EXPLORING THE NEMATICIDAL POTENTIAL OF CASHEW NUT SHELL LIQUID AGAINST ROOT-KNOT NEMATODES: *IN VITRO* AND GREENHOUSE EVALUATIONS

Abstract

Root-knot nematodes(*Meloidogyne* spp., RKNs) remain a major threat to vegetable production and a reason for the overuse of pesticides, particularly in Sub-Saharan Africa. The objective of the present study was therefore to investigate the nematicidal properties of cashew nut shell liquid (CNSL), a by-product of cashew nut processing, against RKNs on vegetables. For this, laboratory bioassays and greenhouse pot experiment were conducted to assess the suppressive effectsof CNSLonRKN reproduction and damage. Four concentrations (0.01%, 0.1%, 1%, and 10%) of cold- and hot-extracted CNSL were used in the laboratory bioassays to evaluate their nematicidal activity against*M. enterolobii* juveniles end eggs. The greenhouse pot experimentconsisted of eight treatments (two CNSL types, one concentration, three application doses, and two controls) assessed in a completely randomized block design with five replicates, using tomato (cv. Padma 108 F1) and soil naturally infested with RKNs. Results from the *in vitro* bioassays showed that CNSL induced juvenile death by up to 90% after 48 h of exposure against 2% in the SDW used as control. Egg hatching was also inhibited by up to 88% after 8 days of exposure to CNLS against 6% for SDW. Under greenhouse conditions, application of 100 ml of CNSL induced a significant reduction in reproduction factor by up to 78.77 % and 91.47% compared with*Crateva adansonii* leaf aqueous extract and untreated positive control treatments, respectively. Cold-extracted CNSL were more effective in suppressing nematode reproduction and root galling compared with hot-extracted CNSL. Tomato plant growth, particularly fresh shoot and root weights, were negatively affected by CNSLapplication, indicating the needs for low concentrations. Although further studies should be conducted to identify effective and non-phytotoxic concentrations of CNSL, these initial results are positive and show promise, particularly for intensive peri-urban and urban vegetable production systems of West-Africa.

Keywords:-Botanical extracts, CNSL, *Meloidogyne* spp., peri-urban and urban agriculture, plant-parasitic nematodes, tomato.

Introduction:-

Root-knot nematodes (*Meloidogyne* spp.; RKNs) are a malignant soilborne curse to vegetables, particularly in tropical and subtropical areas where vegetable production is highly dependent on proper nematode control (Coyne *et al.*, 2018; Hallmann and Meressa, 2018).In Sub-Saharan Africa, RKNs were identified as the single greatest biotic threat in the highly intensive urban and peri-urban vegetable production systems, where vegetable production contributes significantly to food security, family income and employment. These nematodes reduce the absorption and assimilation capacity of the root system, resulting in a decrease in the plant's mineral nutrition, flowering and fructification (Nilusmas, 2020). Furthermore, several authors have reported that initial infestation of tomato plants by RKNs inhibits the expression of resistance genes to some major diseases, and therefore, predispose infested crops to secondary infections by fungi, bacteria, and viruses, leading to increased yield losses (Kidane *et al.*, 2020; Walia and Khan, 2023).

Various strategies are available to protect vegetables in these cropping systems against RKN, but synthetic nematicides remainthe most common coping strategy relied upon by growers. According to Trudgill and Blok (2001), RKNs are very difficult to eradicate due to the wide diversity of their hosts, their very short life cycle, their very high reproduction rate and their endoparasitic nature. This could be also due to the presence of a mixture of RKN species in the same fields, including the highly pathogenic and invasive species *M. enterolobii* (Pagan *et al*., 2015; Affokpon *et al.*, 2017). For example, *M. enterolobii* has been found to exhibit resistance to standard control measures against RKNs (Coffi *et al*., 2025; Poudel *et al.*, 2025). However, pesticide use in sub-Saharan Africa presents a number of challenges, among which the limited or inaccurate understanding by farmers has led to their misuse and overuse (Coyne *et al.*, 2010). Vegetables produced under such situations are

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delivered direct to markets with little or no care in respect to quality or consumer health and safety. As a consequence of their detrimental effects on the environment and human health, environmentally sound alternatives are being sought and investigated. The use of botanical extracts appears to be an important component of integrated methods for managing these nematodes.

