



PLANT NEMATODE SAMPLING KIT

This kit provides information to assist horticultural growers including home gardeners to collect soil/plant samples in order to test for nematodes.

Sample kit contents

- Service information sheet and basic fees
- Guide to soil and crop sampling for nematodes
- Specimen submission form

Tests Available

- Tests for nematodes in soil and/or horticultural plant tissues
- By various extraction methods
- Two basic tests: quantitative soil and quantitative root tests
- Read the instruction sheet before collecting samples
- Select tests required on the specimen form
- Charges apply per sample per test per extraction method
- For more than 10 samples, a bulk discount is negotiable

Taking a sample

- Use the sampling instructions as a guide
- Take samples in general from the root zone of the crop
- With separate samples for healthy and diseased areas
- Sample moist soil before crop removal rather than from fallow
- Place each sample in a clearly labelled plastic bag
- Do not use cloth or paper bags
- Do not put sample forms inside bags containing soil
- Protect samples from heat and drying out
- Samples are best fresh: collect & send early in the week
- Plant samples are best refrigerated
- Pre-booking tests is recommended

Where to send samples

The client is responsible for costs of transporting samples to the laboratory.
Send samples promptly after collecting to:

Greg Walker, SARDI Plant Research Centre

Courier delivery: Waite Precinct, Gate 2b, Level 2
Hartley Grove, Urrbrae 5064
Postal addresses: GPO BOX 397, Adelaide SA 5001
or Locked bag 100, Glen Osmond SA 5064

Contact information: ph. 08-8303 9355; fax 08-8303 9393; email: Greg.Walker3@sa.gov.au

Charges

Note that each test is charged separately for each sample; tick the required sections on the specimen submission form to indicate which tests you require.

a) For all **quantitative soil and root tests**, including basic soil and root tests and special tests such as Citrus Nematode adult female root test (staining), stem/bulb test, leaf test and centrifugal flotation test (Stubby-root Nematode etc.):

\$103.40 per sample (inc. GST) for the first one to two samples per consignment

\$89.10 per sample (inc. GST) for the third and succeeding samples per consignment
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b) For a **qualitative plant test** (visual examination only of roots, tubers etc.):

\$41.25 per sample (inc. GST) for the first one to two samples per consignment (a sample may contain up to 5 plants or tubers)
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\$36.30 per sample (inc. GST) for the third and succeeding samples per consignment
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Quotes for larger numbers (10 or more) of samples or discounts for bulk users are available.

Other tests (bioassays using bait plants, gall index rating, Root-knot Nematode egg mass quantitative root test (staining), Root-knot Nematode egg quantitative root test (hypochlorite extraction), cultivar resistance testing, nematicide testing, or property visits to undertake sampling) by negotiation. Inquiries regarding collaboration and contract research from companies developing nematicidal chemicals, biocontrol products or resistant cultivars are welcome.

You will be invoiced for the cost of these tests – please include full client details including Australian Business Number (ABN) on the attached specimen form.

Quarantine restrictions (samples collected interstate)

Soil/ plant samples collected interstate need quarantine clearance: entry is prohibited if collected from areas where regulated pests/diseases such as Phylloxera, Potato Cyst Nematode, Green Snail or *Fusarium oxysporum* race 3 are known to occur. These restrictions protect our valuable agricultural industries, and breaches are subject to substantial penalties. Documentation is required by law; declaration forms/plant health certificates require time to organize, so plan ahead. Samples arriving without quarantine clearance/documentation are liable to be destroyed. Inquiries can be made to: State Quarantine Service, 46 Prospect Rd, Prospect SA 5082, phone 1300 666 010, fax 08-8344 6033. If you suspect an outbreak of an exotic pest or disease notify your quarantine authority.

Suspected outbreaks of quarantine-status pests/diseases

SARDI strongly encourages reporting of suspected outbreaks of exotic or quarantine-status pests and diseases; the earlier an outbreak is detected the greater the likelihood of successful containment and eradication. If a pest is allowed time to establish it may become impossible or impractical to eradicate: early reporting is in the interests of all agricultural industries. If tests confirm the presence of quarantine-status pests/diseases, re-imbusement of diagnostic charges if levied may be made once positive identification has occurred.

If you see unusual pests or diseases on plants, report it to the exotic plant pest hotline on: 1800 084 881. Visit www.planthealthaustralia.com.au for further information.

The information in this publication can be provided on request in an alternative format or another language for those who need it.

A GUIDE TO SOIL AND CROP SAMPLING FOR NEMATODES

by Greg Walker, SARDI, Plant Research Centre

Plant and Soil Health, Waite Precinct, Hartley Grove, Urrbrae 5064

(Phone: 08-8303 9355; Fax: 08-8303 9393; Email: Greg.Walker3@sa.gov.au)

WHY TEST FOR NEMATODES?

