

NEW TECHNOLOGY TO DETECT LIVING PINEWOOD NEMATODE IN WOOD

By Isabel Leal, Natural Resources Canada, Pacific Forestry Centre

Canada is the fourth largest forest products exporter in the world, in 2016 exports increased by 5.3% to \$34.4 billion. International trade in wood products brings the risk of the movement of tree pests, which can cause ecosystem and economic damage.

Pinewood nematode (PWN), the causal agent of pine wilt disease, has resulted in pine tree mortality in countries such as Japan, Korea, Taiwan, China, Portugal and Spain. The PWN nematode is native to North America pine tree species, but pine wilt disease is not present in Canadian forests. However, trade in pine wood commodities such as logs, untreated wood products, and wood packaging material are potential pathways for the international spread of PWN when its vector, pine sawyer beetles, are also present.



A sawyer beetle transmits the microscopic nematode while feeding on the pine shoots.

L.D. Dwinell, USDA Forest Service, Bugwood.org

This disease is of such concern in 1993, the European Union imposed a ban on untreated lumber to prevent the introduction of PWN into Europe. Most Canadian lumber exports are heat-treated (HT) or kiln-dried (KD), which are treatments that kill PWN.

DETECTION OF PINEWOOD NEMATODE

PINEWOOD NEMATODE

NRCan scientists have developed a method for detecting the presence of living nematodes in wood assuring international importers of the safety of Canadian forest products.

These nematodes are microscopic worms, which are transmitted by sawyer beetles, a wood-boring bark beetle, through their feeding wounds into the resin canals. Here, the nematode will reproduce and hinder the movement of water through water-conducting tissues, eventually causing the tree to die.

To comply with international regulations and prevent movement of PWN to other countries, effective detection is required. Traditionally, detection of PWN involves morphological analysis of species-specific characteristics, after the nematodes are extracted from wood that has been soaked in water. However, this analysis is time-consuming and requires specialized expertise. In addition, the differentiation of PWN from other related species is problematic when only juvenile stages are present. To address these problems several molecular diagnostic methods have been developed that are accurate and faster than traditional methods.

Because only living organisms are regulated as quarantine concerns, molecular tools able to distinguish between living and non-living PWN present in wood are preferable for questions of quarantine regulation in international wood trade.

NEW DIAGNOSTIC METHOD TO DETECT LIVING PWN

Most molecular methods to detect PWN rely on the presence of DNA, a molecule, which can be extracted from both living and dead nematodes. Since only living PWN are a quarantine problem, the presence of false positives from DNA-based methods may result in rejection of safe wood products.



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Propagative pine wood nematode

Photo: Dr. SUN Jiang-hua, Institute of Zoology, Beijing

To address this problem, NRCan designed a molecular method that detects the presence of living PWN in wood, based on RNA, which serves as a viability marker, because it is present in living cells and degrades in dead cells. This RNA-based method reflects the presence of living, disease carrying nematodes.

The ability to quickly and efficiently detect the presence of living PWN using this molecular method will prove useful in disputes involving the detection of PWN, by clarifying whether positive test results originate from live or dead organisms. This will minimize or prevent unnecessary trade restrictions of wood products, thus protecting the access to markets of Canadian wood products from PWN infested areas.

This method is also an effective tool to evaluate the efficiency and success of various phytosanitary wood treatments on PWN survival, such as heat treatment, radiofrequency and microwaves, and could also be used operationally to independently validate kiln efficacy.



Dr. Isabel Leal's technique is providing Canada with a powerful method of certifying that our forestry products are free of live nematodes.

AN INTERNATIONAL STANDARD FOR A PWN DIAGNOSTIC PROTOCOL

The International Plant Protection Convention (IPPC), a United Nations organization dedicated to protecting the world's plant resources from pests and pathogens, recently adopted a globally standardized diagnostic protocol for pine wood nematode, based in part on the research conducted at the Pacific Forestry Centre.



A lumber mill in British Columbia



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Her work involves the development of molecular detection assays specific for various invasive species of phytosanitary concern.

She participated as a member of the drafting group for the IPPC diagnostic protocol for PWN.