Recent research has addressed the pesticidal effects of cashew nut shell liquid (CNSL), a by-product of cashew (*Anacardium occidentale*, L.) nuts abundantly available in any cashew-producing countries. CNSL, either cold- or hot-extracted, has been shown to have pesticidal effects on different developmental stages of various insect pests and pathogenic fungal growth(Garcia *et al*., 2018;Lekha *et al*., 2020; Sotondji*et al.,* 2020). Its insecticidal efficacy against *Aphis craccivora* and *Riptortus pedestris* infestingcowpea, was reported earlier in *in vitro* assay where topical application of concentrations 0.1 % caused mortality comparable to that of the chemicalinsecticide Thiamethoxam 0.03% (Lekha *et al*., 2020). These authors observed that the survived *R. pedestris* exhibited developmental abnormalities and formation of nymphal adult intermediary, and suggested a possible insect growth regulatory effect of CNLS. Recently, the CNLS has been proved to have insecticidal effects on *Spodeptera frugiperda* larva and eggs and its antifeeding properties with average lethal concentrations (LC_{50}) of 1.92% and 2.50% after 2 h for larvae and eggs, respectively, and effective concentrations (EC_{50}) of 0.34% for antifeedant effects (Nyirenda *et al.,* 2024). At a concentration of 320 µg mL-1, CNSL was found to inhibit sporulation and mycelial growth of *Collectotrichum gloesporiodes* and *Lasiodiplodia theobromae* on papaya fruit (Garcia *et al.*, 2018). They reported that no reduction in fungal growthwas observed in the absence of direct contact between the fungus and the CNSL, indicating that the inhibitory effect of CNLS was not due to volatile substances.Anacardic acids are the main constituents of the CNSL, which formscardol, cardanol and polymeric material during the decarboxylation process, some phenolic compounds known for their biological activity against various pests and diseases ((Maia et al., 2015; Garcia et al., 2018). However, reports on the bioactivity of CSNL against plant-parasitic nematodes are scarce.

This study was conducted to investigate the nematicidal properties of CSNL against RKNs on vegetable crops. The specific objectives were to assess *in vitro*the inhibitory effect of both cold- and hot-extracted CNSL on juveniles and eggsof*M. enterolobii*and to determine under greenhouse conditions their effectiveness in protecting tomato against RKNs.

Material and methods:-

Experimental sites:-

The experiments were carried out in the laboratory facilities and greenhouse at the Nematology Unit (UNema) research station in Sekou, Benin (06°37'30.8" N; 002°13'56.9" E; 74 m asl). The research station is located in the Guinean biogeographical zone and characterized by a sub-equatorial climate with two wet seasons (April to mid-July and mid-September to end October) alternated by two dry seasons (Affokpon *et al.*, 2021).

Preparation of nematode inoculum:-

Second stage juveniles and eggs of *M. enterolobii* were used for the *in vitro* bioassay. They were obtained from a pure population maintained on celosia (*Celosia argentea* L.) in the greenhouse of the Nematology Unit following the method described by Coffi *et al.* (2025).

For the pot experiment, infested soil was collected from *in vivo* RKN multiplication plots of the UNema research station. These plots were initially inoculated with a mixture of *M. enterolobii*, *M. incognita* and *M. javanica*and grown with celosia for RKN inoculum production purpose.

Preparation of the CNSL and leaf aqueous extract of *Crateva adansonii*:-

The two types of CNSL, cold and hot extracts, wereproduced from cashew(*Anacardium occidentale*, L.) shells. The products were provided by a local supplier located at Savalou, Benin. For the *experiments*, a stock solution of the CNSL was diluted in sterile distilled water (SDW) using serial dilution method to obtain 10^{-1}, 10^{-2}, 10^{-3} and 10^{-4} suspensions, corresponding to 10%, 1%, 0.1% and 0.01%, respectively (hereafter referred to as concentrations).

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For the pot experiment, aqueous suspension from leaves of *C. adansonii* (Capparaceae*;* DC, 1824) was additionally used as a control treatment. Compost from *C. adansonii* has been recently found to have nematicidal effect on *M. enterolobii* (Sero *et al*., 2025a). To prepare the suspension, freshly harvested leaves, collected from the plantation of Nematology Unit, were carefully washed and cut into small pieces. Then, 200 g was macerated using a hand blender (BOSCH, 400 W) in 500 mLSDW. The resulting solution was filtered through a 200 µm mesh sieve 24 hours later, and used for the experiment.

