



PETIOLE SAP NITRATE-N QUICK TEST FOR DETERMINING NITROGEN STATUS OF TOMATO

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Introduction

Most vegetable farmers use nitrogen fertilizers to increase crop yield and quality (Aldrich 1980; Haskell 1925; Zhao-Hui et al. 2008). However, applying more nitrogen than the crop can take up increases production costs and may contribute to environmental pollution. To meet the crop's need for optimal productivity and to minimize fertilizer cost and loss, plant nitrogen status needs to be evaluated and fertilizer application rate needs to be calculated accurately (Lang et al. 1999).

A soil test does not always accurately reveal the nitrogen status of a crop. Nitrogen is a mobile element and is readily moved down into the soil by irrigation or precipitation (Aldrich 1980; Bevacqua and Cardenas 2002; Burt et al. 1995). Nitrogen can also be tied up by soil microorganisms so that it is not available for crop uptake. Testing nitrogen status in the plant rather than soil is a more reliable indicator of the crop's nitrogen status. The nitrogen level in fresh petiole sap is a very sensitive indicator of the current crop's nitrogen status (Hochmuth 1994) and is especially useful for scheduling nitrogen fertilizer application.

Fresh petiole sap can be measured by a few different methods, such as the colorimetric method using a meter (HACH; Loveland, CO), the nitrate-N test using test strips (Merckoquant, Merck; Darmstadt, Germany), or a Cardy meter (Horiba Scientific; Edison, New Jersey). The meters or test strips for these methods are available in the market and are affordable; however, for the first two methods, it is necessary to dilute the fresh petiole sap with distilled water (1:19 ratio). In contrast, the Cardy meter directly measures the fresh sap—a few drops of the petiole sap are placed on the sensor, and the result is displayed on the digital meter. Thus, the nitrate test with a Cardy meter can be performed easily and in a few minutes in the field.

The objective of this publication is to provide information on how to test petiole sap of tomato to

determine the plant nitrogen level. Instructions are provided on how to use a Cardy meter for this test and also how to interpret the petiole sap nitrate-nitrogen (N) test results.

Measuring Nitrogen in the Plant

The petiole sap nitrate-N test is a rapid and effective method for growers to monitor the nitrogen status of plants throughout the growing season (Burt et al. 1995). This method can be used for tomato grown in the field as well as in high tunnels and greenhouses (Bevacqua and Cardenas 2002; Hochmuth et al. 1991; Hochmuth 1994). The test provides immediate information about the status of crop nitrogen at the time of measurement so growers can make an immediate plan if deficiencies or excesses are present (Burt et al. 1995).

Plant nitrogen status can also be measured in dry tissue samples. While very accurate, there are several limitations to this nitrogen measurement method. Dry tissue analysis requires time for drying the samples, delivering them to a commercial laboratory, and interpreting the results. All these steps are time and money consuming. Further, the crop nitrogen status may change by the time the grower receives the results.

Nitrogen in the Soil and Plant Uptake

Understanding nitrogen behavior in the soil is helpful for understanding why the petiole sap nitrate-N test is an effective test for measuring crop nitrogen. Plants absorb nitrogen in two ionic forms, ammonium (NH_4^+) and nitrate (NO_3^-). The behavior of these two nitrogen ions in the soil differs (Aldrich 1980; Burt et al. 1995). The ammonium ion is held by soil particles because it has a positive charge, which makes it hard to leach, thus, it stays in the soil for a longer time. In contrast, nitrate has a negative charge, so it is more susceptible to leaching. Ammonium tends to be transformed to nitrate through the nitrification process. As a result, soil

tends to have abundant amounts of nitrate and lower amounts of ammonium. Thus, nitrogen in the soil and in the plant is usually measured in the nitrate form, because this is the form that is most commonly present and at the greatest level. The amount of nitrogen is then calculated from the amount of nitrate.

Tomato Nitrogen Demand and Need

Nitrogen is an essential constituent of chlorophyll and amino acids in tomato plants. The first symptoms of nitrogen deficiency are displayed on the lower, older leaves (Hochmuth et al. 1991). Chlorosis, red coloration in the petioles and in the veins of the leaves, and dropping of lower, older leaves, are common reactions of tomato to insufficient nitrogen (Figure 1). Subsequently, premature drop of flowers and small, immature fruit will occur if the deficiency persists. Excessive nitrogen supply, on the other hand, results in bullish growth (thick, curly tomato leaves and compact plant growth; Figure 2), and plants produce fewer flowers and become more susceptible to diseases and abiotic

stress. In addition, fruit set is inhibited, fruit are pale in color, and ripening is delayed (Hochmuth et al. 1991).



