

# Hyperspectral Remote Sensing Detection of Root-Knot Nematode Infestation in Mungbean under Various Management Strategies

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In response to the pervasive threat of *Meloidogyne incognita* infestation in mungbean crops, this study explores the application of hyperspectral remote sensing methodology to detect and assess root-knot nematode presence, while investigating the efficacy of diverse management strategies. Spectral remote sensing was used to record the reflectance pattern of the Root-knot nematode (RKN), *M. incognita* infestation on mungbean plants under various management methods under glasshouse pot conditions. Leaf reflectance was measured on plant canopies of mungbean which revealed that reflectance varies with different inoculum levels of root-knot nematode. The spectral reflectance pattern was maximum at the highest inoculum level of 6000 J<sub>2</sub>/ pot with respect to the wavelength of 250–750 nm and minimum at 0 inoculum level or 100 J<sub>2</sub>/ pot corresponding to a wavelength of 800–1600 nm. In response to nematode management, mungbean plant canopy reflectance showed that at a lower wavelength of 350–750 nm, spectral reflectance were mixed and can be determined easily for low or high pattern. However, at wavelengths >800–1600 nm, the lowest and median reflectance was found with carbofuran 3G @ 1 kg a.i/ha and 2 kg a.i/ha, respectively. Similarly, high and low reflectance pattern for *Trichoderma harzianum* was observed at the concentration of 2 and 1% (w/w) respectively but the reflectance pattern was similar for neem seed oil at two different concentrations of 1 and 2% (v/w). The highest reflectance was observed with untreated un-inoculated control. The valuable information on the use of hyperspectral data gathered from the hand-held spectrometer and small unmanned aerial systems for large-scale mapping of the root-knot nematode infestation could be possible. Hence, the effect of *M. incognita* on the root system of mungbean can be non-invasively diagnosed by using hyperspectral data at the early growth stage of aerial plant canopy.

**Keywords:** Inoculum level, Nematode, Spectral reflectance, Spectroradiometer, Wavelength

## Introduction

Mungbean is an important legume crop of the world which is consumed as supplementary diet, particularly in Asian countries. It is good source of natural protein, dietary fiber, minerals, vitamins and substantial quantities of bioactive compounds.<sup>1</sup> *Meloidogyne incognita*, a kind of Root-knot nematode (RKN), is one of the various biotic limitations that influence mungbean production and cause yield losses.<sup>2,3</sup> They are microscopic obligate parasites that feed on plant roots, leading to the formation of root galls, causing interference in transportation of water and nutrients to the top of the plant ultimately resulting in poor plant growth with low yield.<sup>4</sup> The yield losses to mungbean caused by RKN in typical sandy soil ranged from 19–40% under low rainfall regions.<sup>5</sup> Therefore, plant health monitoring and early detection of RKN disease becomes essential to reduce the yield losses caused by these tiny organisms.<sup>6</sup>

Diagnosis of nematode-infected areas is also expensive because accurate population distribution estimates require a large number of soil samples from the field.<sup>7</sup> One way of indirectly determining the infestation caused by nematodes in large areas within a short periods of time is through the use of remote sensing. Leaf symptoms, low mobility of soil-borne nematode and plant-infested clusters make it possible to detect nematodes using orbital and aerial imaging, making it easier to use of imaging applications for precision farming.<sup>8,9</sup> With regard to nematode-induced stress, a number of sensor-based processes have been tested on ground and air platforms.<sup>10–12,8</sup> Many authors used remote sensing devices/sensors in studies assessing asymptomatic and symptomatic plant conditions.<sup>13–17</sup> Among narrow spectrum bands extensively used in agriculture, hyperspectral bands are widely used to identify characteristics of plant cover, which can be characteristic to detect different types of stress factors.<sup>18–20</sup> In a nutshell, hyperspectral remote sensing can be an invaluable tool for early detection of plant diseases.<sup>21,22</sup>

Through reflectance-based remote sensing, "spectral signatures" of plants and other objects can be identified from their surfaces over a range of electromagnetic spectrum wavebands.<sup>23</sup> Reflectance profile of plant leaves is determinant of the chemical composition and physical properties of plant tissues as well as by spectral properties of the remote sensing tool.<sup>24,25</sup> Diseases and insects are examples of biotic stressors that alter a plant's chlorophyll, chemical concentrations, tissue/cell structure, intake of nutrients and water, and gas exchange, which causes variations in crop canopy reflectance.<sup>26</sup> Reflectance spectra are increasingly being used to assess vegetation quality since hyperspectral remote sensing equipment has been developed extensively and offers additional bands in the visible (VIS), near infrared (NIR), and mid-infrared (MIR) ranges. In the MIR region (400–2500 nm), the majority of hyperspectral detectors gather radiation data within 10 nm of visual bandwidths.<sup>27</sup> In a single scan, hyperspectral remote sensing makes use of hundreds of spectral bands to produce more precise spectral data.<sup>28</sup> There is currently little research being done in India on agricultural pest spectral signatures, so attempts have been made to obtain the pattern of reflectance from the canopy of the mungbean plant infested with root-knot nematodes. Its ability to distinguish between different infection loads has also been evaluated. Efforts have also been made to obtain the mungbean leaf canopy spectral reflectance pattern when management strategies are recommended against root-knot nematodes. This can be applied to diagnose nematode problems more accurately in field and pot investigations. Results indicate that remote sensing may make it possible to detect plants infested with nematodes. Therefore, in the present investigation, an attempt has been made to assess the health of mungbean plants when in interaction with RKN by using foliar hyperspectral data.