In vitro bioassay:-

Two *in vitro* experiments, repeated once, were conducted to assess the suppressive effect of the CNSL on *M. enterolobii* J2 mortality and their inhibitory effect on egg hatching.

Juvenile mortality bioassay:-

The effect of the CNSL on *M. enterolobii* J2s was assessed *in vitro*after 12, 24 and 48 hours of exposure (Ahmad and Siddiqui, 2018) to four concentrations of each CNSL type. Nine treatments were assessed: cold-extracted CNSLat 0.01%, 0.1%, 1%, and 10%; hot-extracted CNSL at 0.01%, 0.1%, 1%, and 10%; and SDW used as control. Using an Eppendorf micropipette, a 100 µl of aliquot 50 J2s was inoculated into individual 55-mm diameter Petri dishes containing 1 mL of a single treatment. The Petri dishes were arranged on the laboratory bench at ambient temperature (28 ± 1°C) following a completely randomised design with four replicates. The number of dead J2s was counted using the technique described by Gebrehana *et al.* (2025) after 12, 24 and 48 hours of exposure to CNSL. For this, nematode suspensions from the CNLS treatments were washed through nested 20 µm sieves using SDW and J2s were transferred to a counting dish for observation and estimation under a light microscope (20 x magnification, Olympus CX31).For each exposure time, J2 mortality was determined as follows:

J2 mortality (%) = (Dead J2 number / Total J2 number) x 100

Egg hatching inhibition bioassay:-

The egg hatching inhibition potential of the CNLS was assessed using nine treatments as for the J2 mortality bioassay: two CNLS types (cold and hot extracts), four concentrations per CNLS type (0.01%, 0.1%, 1%, and 10%), and a control (SDW). Using a tong, one egg mass was placed in individual Petri dishes (55-mm diameter) containing 1 mL of a single treatment. The Petri dishes were arranged in a completely randomised design with four replicates on the laboratory bench at ambient temperature (28 ± 1°C). The number of hatched J2s was counted 7 days after exposure to each treatment (Machal *et al.*, 2025) using a light microscope following a procedure described for J2 mortality experiment.The hatching inhibition rate was determined as follows:

Egg hatching inhibition (%) = (number of unhatched eggs / Total number of eggs) x 100

Greenhouse pot experiment setup:-

The pot experiment, repeated once, included eight treatments: two CNLS types (cold and hot extracts), three application doses (25 ml, 50 ml, and 100 ml), one concentration (10%,based on the *in vitro* bioassay), apositive control (no CNLS application) and a reference control (application of leaf aqueous extract of *C. adansonii* at 50 ml).

Plastic pots (1.5 L capacity) were individually filled with 1000 $cm^3$of unsterilized soil infested with RKNs collected from UNema experimental field, as indicated above. For both experiments, the initial nematode population densities were adjusted to 1140 individual per pot by adding sterilized soil to the field soil. Dependingon the treatments, pots were then inoculated with CNLS and aqueous extracts of *C. adansonii* and thoroughly mixed. Twenty-four hours later, four week-old tomato seedlings (cv. Padma 108 F1) were singly transplanted into each pot. Pots were arranged in a completely randomized design with five replications. Plants were watered manually (twice a day) with tap water and maintained for eight weeks before harvest.

Estimation of nematode reproduction and root damage:-

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At harvest, the effect of the treatments on nematode reproduction and root galling were assessed through the following parameters: soil and root final nematode population densities, galling index and number of egg masses per g root.

The galling index was assessed per plant on the entire root system after plants were uprooted using a 0–10 rating scale, where 0 = no galling damage and 10 = 91-100 % galled roots (Affokpon *et al.*, 2012). To estimate the production of egg masses, part of the root systems was stained with 20% McCormick red food colour for 15 min (Coffi *et al*., 2025). Then, 1 g root was removed and the egg masses were counted under a stereomicroscope

Final nematode population densities were determined in roots and soilat harvest. Briefly, J2s and eggs were extracted from the total root system and from 250 cm^3 soil using the centrifugation technique described by Affokpon *et al.* (2011). Nematodes were counted under a compound microscope (Olympus CX31) from 3 ml aliquots removed from50 ml and 100 ml of the soil and root suspensions, respectively. Nematode reproductive factor (RF) was then calculated per pot as the ratio between the final number of soil and root nematodes (J2s and eggs) and the initial nematode inoculum.

Measurement of Crop Growth Characters:-

At eight weeks after tomato transplanting, shoot length, stem girth, fresh shoot weight and fresh root weight were measured to determine the effect of the CNLS application on tomato growth.