Figure 1. Nitrogen deficiency in tomato leaves (photo by Edward Scheenstra).



Figure 2. Failure to set fruit or blossom drop and delayed ripening due to excess nitrogen supply (photos by Edward Scheenstra).

Less than 30% of the total tomato plant requirement for nitrogen is prior to fruit set, while 70% or more of the total plant nitrogen requirement occurs after fruit set begins. Much of the nitrogen that is applied pre-plant will not be taken up immediately from the soil, as the nitrogen requirement of young plants is relatively low. This situation makes pre-plant applied nitrogen more susceptible to leaching (Thompson and Kelly 1957). Applying a large portion of nitrogen fertilizer early in the season, or pre-plant, can result in an insufficient nitrogen supply to the plant later on when the plant needs it, as the nitrogen can leach from the soil, and, thus, no longer be available for uptake. Applying nitrogen during the crop production season via fertigation is an effective way to meet crop nutrient needs.

How to Sample a Tomato Leaf

The tomato leaf is a compound leaf, and it consists of leaflets and rachis (short petiolules). The petiole is the stem that connects the entire leaf to the main stem (Figure 3). It is easy to distinguish the petioles, and petioles are easier to press to extract plant sap than leaf tissue. For these reasons, the petiole is used for the sap nitrate-N test.

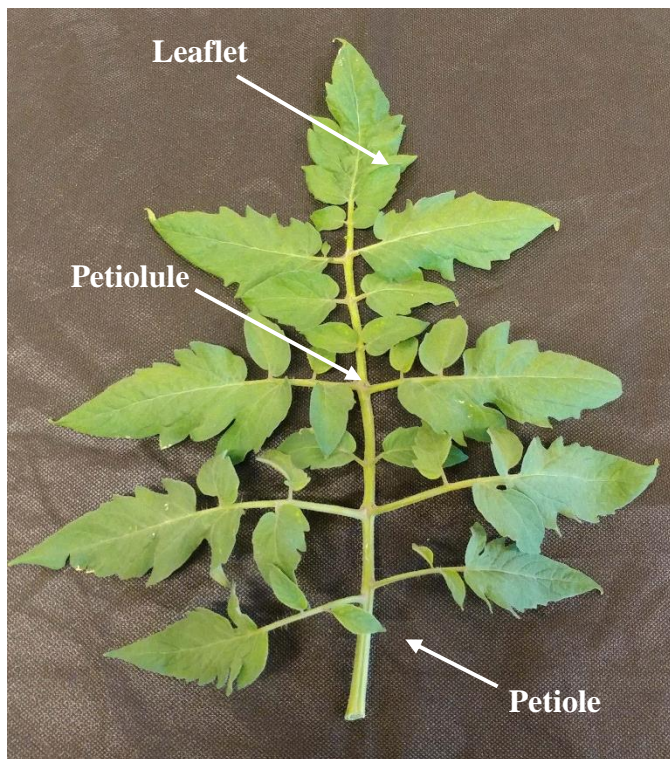


Figure 3. Tomato leaf morphology (photo by Edward Scheenstra).

Time of Day for Collecting the Sample

The plant petiole sap nitrate level is affected by the time of day (Hochmuth 1992). Most studies indicate that the most consistent readings will be obtained between 9 am and 2 pm. At this time of day, the nitrate-N level is highest in the petiole sap for tomato, due to increases in photosynthesis, which reaches the maximum around noon as a function of the intensity of sunlight. Samples collected using a standardized method and timing will provide the most accurate and consistent results over the growing season (Brust 2008; Hochmuth 1994; Thompson et al. 1996; Volpe et al. 2011).

Part and Age of Leaf to Sample

For consistency and accuracy, collect leaves that have the same physiological maturity. The fifth or sixth leaf from the top of the plant, which is the youngest fully mature leaf (Figure 4), is considered the best plant tissue to sample. This leaf is the newest mature leaf, so it has just completed its growth and development and is a reliable indicator of the plant's nutrient status (Geraldson et al. 1973). Avoid collecting leaves that appear to have any defects, such as disease or insect damage (Kaiser et al. 2013).

