Department
for Environment
Food & Rural Affairs

Regulation proposal for tobacco ringspot virus (*Nepovirus nicotianae*) on *Glycine max* (soybean) - seed of oil and fibre plants

November 2024

Objective

To review the status of tobacco ringspot virus in GB legislation

Assessment

The following is a summary of an assessment undertaken by Defra following the method outlined by EPPO (European and Mediterranean Plant Protection Organisation) (Picard *et al*., 2017).

Regulated non-quarantine pest (RNQP) assessment for Great Britain: tobacco ringspot virus (*Nepovirus nicotianae*) on *Glycine max* (soybean) - seed of oil and fibre plants

Background

Tobacco ringspot virus (also known as *Nepovirus nicotianae* or TRSV) is currently a Quarantine Pest (QP) for GB (Great Britain). Available evidence suggests that this pest is present in GB and it is not under official control. As such, TRSV does not meet the requirement for QP status. Assessments were therefore undertaken to see if this pest could become an RNQP (Regulated Non-Quarantine Pest) and if so, which hosts should be listed under the regulations. TRSV has a scattered worldwide distribution, with most impacts occurring in North America where the nematode vectors are widespread.

Current listing of pest in GB legislation

Quarantine Pest (Annex 2, Part A)

Current regulated plants for planting – host plants

None

Taxonomy

Pest name

Nepovirus nicotianae; tobacco ringspot virus; TRSV

Will the pest be listed at species level?

Yes

Status in GB

Is this pest present in GB?

Yes: There is a long history of TRSV causing symptomless infection of *Pelargonium* (geranium) stocks in the UK, with unpublished records beginning in 1979 and the most recent survey being from 2003 (Defra, unpublished data). The results of the most recent survey did indicate that levels of viral contamination had dropped, but there is no evidence that TRSV has ever been fully eradicated from *Pelargonium* (especially since the virus can be transmitted via seed and pollen in *Pelargonium*, Scarborough & Smith, 1977).

Pathways

Are the listed plants for planting the main pathway for the "pest/host/intended use" combination?

Yes

TRSV can spread over longer distances via seeds, by grafting of woody hosts, by vegetative propagation of herbaceous hosts, and via adherent soil containing viruliferous nematode vectors and/or infected seeds (EPPO, 2022).

Seed: Most research on seedborne TRSV has been carried on soybeans, in which seed infection levels of 100% have been recorded. Studies have showed that the virus can be present in the embryo and perisperm, but not the seed coat. Electron microscopic studies have also detected the virus in the inner layer of the pollen wall and the generative cell, in the walls and cells of the embryo sac, integuments and nucellus. Infection reduced pollen germination and slowed germ tube growth, thus impairing its ability to fertilize and transmit the virus. Transmission of the virus seemed to be dependent on infection of the megagametophyte from the mother plant which would have to occur before or at flowering. This conclusion was supported by the finding that higher levels of seed infection occurred in plants infected before, rather than after, flowering. The percentage of infected seeds and the rate of transmission did not change after storage for 5 years either at room temperature or at 1-2°C (CABI, 2022 and references therein).

Pollen: Pollen transmission of TRSV has been reported but data are limited. A study from 2007 reported pollen transmission in soybean, although other scientific evidence is lacking. In soybean, infected pollen have been shown to impair fertilisation, thereby eliminating virus transmission (EPPO, 2022 and references therein).

Vectors: TRSV is spread by nematodes in the *Xiphinema americanum senso lato* complex. *Xiphinema americanum* is considered the main vector in the virus' native distribution (EPPO, 2022; CABI, 2021). Transmission of TRSV in soybean has also been reported for aphids, a beetle (*Epitrix*), a grasshopper, thrips spp. (notably *Thrips tabaci*) and a spider mite. Generally, the transmission efficiency of these vectors was low in laboratory testing and their significance in the field unclear. These studies date from the 1950s to 1980s (Tolin & Polston, 1978; EPPO, 2022; more detail in CABI, 2022). Additionally, TRSV has been reported to systemically spread and propagate within European honeybees (*Apis melifera*). Since these results have been debated, the relevance of this insect species as a vector remains unclear. It should be noted that the ability of TRSV to be transmitted by both nematodes and aerial vectors would constitute an exception among nepoviruses.

In GB, the nematode vectors of TRSV are not known to occur, though the rapid PRA for these nematodes (Fera, 2014 unpublished) acknowledged that some populations may have been inadvertently imported in large, containerised plants. If nematode vectors were to enter GB, they are very likely to be able to establish both outdoors and in protected conditions (Defra, 2018). Arthropod vectors may be present in GB, but as their efficacy as vectors is unclear, it is assumed that the transmission of TRSV from ornamental plants (*Pelargonium*) to soybean crops via these vectors is unlikely.

Imported seeds are therefore considered to be the main means of spread of TRSV on *Glycine max*.

Economic Impact

Are there documented reports of any economic impact on the host?

Yes.

The main sources of infection within crops in the USA is still unclear and sources of information were often found to contradict each other on the importance of seed transmission, nematode transmission, and transmission by immature *Thrips tabaci*. Information on impacts was also dated or not well referenced. Currently, impacts are assumed to be low, due to lack of reports, perhaps because more resistant varieties are in use.