Statistical analysis:-

R software version 4.4.2 (R Core Team, 2024) was used to perform the statistical analyses. Data on nematode counts were transformed to log (x+1) to conform to normal distribution prior to statistical analysis (Gomez and Gomez, 1984). All data weresubject to ANOVA andmeans were separated using the Fisher LSD procedure at 5%. The lethal concentrations (LC_{50}) were calculated from J2 mortality and egg inhibition percentages, which were corrected using Abbott's formula; the "drm" function from the drc package was used (Ritz *et al.*, 2016), as it allows the fitting of different non-linear dose-response models to binary data.The arguments fct = LL.2 and type = 'binomial' was applied, indicating that the two-parameter logistic function and binomial data type were selected. The LC_{50} was obtained using the ED function from the 'drc' package. For both *in vitro* and pot bioassays, datafrom the repeated experiments werepooled for the ANOVA after determining that the interactiontreatment x experiment was not significant (Affokpon *et al.*, 2011).

Results:-

In vitro bioassay:-

Effect of cashew balm on *M. enterolobii* juveniles and eggs:-

Regardless of the types of CNSL, in most cases, their suppressive effect on *M. enterolobii* J2s increased consistently with exposure time and CNSL dilution (Figure 1). Comparing the two CNSL types, for the same dilution, cold-extracted CNSL has induced higherJ2 mortality rates than hot-extracted CNSL, except for the concentration 10%. Cold-extracted CNSL at concentrations 1% and 10% caused more than 50% dead of J2s at 12h exposure and reachedapprox. 85% dead at 48h exposure time for the concentration 10%. In hot-extracted CNSL treatments, only the concentration 10% caused J2 mortality rate higher than 50%, regardless of the exposure time, but reached 90% at 48 hours. In contrast, the J2 mortality rate in the SDW average 2%. The linear regression analysis showed that the lethal concentrations (LC_{50}) against *M. enterolobii* J2safter 48 hours of exposure were 3.368 µl/ml and 18.156 µl/ml for cold- and hot-extracted CNSL, respectively (Figure 2).

Effect of CNSL on*M. enterolobii* eggs:-

Regarding the effects of CNSL on *M. enterolobii* eggs, the hatching inhibition rate in CNSL suspensions after 8 days of exposure was significantly higher ($P<0.05$) than that recorded in SDW (6.13%) for the same period, except for the cold-extracted CNSL at concentration 0.01% (Figure 3). The highest egg hatching inhibition rates were observed at 10% CNSL concentration, with 88.25% and 84.88% for cold and hot extracted CNSL, respectively. The lethal concentrations (LC_{50}) for hatching inhibition of*M. enterolobii*egg after 8 days of exposure were 1.133 µl/ml and 4.156 µl/ml for cold- and hot-extracted CNSL, respectively (Figure 3).

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Greenhouse pot experiment:-

Effect of cashew balm on nematode reproduction and root infestations:-

Application of CNSLin unsterilized soil planted with tomato resulted, in most cases,in significant reductions of nematode final population densities, reproductive factor, gall index, and number of egg masses compared with the untreated positive control (Table 1). The highest reductions of the final nematode (J2s and eggs) population densities were recorded from soil treated with 100 ml of cold-extracted CNSL with 78.77 % and 91.47% reductions, compared with *C. adansonii*leaf aqueous extractand untreated positive controls. For the same CNSL application doses, final nematode population densities in cold-extracted CNSL treatments were at least 20% lower than those in the hot-extracted CNSL treatments. Nematode reproductive factor (RF) was also significantly inhibited following CNLS application, with reduction rates similar to those of nematode population densities (Table 1). The same trends were also observed when comparing the average RF values between CNSL types for the same application dose. Regarding the gall index, significant inhibition of root galling was observed in tomato plants from CNSL treated pots compared with the untreated positive pots, except for the low dose (25 ml of CNSL). Cold-extracted CNLS applied at 100 ml / pot induced the highest gall index reduction by up to 54%, 78%, and 85% compared with the same dose for hot-extracted CNLS, *C. adansonii* treatment and the positive control, respectively (Table 1). Egg mass production was also significantly inhibited following CNSL application at doses 50 ml and 100 ml per pot for the cold extract and 100 ml/pot for the hot extract. However, there was no significant difference in number of egg masses per g roots between CNSL types at application dose of 100 ml/pot(Table 1).