Sample Collection and Handling

In a field of 5 to 10 acres, where plant growth is uniform, collect 20 leaves randomly from representative areas of the field and bulk the leaves (Hochmuth 1994). In a non-uniform field, where plant growth differs from area to area, collect 3–6 leaves per area, and bulk the leaves from each area but keep the areas separate. It is better to test each area separately, and, if different nitrate levels are obtained, you may adjust fertilizer rates to each area as needed. In both situations, divide the leaves into subsamples of 3–4 leaves, as this will provide a sufficient amount of petiole sap for the nitrate-N test per subsample.

To obtain a more reliable reading, it is best to conduct the petiole sap nitrate-N test in a controlled indoor area rather than in the field. Environmental conditions in the field, such as elevated temperature,



Figure 4. Position of the fifth youngest but fully mature leaf from the top of tomato plant (photo by Edward Scheenstra).

dust, or direct exposure to sunlight might give inaccurate results. Place the leaves in plastic bags, place the bags on ice in an insulated cooler immediately after collection, and then transport the leaves to an indoor area. Separate petioles from the leaf tissue using a razor blade or pocket knife (Figure 5). Petiole samples can be kept in plastic bags for up to 2 hours at room temperature (70°F) or in plastic bags on ice in an insulated cooler up to 8 hours. Petioles also can be frozen in a home freezer



Figure 5. Leaf petiole samples separated from the leaf tissue using a pocket knife (photo by Edward Scheenstra).

(-10 to -20°F) overnight. Cold or frozen petioles need to be at room temperature before extracting the sap, however. The petiole sap nitrate-N test should be done within one or two minutes after sap extraction (Hochmuth 1994).

Petiole Sap Extraction and Nitrate Measurement

Extract the petiole sap from three to four petioles at a time. Use a garlic press or a plastic hammer and plastic bag to press the sap from the petioles. Avoid exposing the petiole sap to the air for more than a few minutes, as this might affect the nitrate reading. Place four to five drops of the petiole sap on the sensor of the Cardy meter and record the amount of nitrate (ppm) registered on the meter (Figure 6) (Andersen et al. 1999; Hochmuth 1994; Thompson et al. 1996). According to the manufacturer's recommendations, the Cardy meter should be calibrated before the first reading. After every ten readings, utilize a standard solution and recalibrate if needed (Thompson et al. 1996). The standard solution comes with the purchase of the Cardy meter, or it can be ordered as needed.

