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NEMATICIDAL ACTIVITY OF AQUEOUS EXTRACTS FROM LEAVES OF *RUTA GRAVEOLENS* ON *XIPHINEMA INDEX*

by
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Summary. Aqueous extracts from leaves of *Ruta graveolens* had a high nematicidal effect against *Xiphinema index*, *in vitro*. The LD-50 values, obtained by probit analysis, ranged between 83.14% and 1.79% of a standard solution. Nematode mortality increased with the increase of the leaf extract concentration and the exposure time. There was a high negative correlation ($r = -0.973$) between the \log_{10} of these two parameters.

Several natural products from plants possess nematicidal or nematostatic properties (Gommers, 1981).

An *in vitro* experiment was undertaken to investigate the effect of aqueous extracts from leaves of *Ruta graveolens* L. on *Xiphinema index* Thorne et Allen, a nematode commonly occurring in Italian vineyards (Roca and Lamberti, 1978).

Materials and methods

The leaf extract of *Ruta graveolens* was prepared by soaking 50 g of green leaves in 200 ml distilled water for 24 hrs, interval thought sufficient to solubilize the active principles. The leaves were then comminuted in a blender and the suspension filtered through a filter paper. This filtrate, which had pH 7.3, was designated as standard (S). It was diluted with distilled water to provide 2%, 4%, 8%, 16%, 32% and 64% concentrations of the standard.

X. index, was cultured on commercial fig (*Ficus carica* L.) in a glasshouse at 20-25 °C; nematodes were extracted by the Cobb's decanting and sieving method.

Three millilitres of each leaf extract concentration were poured into 3.5 cm diam Petri dishes in which 20 female *X. index* were placed. The same number of nematodes placed in distilled water served as a control. The Petri dishes were left at room temperature (20-25 °C) and observed for nematode mortality after 3, 6, 12, 24 and 48 hrs.

There were three replicates of each treatment according to a complete randomized design.

Nematodes that did not display motility, after transfer to distilled water for 1 hour and when stimulated on head or vulva, were considered dead.

Data were subjected to probit analysis to estimate LD-50 of doses applied for each exposure period.

Results and discussion

After 3 hrs exposure the maximum nematode mortality (30.6%) occurred at the highest concentration of leaf extract tested (64%). To obtain a 50% mortality of *X. index* with 3 hrs exposure a concentration of 83.14% S would be required (fiducial limits 116.80-59.09) (Fig. 1).

Doubling the exposure time (6 hrs) resulted in a remarkable decrease of the concentration required for 50% mortality. In this case a concentration of 17.29% S (f. l. 19.56-15.19) was necessary to achieve the LD-50, while a maximum mortality of 96.6% was obtained in the 64% S concentration.

After 12 hrs exposure the LD-50 value was estimated as 11.37% S concentration (f. l. 12.78-10.12), while 100% mortality was obtained at the 32% S concentration.

Exposure for 24 and 48 hrs gave LD-50 values of 6.90% S (f. l. 7.75-6.15) and 1.79% S (f. l. 2.28-1.40) respectively (Fig. 1), and 100% nematode mortality at 16% and 8% S respectively. Nematode mortality was not observed in the control.

The relationship between LD-50 and exposure period (Fig. 2) shows a high negative correlation ($r = -0.973$) between the \log_{10} of these two parameters.

Leaf extract of *R. graveolens* has a greater nematicidal effect on *X. index* than the extract from pods of chilli pepper (Sasanelli and Catalano, 1991) under similar experimental conditions. Although the active ingredient involved has not been identified, the results represent the first record of the nematicidal properties of *R. graveolens* leaves. This plant contains high levels of alkaloids (structurally and biogenetically included in the family of antranilic acid), terpenes (limonene, pinene and cineole), coumarins (xanthotoxin) and flavonoids (rutin).