Effect of the CNLS application on plant growth:-

The potential effect of the CNSL on plant growth was assessed using tomato(cv. Padma 108 F1). There were significant reductions in shoot length (SL), stem girth (SG), fresh shoot weight (FSW) and fresh root weight (FRW) in CNSL treated pots compared with the control pots, except hot-extracted CNSL applied at 25 ml/pot for SL and SG (Table 2). Notable reductions were particularly observed for cold-extracted CNSL, regardless of the application doses, and for hot-extracted CNSL applied at 50 ml and 100 ml per pot. SL, SG, FSW, and FRW were 74%, 82%, 86%, and 69.13%, respectively, lower in pots treated with cold-extracted CNSL at 100 ml than in untreated control pots(Table 2).

Discussion:-

Meloidogyne enterolobii has been reported to overcome standard control measures developed against RKNs, including chemical control and use of resistant varieties (Coffi *et al*., 2025; Poudel *et al.*, 2025). This aggressive RKN species is being spread in Sub-Saharan Africa (Pagan et al. 2015; Kolombia *et al*., 2016; Affokpon *et al.*, 2017), and is therefore considered a serious threat for food security in this region. The results of the current study indicated the suppressive effect of CNSL against *M. enterolobii* juveniles and an inhibitory effect on egg hatching. To the best of our knowledge, no or very fewstudies have been conducted on nematicidal properties of CNLS. In contrast, this product has been tested for its pesticidal effect against nematode parasites of animal (Ademola and Eloff, 2011), insect pests (Sotondji, *et al*., 2022; Nyirenda *et al*., 2024; Sivaranjani *et al.*, 2026), mites (De Carvalho Castro *et al*., 2019), and pathogens (Garcia *et al.*, 2018). Phytochemical analysis of CNSL indicated the presence of various bioactive compounds, including anacardic acid, phenols, tannins, terpenoids, amino acids, flavonoids, and saponins (Kumar et al, 2022; Ashong *et al.*, 2025). According to Gracia *et al*. (2018), after the entrance of CNSL into the intracellular environment facilitated by the formation of chains, its phenol groups work to inactivate enzymes of energy synthesis, therefore possessing toxic potential for pathogens. This is in alignment with Dix (1995) who explained that the phenol groups inactivate enzymes related to energy production and the synthesis of basic metabolites. Assessing the insecticidal potential of CNSL on *Aedes Aegypti* larvae, Passari et al. (2015) observed severe change in the midgut, leading to irreversible disruption of the complete larval development. In our study, the *in vitro* exposure of *M. entrolobii* to CNSL resulted in up to 90% mortality rate of J2s and inhibition of up to 88% of egg hatching. These results reflected the low LC_{50} values for both J2 mortality and egg hatching inhibition, providing evidence of the high nematicidal potential of CNLS. Interestingly, the suppressive effects recorded in this study outperformed the nematode suppression rates observed in the *C. adansonii* control treatment and those reported on some plants extracts. Sero *et al.* (2025a) observed a maximum of 61.40% *enterolobii* J2 mortality after 72 h of exposure to compost tea produced from *C. adansonii* and 29.80% inhibition of egg hatching after 8 days of exposure. In the

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best cases, Mishra *et al.* (2017) observed over 50% inhibition of *M. incognita* egg hatching after 15 days of exposure to vermicompost tea from agricultural residues, which are lower than that caused by CNLS. At some concentrations (below or equal to 1%), the larvicidal effects on *M. enterolobii* was higher in cold extracted CNSL than in hot extracted, indicating that at medium concentrations, the effectiveness of CNLS is enhanced by the methods of extraction. These findings aligned with previous observations which indicated that the composition of CNSL is determined by the mode of extraction (MAIA *et al*., 2015; Ashong *et al*., 2025). Several authors revealed differences in chemical compositions of CNSL following the extraction methods, with cold extracted CNSL having the highest content of anacardic acid (Patel et *al*., 2006; Rodrigues *et al.*, 2010, Sharma *et al.*, 2020).