## Materials and Methods

### Study Area and Materials

Experiment was conducted in green house condition in 6" earthen pots filled with 1 kg of steam sterilized soil. Mungbean cv Pusa vishal was used in the experiment. Root-knot nematode, *M. incognita* pure culture was isolated from the brinjal seedling and the inoculation was made at different range of Second-stage juveniles (J2s) per pot and it was placed close to the base of the mungbean seedling by making a small hole.

Each treatment was replicated six times and the experiment was arranged in Completely Randomized Design (CRD).

### Pot Experiments

Pot experiments for mungbean infested root-knot nematode damage at different inoculum levels (Table 1) along with various management options (Table 2) were carried out at ICAR-Indian Agricultural Research Institute, New Delhi. Five seeds per pot were sown in earthen pots of size 15cm diameter. After germination, they were thinned at three leaf stage to maintain one seedling per pot of almost equal height.

### Spectral Readings and Analysis

Reflectance spectra ranging from 0 to 2500 nm for different levels of RKN damage on mungbean plants were measured using a portable field spectroradiometer as shown in Fig. 1 [FieldSpec, Analytical Spectral Devices (ASD), USA]. Prior to the observation, the instrument was calibrated against solar radiation by means of a spectral on, reference panel. Reflectance spectrum was achieved by comparing radiance of target plants (Mungbean) to that of the spectrum. Instrument was adjusted to

Table 1 — Different inoculum levels of root-knot nematode, *M. incognita* on mungbean per pot

Treatment ID	Treatment Description (Inoculum range of Second-stage juveniles (J2s))
T1	0
T2	100
T3	1000
T4	2000
T5	4000
T6	6000
T7	Untreated

Table 2 — Treatments applied for management of root-knot nematode, *M. incognita* of mungbean per pot

Treatment ID	Treatment Description
T1	Carbofuran 3G 1 kg a.i./ha <sup>-1</sup> + N + R
T2	Carbofuran 3G 2 kg a.i./ha <sup>-1</sup> + N + R
T3	<i>T. harzianum</i> 1 % w/w + N + R
T4	<i>T. harzianum</i> 2 % w/w + N + R
T5	Neem oil 1 % v/w + N + R
T6	Neem oil 2 % v/w + N + R
T7	Untreated inoculated control + N + R
T8	Untreated uninoculated + R
T9	Untreated inoculated + N
T10	Untreated uninoculated

(Abbreviations: N = Nematode and R = Rhizobium)



Fig. 1 — Portable field spectroradiometer

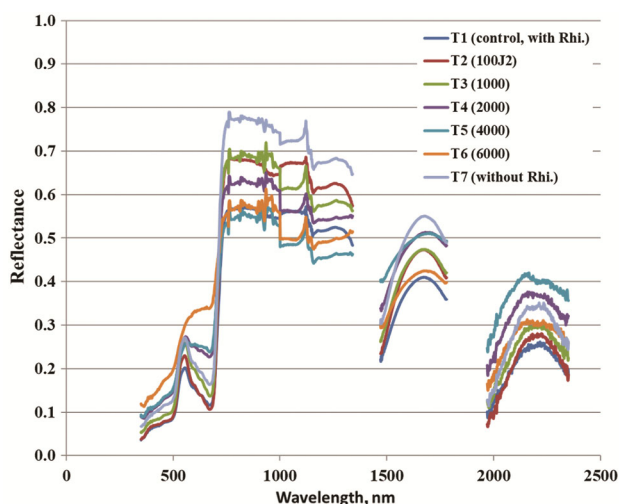


Fig. 2 — Effects of root-knot nematode inoculum levels on the light reflection pattern from mungbean plant leaf canopy (Cv. Pusa Vishal) after 25 days of inoculation.

produce a mean of 50 spectra from one target at a time. All earthen pots were brought under sun prior to observation where exclusion of background shade was ensured. With a 25° field of view, the sensor was held 80 cm above canopy to ensure full cover of plant canopy. Reflectance from mungbean plants was recorded from fixed positions under cloud-free sunlight conditions between 1100 h and 1300 h IST. Mungbean plant reflectance was recorded after 25 days of inoculation and pooled for analysis.