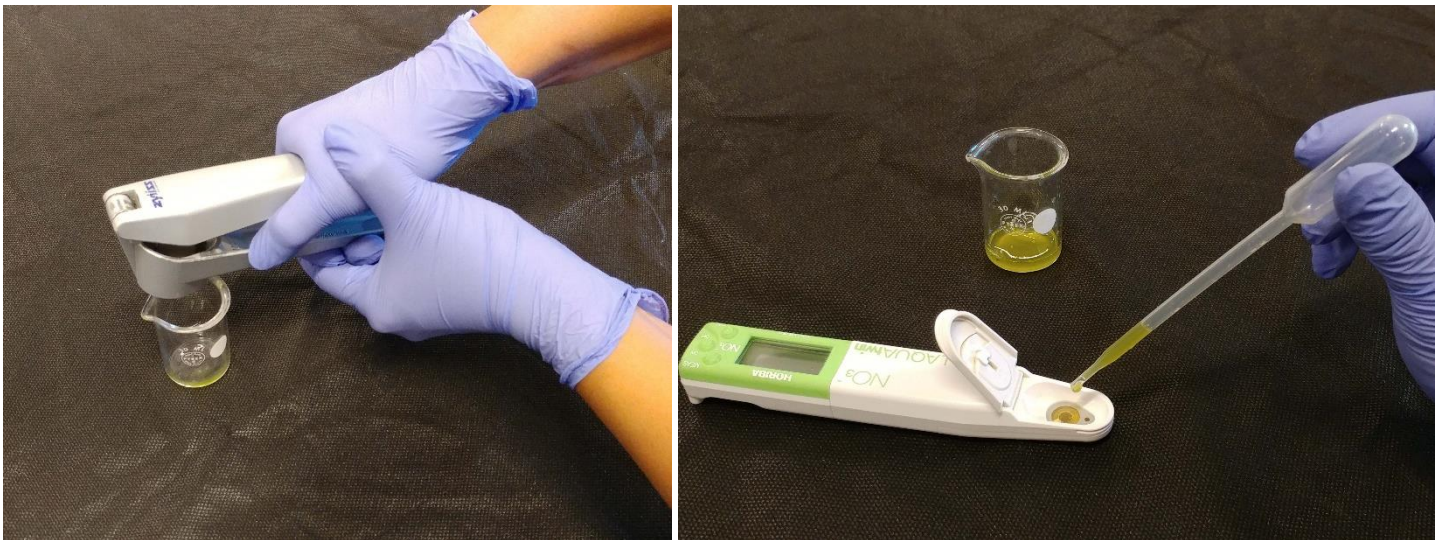


Figure 6. Extracting the petiole sap by using a garlic press and estimating nitrate amount (ppm) in the sap by using a Cardy meter (photos by Edward Scheenstra).

Convert Nitrate Values to Nitrate-N Values

The Cardy meter will provide a reading for the amount of nitrate in the sap. Calculate the amount of nitrogen in the sap sample using the following equations (Hochmuth 1994):

$$\text{NO}_3\text{-N (ppm)} = \text{NO}_3 \times 0.226$$

$$\text{Or, NO}_3\text{-N (ppm)} = \text{NO}_3 \div 4.43$$

The nitrogen molecule has a mass unit of 14, and the nitrate molecule NO_3 has a formula mass unit of

62: $\text{N} + 3(\text{O}) = 14 + (3 \times 16) = 62$. The amount of nitrogen in nitrate is $14 \div 62 = 22.6\%$, or $62 \div 14 = 4.43$.

For example, sap was extracted from a sample of field-grown tomato petioles at the first open flower stage, and the Cardy meter reading was 2000 ppm nitrate. The amount of nitrate-N in the sap sample is calculated to be: $2000 \times 0.226 = 452$ ppm, or, $2000 \div 4.43 = 452$ ppm. This amount of nitrate-N is under the sufficient range of 600–800 ppm (Table 1). Thus, the amount of nitrate in the leaf petiole is categorized as insufficient, and it is recommended to apply additional nitrogen fertilizer.

Table 1. Tomato fresh petiole sap nitrate-N concentration guidelines (source: Hochmuth 1994).

Production location	Tomato growth stage	Nitrate-N fresh petiole sap concentration (ppm)
Field	First flower buds	1000–1200
	First open flowers	600–800
	Fruit 1-inch diameter	400–600
	Fruit 2-inch diameter	400–600
	First harvest	300–400
	Second harvest	200–400
Greenhouse	Transplant to second fruit cluster	1000–1200
	Second cluster to fifth fruit cluster	800–1000
	Fruit harvest	700–900

Determining Nitrogen Sufficiency

Sufficient nitrate-N concentration in the petiole sap for tomato is different for each plant growth stage (Table 1) (Hochmuth et al. 1994). Nitrate-N sufficiency levels also differ between field-grown tomato and greenhouse-grown tomato. There are no guidelines for high tunnel-grown tomato nitrate-N sufficiency level; however, growers can follow the field-grown tomato guide, because field- and high tunnel-grown tomato tend to have a determinate growth habit and are grown directly in the soil, in contrast to greenhouse tomato cultivars that are indeterminate and grown in soilless media. If the sample readings indicate plants are low in nitrogen, apply fertilizer through the irrigation system (fertigation). If sample readings indicate plants have sufficient or excess nitrogen, do not apply any further nitrogen. If sample readings continue to show an excess in the level of nitrogen during the crop season, adjust the fertilizer application rate in the next growing season so that excess fertilizer is not applied.

Summary

The petiole sap nitrate-N test is a good investment for guaranteeing that tomato plants receive enough nitrogen during the growing season but fertilizer is not overapplied. It is important to remember that proper collection of plant samples is necessary to obtain accurate and correct values. It is sometimes difficult to select the representative area in a field to collect the sample from. When in doubt, collect more leaf samples. In addition, there are several factors that can significantly affect nitrate values other than excessive or deficient N fertilizer, such as stress on plants caused by drought or cold damage. The suggested resources provided in this publication provide more information about testing nitrate-N in plant petiole sap.

Suggested Resources

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