In the greenhouse pot experiment, CNLS application at some doses was found to inhibit nematode reproduction better than *C. adansonii*. Recently, Sero *et al.* (2025a) has reported the nematicidal effect of compost tea produced from *C. adansonii* against *M. enterolobii* in *in vitro* bioassay. The present study confirms the superiority of CNSL over *C. adansonii* under naturally nematode-infested environment. In a pot experiment assessing the suppressive effect of compost produced from *Chromolaena odorata* on *M. enterolobii*, Sero et al. (2025b) reported a reproduction factor of 2.91 over 10, while it was 1.15 in our study. Interestingly, this outperformance of CNSL was observed using unsterilized soil infested with mixture species of *Meloidogyne* spp., reflecting the nematode situation in the West African vegetable production systems. These impressive results position CNSL as a promising alternative option for RKN management in vegetable crops.Between the two types, cold-extracted CNSL, particularly at doses 50 ml and 100 ml per pot) proved more effective than hot-extracted CNSL in suppressing nematode reproduction and inhibiting root gall formation. These results reflected those recorded in the *in vitro* bioassays, confirming that the mode of extraction affects the CNSL composition (MAIA et al., 2015), and in this case, the content of anacardic acid (Patel et *al*., 2006; Rodrigues et al., 2010, Sharma *et al.*, 2020).

Adversely, all the growth parameters assessed in this study were negatively affected by CNSL, regardless the types. These results suggest a negative effect of CNSL soil application on tomato plants.Investigation of the phytotoxicity effect of CNSL on the germination and growth of lettuce, tomato and coffee revealed that CNSL negatively affected the germination and vigor of lettuce and tomato, particularly the root development (Matias *et al.*, 2017), reflecting the situation in the present study. In contrast, Sotondji *et al.* (2022) has reported an increase of marketable cabbage yields (average 14 t/ha vs. 1.70 t/ha for untreated plots) following application of cold-extracted CNSL at 15%. However, the CNLS has been applied as foliar treatment, while it has been applied in soil in the current study. Likewise, the assessment of the effects of CNSL on rice in pot cultivation, showed no significant effect on the plant growth and dry matter production following soil application of CNLS at 0.1% (Minamikawa *et al.* 2021). Therefore, the detrimental effect of CNSL observed in our study might be due its application in close environment (pot experiment) at high concentration (10%) with no possibility of leaching. These observations call for further studies assessing various concentrations (dilution rates) and application doses.

Conclusion:-

The present study provides evidence that CNSL has nematicidal properties against *M. enterolobii* juveniles and eggs *in vitro*. Furthermore, this cashew nut by-product demonstrates clearly itsinhibitory effects on RKN reproduction and tomato root galling in soil naturally infested with mixed species of RKNs. It is important to note that cold-extracted CNLS outperforms hot-extract CNSL in suppressing RKNs and the damage they cause to tomato roots. However, at concentration of 10%, CNSL, regardless of the mode of extraction, has negative effect on the development of tomato plants, particularly fresh shoot and root weight. Hence, further investigations are required to identify concentrations suitable to suppress nematode reproduction while ensuring plant growth.

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3. Figures

List of figures and legends

Figure 1: Percentage of dead second-stage juveniles (J2s) of *Meloidogyne enterolobii* at different exposure times following treatment with different concentrations of cold- and hot- extracted cashew nut shell liquid (CNSL)

Legend Figure 1

C1, C2, C3, and C4 represent CNSL concentrations of 10%, 1%, 0.1%, and 0.01%, respectively.

SDW: Sterile Distilled Water (control)

Figure 2: Lethal concentrations (LC_{50}) of cold- and hot- extracted cashew nut shell liquid (CNSL) causing 50% mortality of *M. enterolobii* J2s after 48 hours of exposure

Figure 3: Lethal concentrations (LC50) of cold- and hot- extracted cashew nut shell liquid (CNSL) inhibiting 50% hatching of *M. enterolobii* eggs after 8 days of exposure

