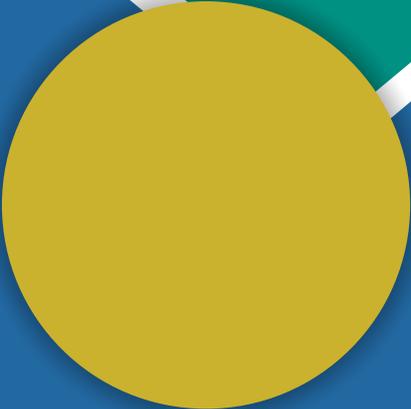


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The subject invention provides a novel control of root knot nematodes through *Pasteuria* root powder. The invention is in powder form which has the properties for the control of root knot nematodes. This root powder is comprising of the acclimatized *Pasteuria* endospores (bacteria) and can be successfully utilized against different species of root knot nematodes.

A systematic survey was conducted to obtain a reliable estimate of nematode and their level of infestation with two vegetables i.e. cucumber, tomato at grower's field located at major vegetable production areas of Faisalabad, Jhang, Khanewal, Lahore, Multan, Rawalpindi and Toba Tek Singh Districts. Root and soil samples were carefully collected from vegetable fields selected at random (twenty samples/crop/District). Samples were put in polythene bags and transported to Nematology Laboratory of Department of Plant Pathology, University of Agriculture, Faisalabad, stored in refrigerator at 5°C (40°F) until processed. Data were recorded on root knot nematodes infestation and presence of any source of indigenous isolates of *P. penetrans* present in the soils of Punjab Province.

During the survey 284 soil and root samples were collected. Out of which 117 (41.19%) were infested with root knot nematodes, while the other samples were devoid of RKN infestation. The incidence of infestation ranged from 30 to 52.38% in sampled areas. Maximum incidence was recorded in cucumber as compared to tomato crop ranging from 30-52.38% and 30-45%, respectively. The root and rhizosphere soil analysis also demonstrated the presence of five other plant parasitic nematode genera, *Criconema*, *Helicotylenchus*, *Hoplolaimus*, *Pratylenchus* and *Xiphinema*. Their root and soil population was very low and these nematode genera are of minor importance.

Identification of *Pasteuria* from roots of infected plants was done by isolating and crushing the female body under a cover slip in water and making observation of the nematode body contents at magnifications between 250 – 400X, while from soil identification was carried out by extracting nematode from soil and observed for juveniles encumbered with spores. The results revealed that indigenous isolate of *Pasteuria* were not found during the survey of cucumber and tomato.

In-vitro trial was conducted on 117 populations of root knot nematodes associated with two vegetables from seven Districts of Punjab. Different root knot nematode populations were maintained on susceptible tomato throughout the experiments. A suspension of 50 J2s in 1 mL of water was pipetted to Petri dish. Two suspensions of *Pasteuria* endospores (PP-3 and PP-Japan) were prepared in water (10^4 and 10^5 spores per ml) using Haemocytometer. One ml of the spore suspension was pipetted to each Petri dish containing one ml of nematode suspension and incubated at $30 \pm 2^\circ\text{C}$ in an incubator.

The Petri dishes were checked after 45 minutes of exposure time and the number of spores attached/J2s on first 10 nematodes was counted under the microscope. The results revealed that the adhesion suitable for reproduction of the *Pasteuria* increased by increasing spore densities of both isolates (PP-3 and PP-J). Maximum attachment was at $32 \pm 2^\circ\text{C}$ as compared to the ambient temperature. Also the response of *Pasteuria* isolates (PP-3 and PP-J) was different with different RKN populations. PP-J showed maximum attachment with RKN populations either tomato or cucumber as compared to PP-3.

From these results fourteen (14) RKN populations were selected on the basis of suitable adhesion to *Pasteuria penetrans* (seven from each host). Culture of these 14 populations was maintained in greenhouse throughout the study. Encumbered juveniles of these populations with PP-3 and PP-J were inoculated to three weeks old tomato plants transplanted in 15 cm Dia. earthen pots. These plants were allowed to grow for 35-40 days and data were recorded on nematode reproduction variables. It was observed that PP-3 produced maximum numbers on infected females, number of *Pasteuria* endospore /female and endospores/ gram root powder as compared to PP-J.

Shelf life of *Pasteuria* (locally produced and exotic) was assessed on quarterly basis for one year. The results revealed that the *Pasteuria* which was produced locally has significantly greater encumbrance rate with J2s, effect on killing, controlling/infecting nematodes, spores/female and spores/gram root powder. These results proved that *Pasteuria* produced on the local population of RKN is acclimatized in the environment of Punjab Province. *Pasteuria* endospores are not damaged by drying. Therefore, they can be stored for long periods at room temperature.