## Results and Discussion

### Spectral Reflectance in Relation to Root-knot Nematode Infestation

The hyperspectral reflectance of the plant cover is affected by the conditions, which follow from the influence of many biotic and abiotic factors unique to the particular ecosystem. That might be a step in the process of gathering ground truth using remote sensing. It has been observed that the reflectance pattern in the plant system is guided by the levels of chlorophyll, water, and minerals as well as overall

turbidity. The aim of this study was to determine the pattern of spectrum reflectance from a mungbean plant's canopy infected with varying levels of root-knot nematode, *M. incognita*, as well as when management options are implied against root-knot nematodes

In the glasshouse pot experiment, reflectance was assessed following a 25-day inoculation period. Spectrophotometric measurements of reflectance on plant canopies, as shown in Fig. 2, revealed that it varied with inoculum levels. We verified the actual signs of nematode infestation in mungbean, and greater damage was noted in the form of galls at inoculum levels between 2000 and 6000 J<sub>2</sub>s, indicating a higher infestation. The spectral reflectance curve, shown in Fig. 2, showed that, generally speaking, uninfected plants exhibited lower reflectance than infested plants, with peak reflectance occurring between 250 and 800 nm. However, infected plants with the maximum amount of inoculums — 6000 J<sub>2</sub>s/pot exhibited a greater reflectance of 0.4–0.5 in the 250–750 nm wavelength, while the lowest reflectance was observed with either zero or 100 J<sub>2</sub>s/pot. The uninfected plants' peak reflectance, which was measured at a greater wavelength of 800–1600 nm, was higher than that of the infected plants (the trend reversed). Under glass house pot studies, reflectance in the 250–800 nm wavelength range was found to be directly correlated with RKN inoculum levels and damage, with reflectance increasing as inoculum levels climbed from 100 to 6000 J<sub>2</sub>s/pot. On the contrary, in the wavelength 800–1600 nm), reflectance decreased with an increase in RKN inoculum levels. As a result, mungbean plant reflectance showed a negative correlation with RKN damage.

Higher mungbean plant reflectance in the VIS correlated with more RKN damage, which indicated a decrease in leaf chlorophyll content as a result of the nematode feeding inside the root tissues, or in vascular bundles. Visual investigations revealed a discernible decrease in chlorophyll, as an RKN-damaged plant had indications of root galls below ground, which often results in leaf wilting, shortening, and yellowing above ground. Being greener than an infected crop, the uninfected crop may be able to absorb more radiation. Prior reports have indicated that nematode-induced biochemical alterations in photosynthetic pigments cause higher plant reflectance in the VIS region.<sup>29–31</sup> Gausman *et al.*<sup>32</sup>

noted variations in light reflectance patterns in cotton leaves grown with a large number population of nematodes (Stressed) vs leaves that are grew with a low/nematode free (*Rotylenchulus reniformis*) population (Unstressed) in the wavelength range of 0.5 to 2.5  $\mu\text{m}$ . In greenhouse and field crops, stressed leaves had a lower reflectance profile than non-stressed leaves of the same age. The primary causes of the variations in the reflectance profiles between stressed and unstressed leaves in the visible (0.5 to 0.75  $\mu\text{m}$ ), near infrared (0.75 to 1.35  $\mu\text{m}$ ), and infrared water uptake (1.35 to 2.5  $\mu\text{m}$ ) regions were the concentration of chlorophyll in the leaves, the mesophyll structure, and the water content, respectively. Similarly, *Rotylenchulus reniformis*, a reniform nematode, has been identified in cotton fields by Palacharla *et al.*<sup>33</sup> using hyperspectral data. Additionally, Hillnhutter *et al.*<sup>34</sup> demonstrated the application of remote sensing for the identification of soil nematodes.

#### Spectral Reflectance in Relation to Infestation under Various Management Strategies

The management experiment was conducted to validate the actual symptoms of nematode infestation. 2000 second stage juveniles ( $J_2$ s) were used as inoculum levels, and the results showed that the infestation was varied with different treatments which could be seen in Fig. 3. At lower wavelengths, 350–750 nm, reflectance is mixed; at higher wavelengths, 800–1600 nm, the results showed a lowest 0.30 and a median 0.40 reflectance, which corresponded to high and low carbofuran treatment concentrations (@2 & 1 kg a.i/ha). We have observed high reflectance of