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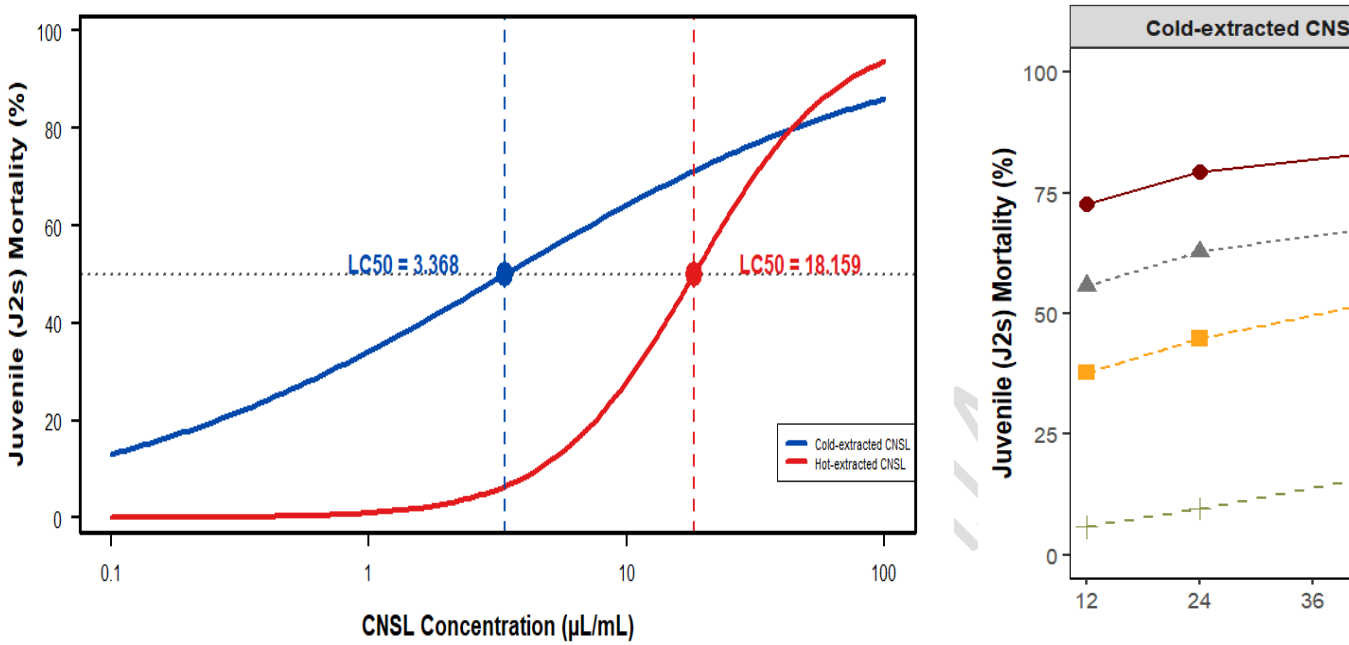

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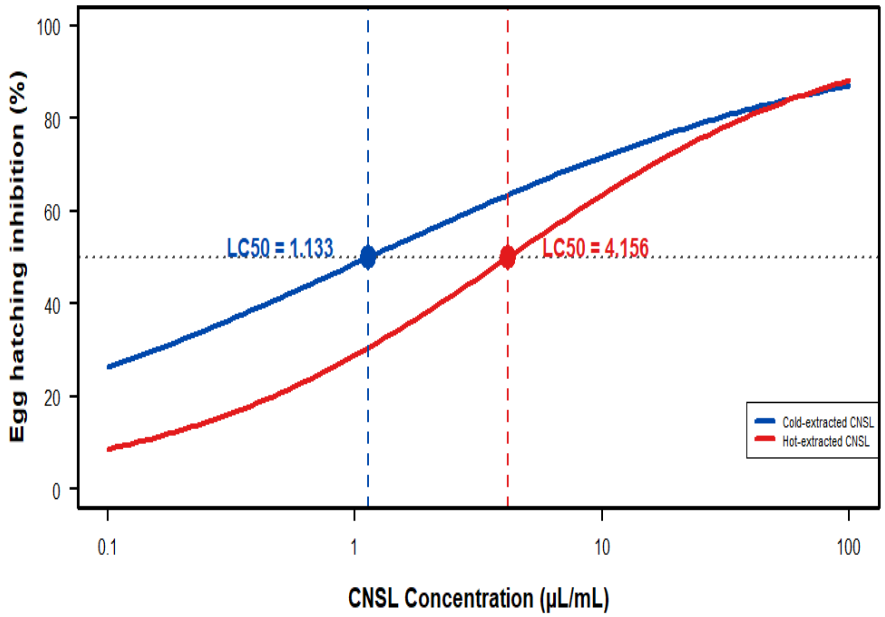

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4- Tables

List of tables and legends

Table 1: Effect of different doses of cold- and hot-extracted cashew nut shell liquid (CNSL) on root-knot nematode reproduction and gall index eight weeks on tomato plants (cv. *Padma 108 F1*) 8 weeks after transplanting in *Meloidogyne* spp. infested soil

Legend Table 1:

Data are means of ten replicates. Values in columns followed by different letters are significantly different ($P \leq 0.05$) based on Fisher's Least Significant Difference (LSD) test using one-way ANOVA. [a]Statistical analysis based on log_{10} (x+1), backtransformed data presented. [b]RF (nematode reproductive factor)= final nematode density (eggs and juveniles) per pot / initial nematode inoculum per pot.[c]Gall index was assessed on a scale of 0-10, where 0 = no galling and 10 = 91-100 % galled roots (Affokpon et al., 2012).