Plant parasitic nematodes (or eelworms) are microscopic worms found in soil which feed on plants, reducing crop growth and yields. They commonly attack the roots producing symptoms of general root insufficiency: unthrifty growth, leaf yellowing and wilting. All horticultural crops are attacked by one or more nematodes but particular nematodes attack only certain kinds of crops, and crop varieties can vary from highly susceptible to highly resistant. Root-knot Nematode is one type of nematode which attacks a wide range of crops including vegetables, grapevines, fruit and nut trees, ornamentals, and some field crops, plus many weeds. Plant parasitic nematodes cause estimated losses to world agriculture of \$127 billion per year. Other nematodes attacking horticultural crops include Citrus, Cyst, Dagger, Pin, Ring, Root-lesion, Spiral, Stem and Bulb, Stubby-root, and Stunt Nematodes. Losses from nematodes are often underestimated because they are hidden in the soil. Testing for nematodes is needed to diagnose nematode-related problems, and is also useful before planting to help decide whether resistant varieties or rootstocks, soil fumigation or treatment with a nematicide may be beneficial.

THE NEMATODE TEST

Soil and/or plant samples are tested for nematodes at the Plant Research Centre. The living nematodes are extracted from the samples over a period of up to five days. The plant parasitic nematodes are then counted using a microscope, and the results are sent to the grower. Some nematodes occur mainly in the soil around roots, while others occur both in the soil and on or inside roots, so it is best to test both soil and roots. For quantitative tests, indicate on the specimen form whether you also want a count (usually in soil) of the total free-living (or saprophytic) nematodes. These nematodes do not damage plants and are important in nutrient recycling and in interactions with other soil animals; some growers want an estimate of their numbers as an indication of overall soil health.

A qualitative test (visual examination) can be made of roots etc. to determine the presence of galls or nematode egg masses, females or cysts. Quantitative soil or root tests use different extraction methods to determine numbers of nematodes per unit (fresh or dry) weight or volume of soil or roots. If you want a basic quantitative soil or root test select the options marked 'Quantitative soil test' or 'Quantitative root test' respectively on the specimen sheet. Quantitative tests can also be used on stem, bulb, tuber or leaf tissue for nematodes that infect these parts of plants and there are specific extraction methods that are specific for particular stages of specific nematodes (e.g. staining test for adult female Citrus Nematodes on roots) or groups of nematodes (e.g. centrifugal flotation for less motile nematodes including Stubby-root Nematodes). For Citrus Nematode, the basic soil and root tests detect the worm-like motile stages (which are more sensitive to changes in soil moisture and temperature) whereas the staining test detects sedentary females embedded in the roots (note that the latter stage may still be detected after application of a nematicide, even though they may be dead, until such time as breakdown has occurred). If you plan to re-sample at a later date or compare different areas, soil measurement on a volumetric or equivalent dry weight can reduce the effects of variation in soil moisture.

WHEN TO SAMPLE

Growers should take soil samples regularly for nematode testing, particularly before planting and at harvest, and should keep records of population levels in different blocks to assess the effectiveness of management programs. Nematodes are often found in highest numbers in the root zone of the crop towards the end of the growing season or in late summer to mid-winter. For Root-knot Nematode on grapevines, juveniles were found in one study to peak in soil in winter, and then to decline in spring as they entered roots. Numbers decline in weed-free, fallow ground. Soil should preferably be moist but not wet at the time of sampling; nematode detection is most difficult in dry, fallow soil. Serial samples are best collected at an equivalent stage in the irrigation cycle or when soil moisture levels are similar.

Before planting, sample after cultivation but allowing sufficient time (at least 2-4 weeks) to both test the samples and to make arrangements for soil treatment etc. To determine the effectiveness of treatment, soil can be re-sampled 2-3 weeks after fumigation or 4-6 weeks after application of a non-fumigant nematicide (e.g. NemaCur^(R)). A repeat sampling at mid-season may further help to determine the effectiveness of a nematicide. It is a good idea to take another sample at harvest or at the end of the

growing season to determine the likely carry-over into the next crop and to help plan rotations including use of break crops, resistant varieties and fallows.

Where diagnosis is required for unthrifty growth, plant and soil samples may be collected at any time during the growing season. Samples should be collected from near the edge of declining patches and, for comparison, from areas of healthy plants. Plants that have already died are best avoided since nematode populations in their vicinity may already have declined. It is far easier to detect nematodes before ripping out a crop than afterwards in dry, fallow soil so plan ahead if possible.

Whole plants, roots or tubers should be included to assist diagnosis.

HOW TO SAMPLE

Nematodes are usually unevenly distributed in a field and soil sampling procedures need to take this into account to obtain a truly representative sample. Very low or patchy populations are difficult to detect. The reliability of nematode counts is only as good as the sampling procedure used. Particular attention should be paid to the following:

Sampling Tools

A soil tube or auger is useful for taking many sub-samples (cores) while reducing the total amount of soil collected. A spade can be used if most of the soil is removed by hand from along the vertical edge of the blade, and is useful for collecting roots and associated, relatively undisturbed soil. Whatever tool is used, it should be cleaned before sampling a separate plot or field, and soil disturbance should be minimised to avoid abrasion damage to the nematodes.