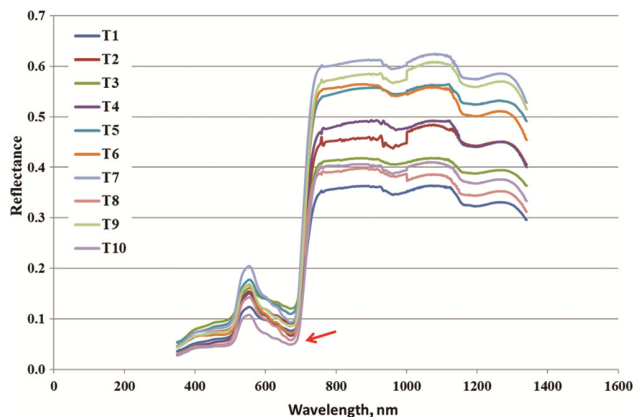


Fig. 3 — Impact of various management strategies on the spectral reflectance pattern of light from mungbean plant leaf canopy (cv. Pusa vishal) (Along with root-knot nematodes)

0.7 and low reflectance of 0.5 with *T. harzianum* treatment (@ 2% & 1% w/w) at high and low concentrations, respectively. Conversely, reflectance for neem oil treatment (@ 1% & 2% v/w) was comparable at 0.50 for both concentrations. The untreated, uninoculated has the highest reflectance (0.64). The results revealed that the spectral reflectance curve of mungbean plant damaged by root-knot nematodes were different from that of the healthy plants. Likewise, Lawrence *et al.*<sup>35</sup> reported management of nematode by means of remote sensing technology, self-organizing maps, and applications of variable-throughput nematicides. The use of remote sensing in soybean nematode, *Heterodera glycines* with infrared reflectance through Landsat was investigated and a 60% change in data was observed.<sup>11</sup> Results indicate that remote sensing may be successful in detecting nematode-infested plants. Both severely infested and asymptomatic potato tubers have been discovered to contain *Meloidogyne luci*. Hyperspectral photography and real-time PCR, a molecular method for detecting nematode DNA, were used to achieve this.<sup>36</sup>

Furthermore, Samiappan *et al.*<sup>37</sup> evaluated the effect of RKN on reflectance of leaves in a controlled environment using a spectrometer. Additionally, to differentiate RKN-infested cotton from the control group, they employed statistically based supervised learning techniques. It was found that the early growth stage of cotton may be non-invasively diagnosed using hyperspectral data after examining the classification accuracy utilizing the visible and near-infrared spectra (350–2500 nm) and partial spectra (350–1000 nm) as a supervised classifier data set.

According to studies by Ramamoorthy *et al.*<sup>38</sup>, Root-Knot Nematode (RKN) stress can be identified ten days after infection. RKN, drought stress (DS), and DS + RKN can be identified with greater than 98% accuracy by utilizing bands that fall between 350–1000 nm and 350–2500 nm. The genotypes "M8" and "Rk-Rn-1" showed different reactions to DS, RKN, and DS + RKN. The characteristics linked to pigments, physiology, and biomass was reduced by all three stressors, with a few noticeable exceptions; the degree of the reduction was higher in "M8" than in "Rk-Rn-1." This research also showed that physiological and hyperspectral responses to RKN and DS may be used to assess plant health issues before outward signs manifested.

Now it is interesting to compare the reflectance pattern for the different kinds of treatments viz. nematicides vs bioagents vs neem products. It is well known fact that nematicides like carbofuran which is systemic in action as far as acting against nematodes while it is also known to interfere in crop growth by boosting the growth by altering physiological parameters like of phenol levels as they are broken down by the microorganisms.<sup>39</sup> Now reflectance is governed by minimizing the nematodes or because of the facts that of physiological attributes. In the same fashion, role of biological agent has multiple actions like reduced nematode count, improved nutrition status and secondary metabolites generation all has bearing on the reflectance. In case of neem oil that has role against nematode development also plays a role for plant growth improvement. Thus reflectance was influenced more by reduction in nematode or by other phenomenon is a further point of investigation. The overall findings point to the possibility of using remote sensing to identify plants that are nematode-infested based on the degree of infestation. It's crucial to pursue it further for the other nematode and crop varieties.

### Conclusions

The findings of this study demonstrated that, in both greenhouse and field conditions, mungbean crop damage induced by root-knot nematodes could be distinguished using the visible and near-infrared (VIS) areas of the electromagnetic spectrum. It would be easier to count the nematodes or use reflectance at wavelengths that are known to be sensitive to the damage produced by RKN when assessing RKN damage in field or pot conditions. The growers may be able to identify RKN infections early and take swift action to combat the pathogen (RKN) by using the hyperspectral remote sensing approach. It is essential to comprehend the reflected spectral reactions of foliage from plants growing under various stress situations, such as salt stress, water stress, nematode infestation, nutrient shortages, insect infestations, and illnesses/diseases. Understanding these reflectance traits could make it easier for remote sensing to identify stressed plants and tell them apart from healthy ones. To improve knowledge of the reflectance produced by stressed plant leaves as a result of various illnesses, such studies ought to be supported.

### Conflict of Interest

The author declares no conflict of interest.

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