopathology study was done by Kaplan (1981) showed that plant resistant on *T. semipenetrans* were charactered with there are hypersensitive respons on infection site of nematode, and caused periderm injury formation.

CONCLUSIONS

Some citrus cultivars have resistance level that different to *T. semipenetrans* attacking. *C. jambhiri*, *C. reticulata*, and *C. aurantium* were resistant to *T. semipenetrans*.

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