Table 2: Shoot length (SL), stem girth (SG), fresh shoot weight (FSW) and fresh root weight (FRW) of tomato plants (cv. *Padma 108 F1*) 8 weeks after transplanting in *Meloidogyne* spp. infested soil

Legend Table 2

Data are means of ten replicates. Values in columns followed by different letters are significantly different ($P \leq 0.05$) based on Fisher's Least Significant Difference (LSD) test using one-way ANOVA

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584 **Table 1:** Effect of different doses of cold- and hot-extracted cashew nut shell liquid (CNSL) on root-knot
585 nematode reproduction and gall index eight weeks on tomato plants (cv. *Padma 108 F1*) 8 weeks after
586 transplanting in *Meloidogyne* spp. infested soil

Doses of CNSL (ml/pot)	**Final nematode densities per pot[a]**	***RF = Pf/Pi[b]***	**Gall index[c] (0-10)**	**no egg masses/ g of roots[a]**
Cold-extracted CSNL 100	1301 f	1.15 e	1.2 d	4 c
Cold-extracted CNSL 50	3586 d	3.17 d	2.6 c	6 c
Cold-extracted CNSL 25	12094 b	10.69 b	7.6 a	18 ab
Hot-extracted CNSL 100	1642 e	1.45 e	2.6 c	4 c
Hot-extracted CNSL 50	5944 c	5.25 c	5.0 b	17 ab
Hot-extracted CNSL 25	15034 a	13.28 a	7.8 a	22 a
C. adansonii leaf extract (20g/50 ml)	6128 c	5.41 c	5.6 b	10 b
Positive control (nematode alone)	15251 a	13.47 a	8.0 a	26 a
Fisher	***249.4***	***280.5***	***156.5***	***16.12***
P value	***< 0.001***	***< 0.001***	***< 0.001***	***< 0.001***

587 *Data are means of ten replicates. Values in columns followed by different letters are significantly different ($P \leq$*
588 *0.05) based on Fisher's Least Significant Difference (LSD) test using one-way ANOVA. [a]Statistical analysis*
589 *based on log_{10} (x+1), backtransformed data presented. [b]RF (nematode reproductive factor)= final nematode*
590 *density (eggs and juveniles) per pot / initial nematode inoculum per pot.[c]Gall index was assessed on a scale of 0-*
591 *10, where 0 = no galling and 10 = 91-100 % galled roots (Affokpon et al., 2012).*

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608 **Table 2:** Shoot length (SL), stem girth (SG), fresh shoot weight (FSW) and fresh root weight (FRW) of tomato
609 plants (cv. *Padma 108 F1*) 8 weeks after transplanting in *Meloidogyne* spp. infested soil

Doses of CNSL (ml/pot)	**SL (cm)**	**SG (mm)**	**FSW (g)**	**FRW (g)**
Cold-extracted CSNL 100	15.3 d	1.11 d	2.69 f	2.76 e
Cold-extracted CNSL 50	39.5 c	2.97 c	7.50 e	3.30 de
Cold-extracted CNSL 25	51.6 b	4.04 b	13.24 c	7.09 c
Hot-extracted CNSL 100	15.0 d	1.59 d	3.12 f	2.66 e
Hot-extracted CNSL 50	39.9 c	3.03 c	10.62 d	4.67 d
Hot-extracted CNSL 25	52.5 b	5.81 a	14.12 c	6.99 c
C. adansonii leaf extract (20 g/50 ml)	64.4 a	6.26 a	26.77 a	11.80 a
Positive control (nematode alone)	59.1 ab	6.34 a	19.44 b	8.94 b
Fisher	***30.99***	***55.5***	***78.55***	***44.89***
P value	***< 0.001***	***< 0.001***	***< 0.001***	***< 0.001***

610 *Data are means of ten replicates. Values in columns followed by different letters are significantly different (P ≤*
611 *0.05) based on Fisher's Least Significant Difference (LSD) test using one-way ANOVA*
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