Depth of sampling

Nematodes are most abundant within the root zone and, for shallow-rooted crops, samples can be taken to a depth of about 20 cm, but few nematodes are present in the first few cm of dry, sun-exposed soil so this can be omitted. Some nematodes are more abundant at greater depth in fallow soils during summer, and a second sample may be taken pre-planting from depth 20 to 40 cm or more, especially where deep-rooted crops, e.g. trees, vines, lucerne, were previously growing. In established crops, sample in the row from the root zone, near the plant stems or inside the dripline, collecting both soil and roots. In grapevines, a sampling depth of 0 to 30 cm is often used but some species reach highest populations at 30 to 60 cm in deep soils.

Number of sub-samples/samples

For plots up to 100 square metres in area, at least 20-30 sub-samples (cores) should be taken per sample. For uniform areas up to 1 ha, at least 50 sub-samples should be taken. Sub-samples should be collected in a bucket or bag while walking up and down the field or orchard; various sampling schemes such as collecting a sub-sample at regular intervals along a “W” or “zig-zag pattern” have been used. Sub-samples that are too large make it difficult to obtain a representative composite sample of manageable size. Fields should be divided into areas of 1 ha or less for sampling and separate samples should be taken from areas with different cropping histories, soil types/treatments applied, or crop health.

Handling and storage of samples

With samples, fresher is better. Soil sub-samples should be gently mixed before placing at least 500 g in a plastic bag; do not use cloth or paper bags. For a root test at least 20 g of fine feeder roots will be needed, especially include roots showing symptoms such as galling, thickening, bunching, soil adherence etc. Seal bags to prevent drying and protect samples from heat; perishable plant samples especially need to be sent promptly &/or be refrigerated. Soil does not need to be refrigerated unless liable to be exposed to high temperatures or if it cannot be despatched promptly. Label the bag as appropriate and complete a *Specimen Form* with the test required, block/sample numbers, depth, crop type, name, address, phone/fax number etc. Do not put paper labels inside the bags with the soil. **Send the samples promptly to Greg Walker (SARDI Plant Research Centre, Gate 2b, Level 2, Waite Precinct, Hartley Grove, Urrbrae 5064 [Locked Bag 100, Glen Osmond SA 5064]).**

To ensure samples are promptly processed, pre-book tests and preferably collect and despatch samples early in the week.

SPECIMEN FORM

NEMATOLOGY DIAGNOSTIC SERVICE HORTICULTURE PATHOLOGY

PLANT RESEARCH CENTRE
Waite Precinct, Gate 2b, Level 2
Hartley Grove, Urrbrae 5064

Attention: Greg Walker

Telephone: (08) 8303 9355/8303 9497

Fax: (08) 8303 9393

Email: Greg.Walker3@sa.gov.au

ID NO.:

Laboratory use only.

SARDI



SOUTH AUSTRALIAN
RESEARCH AND
DEVELOPMENT
INSTITUTE

CHARGE TO EITHER: Client.....or submittee.....(TICK OPTIONS AS REQUIRED)

REPORT TO: Client.....or submittee.....

REPORT BY: Mail.....Fax.....Email.....Phone.....

SUBMITTED BY: **Date sent:**...../...../..... [Date received:...../...../.....]

Name & ABN:.....

Location/Address:.....

Telephone: ()..... **Fax:** ()..... **Email:**.....

CLIENT INFORMATION (if different from above):

Name:.....

Business Name & ABN:.....

Postal Address:.....

Suburb/Town:..... **State:**..... **Postcode:**.....

Telephone: ()..... **Fax:** ()..... **Email:**.....

Location of property (if other than postal address):.....

SAMPLE DETAILS FOR NEMATODE TEST (Label bag clearly): (If insufficient space use back of form)

Plant/crop type:..... **Scion/rootstock cultivar:**.....

Cropping history:

Symptoms:.....

Purpose of test: Diagnosis.....Monitoring population.....Preplanting.....Chemical efficacy.....

Test requested (each charged separately per sample): Visual examination of roots/tubers etc. only.....

Quantitative soil test.....Quantitative root test.....Citrus Nematode adult female root test (staining)

.....Quantitative stem/bulb test.....Leaf test.....Centrifugal flotation (Stubby-root Nematode

etc.)Other (specify) **Bag labelled**.....

For quantitative tests: Plant parasitic nematodes only.....or total free-living nematodes too.....

Preferred soil measurement: By fresh weight.....By dry weight.....By volume.....

Collected by: Spade.....Auger.....Other.....At depths:mm tomm

No. of cores/sub-samples collected.....From area of:ha or.....m²

Soil texture (e.g. sandy loam):Soil pH:Last irrigated on:/...../.....

Too busy for above details?: give me the basic soil test.....&/or root test.....No. samples.....

Nematicides/fumigants applied:On date(s):