Symptoms:

Severe stunting, curvature of the terminal bud, necrosis of the other buds and pod development reduction or abortion (CABI, 2022 and references therein).

TRSV causes bud blight, shepherd's crooking, stunting, wilting, delayed maturity, bud and lead proliferation, reduced pod number, and discoloured, distorted, and dead leaves. Discoloured pith, petiole distortion and discolouration, leaf vein discolouration, and poor root nodulation (Crop Protection Network, 2019). It also causes root nodulation (Orellana *et al*., 1978).

Symptoms vary according to cultivar of soybean and strain of TRSV (Tu, 1986).

Impacts:

TRSV causes a severe disease (bud blight) of soybean in most of the Midwest soybean-growing areas of the US. Although widespread, the incidence of the disease fluctuates greatly from year to year (Bergeson *et al*., 1964)

From Tolin & Polston (1978): "*Though TRSV occurs less frequently than other viruses in soybean, it causes greater damage when it does occur. Reductions in yield are due to a decrease in the number of pods reaching maturity, the number of seeds per pod, and the quality of the seed. Severe widespread infections have occurred in the Midwest, but only isolated infections have been reported in Virginia. A 90% reduction in yield has been observed in diseased experimental soybeans in Virginia. TRSV causes a major disease of soybean in the Midwest*."

Of the many diseases caused by TRSV, bud blight of soybean is the most severe and causes the greatest losses. Yields may be reduced by 25 to 100%. In general, losses are greatest when young plants are infected (before flowering) or when seeds with a high percentage of TRSV are sown. Yields also are lowered through reduced pod set and seed formation on infected plants (University of Illinois, 2017).

When healthy and infected seed was mixed, no significant yield differences occurred until 50% infected seed was in the mixture, resulting in over 30% infected plants. Below this, the lessened competition enabled uninfected plants to produce more seed, compensating for the loss from infected plants (Athow, 1961).

TRSV can be common in fields and yet yield losses generally insignificant, as is the case in North Carolina (NC State, 2020).

Bud blight is a cyclic disease. Athow (inaccessible paper) reported high losses of soybean in 1943-1947 and low losses from 1955-57 (Mundt, 1973).

What is the likely economic impact of the pest irrespective of its infestation source in the absence of phytosanitary measures?

Minor impacts are expected on soybean with high uncertainty.

The UK rapid Pest Risk Analysis for TRSV (Defra, 2018) concluded that, on all hosts, the potential economic impacts would be small with high confidence, and they were expected to be largely limited to ornamentals (similar to impacts seen in the past in the UK and EU countries).

Symptoms can be severe in soybean in some circumstances, reducing pods and yields which would reduce market price for human consumption. Infected crops could still be used as fodder, but the reduced number of beans might also affect price for this market. Fast means of spread (via aerial vectors) are not well documented, therefore how easily the virus could spread to weeds in field margins (which could act as a reservoir for the virus) and become established in the long-term is not clear.

One other key uncertainty is whether the GB specific varieties of soybean have virus resistance. Varieties have been bred for early planting (to suit our climate) and for harvestability. The largest supplier in GB '*always aim[s] to produce seed in the UK, to ensure quality at all times*' (Soya UK, no date).

Is the economic impact due to the presence of the pest on the named host plant for planting, acceptable to the propagation and end user sectors concerned?

No

Risk Management Measures

Are there feasible and effective measures available to prevent the presence of the pest on the plants for planting at an incidence above a certain threshold (including zero) to avoid an unacceptable economic impact as regards the relevant host plants?

Field inspections can be used to detect TRSV infections in soybeans, since these infections generally show clear symptoms (EPPO, 2022).

EPPO PM 7/2 (2) *Tobacco ringspot virus* states that testing is easiest in the soft tissue of the plant. Therefore, germinated seeds need to be tested (EPPO, 2017). CABI (2022) however, reference a paper where TRSV was detected in soybean seed using RT-PCR that had been combined with a gold immunochromatography assay (GICA) (Zhang *et al*., 2009 – paper not accessed, see references).

None of the following treatments inactivated the virus in soybean seed: hot water at 56°C for 10 min, hot air at 52°C and 56°C, or aerated steam at 60°C (CABI, 2022 and references therein).

Data Quality

Is the quality of the data sufficient to recommend the pest to be listed as an RNQP?

Yes, though there is some uncertainty as to the potential impacts of TRSV on soybean.

Proposal for regulation

We propose to remove TRSV from the QP list and instead regulate TRSV as an RNQP on seed of *Glycine max*, by adding it to Annex 4, Part G, of the Phytosanitary Conditions Regulation[1]. As a result, this seed would need to be free from TRSV to be imported into, or moved within, Great Britain.

[1] Commission Implementing Regulation (EU) 2019/2072 of 28 November 2019 establishing uniform conditions for the implementation of Regulation (EU) 2016/2031 of the European Parliament and the Council, as regards protective measures against pests of plants, and repealing Commission Regulation (EC) No 690/2008 and amending Commission Implementing Regulation (EU) 2018/2019

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This regulation proposal has been undertaken taking into account the environmental principles laid out in the Environment Act 2021. Of particular relevance are:

- The prevention principle, which means that any policy on action taken, or not taken should aim to prevent environmental harm.
- The precautionary principle, which assists the decision-making process where there is a lack of scientific certainty.