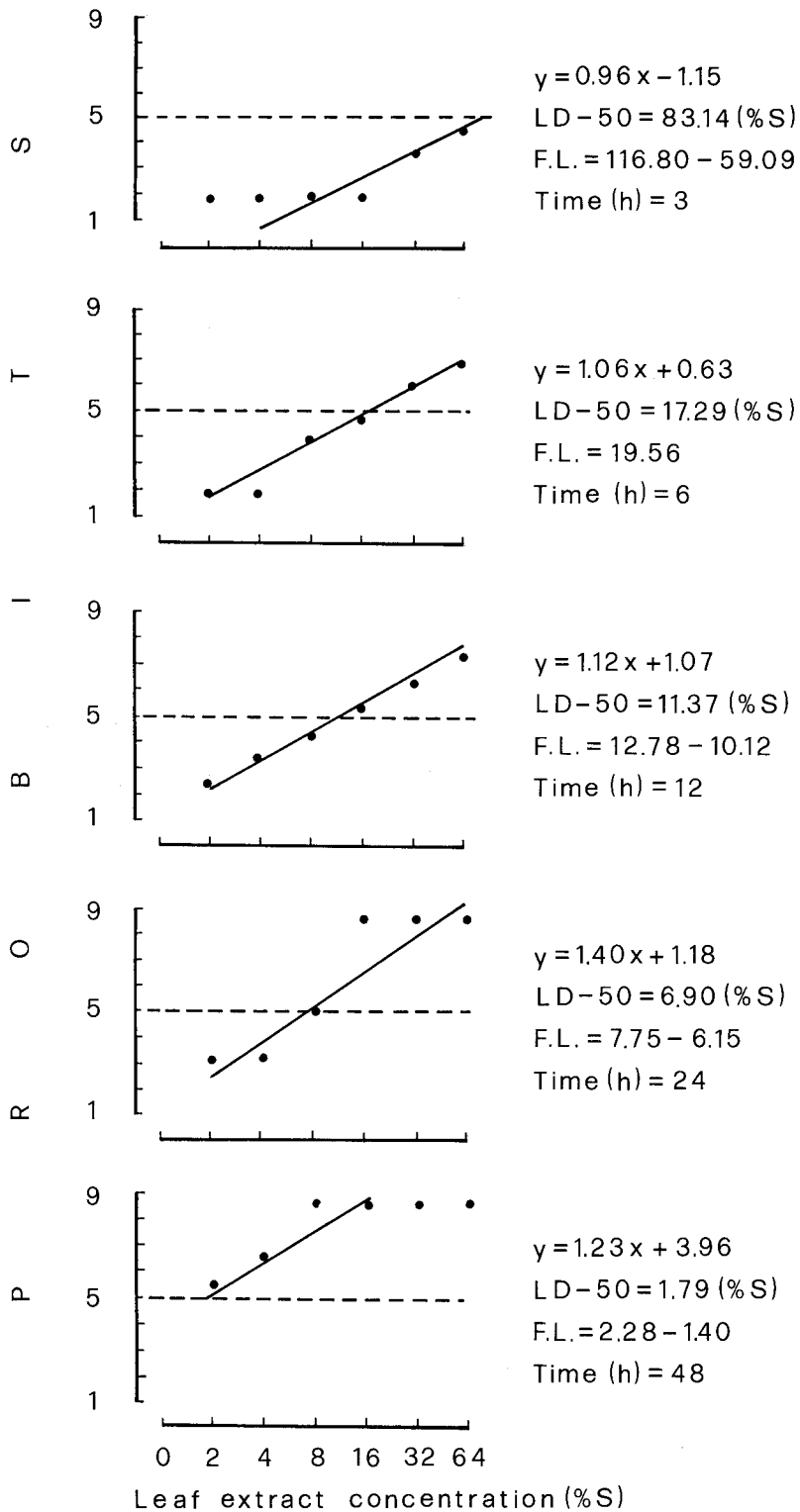


Fig. 1 - Relationship between the log₁₀ concentration of leaf extract of *Rutta graveolens* and probit mortality of *Xipbinema index*, at different exposure times.

It is well known that many alkaloids have nematicidal and insecticidal activities (Onda *et al.*, 1965; Fassuliotis and Skucas, 1969; Allen and Feldmesser, 1970; 1971; Reese and Holyoke, 1987). However, other substances present in *R. graveolens*, like limonene, pinene, xanthotoxin and rutin are toxic to insects (Holyoke and Reese, 1987).

It is not yet known whether the nematicidal activity of leaf extracts of *R. graveolens* is attributable to a single compound or to a synergism between several compounds. Therefore, further studies are needed to obtain more information on the natural products from *R. graveolens*.

Although our results show promise it is too early to suggest the use of *R. graveolens* as a green manure for nematode control and investigations under field conditions are necessary.

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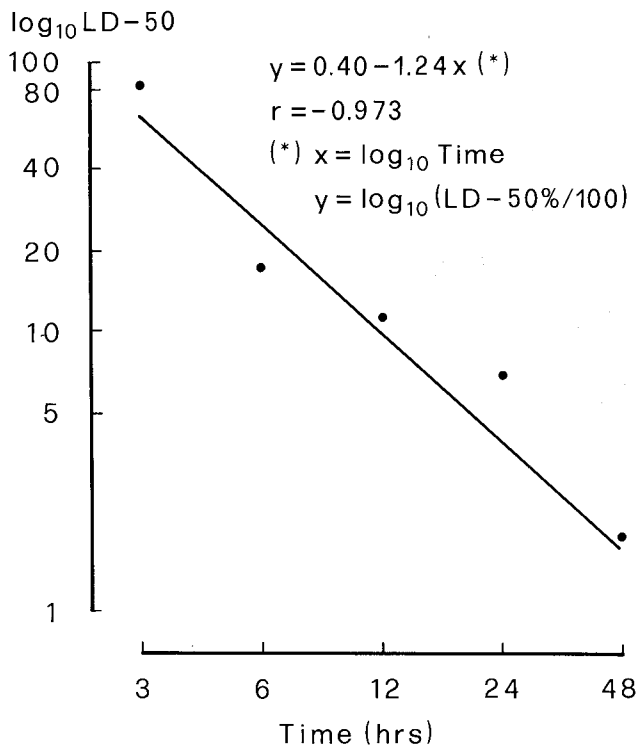


Fig. 2 - Relationship between \log_{10} LD-50 and \log_{10} exposure time.

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