



Use and Accuracy of the Diphenylamine Field Kit for Determining the Presence of Toxic Nitrate Levels in Forage Samples

Brent Bean

Texas Agricultural Extension Agronomist — Amarillo

The diphenylamine quick test can be used in the field to determine if nitrate is at level in forage that may pose a risk to livestock. The test is primarily an indicator of the presence of high nitrate concentration and is not quantitative. If the test indicates the presence of nitrate, samples should be sent to a lab for quantitative analysis. When using the kit, or collecting samples for lab analysis, always collect several samples from different parts of the field.

To use the kit, plants to be tested should be cut at the soil level and the stalk split lengthwise. Lay the stalk on the ground with the exposed pith facing upwards. Allow test solution to fill the dropper and let a drop fall on the pith 2 to 3 inches from the base of the stalk. Place additional drops further up the stalk. Generally nitrate levels will be higher closer to the base of the plant. The solution will turn dark blue or black within 20 seconds if nitrates are present at a high concentration. The deeper the color the higher the concentration of nitrate. A mild tinge of blue or no color change would indicate little nitrate is present.

The accuracy of the kit was examined in Oklahoma by comparing the results obtained with the kit to laboratory analysis. The kit proved to be very accurate in detecting nitrate in samples having a concentration level that would be expected to be toxic to livestock. Where nitrate concentration was 6,000 - 10,000 ppm the kit indicated the presence of nitrate 86% of the time in at least one of two plants tested. At nitrate concentration levels greater than 10,000 ppm the kit identified the presence of nitrate 95% of the time. The draw back to the accuracy of the kit is that false positives are often given when nitrate levels are not high enough to pose a threat to livestock. False positives were recorded 46% of the time in at least one of the two samples tested but only 24% of the time in both samples. Increasing the number of samples tested will increase the accuracy to the test. One reason for the high level of false positives is that the stem contains much more nitrate than leaves. In the laboratory test the nitrate level is diluted when leaves are added to the sample being tested.

Percentage of 1064 Samples Giving Positive (Blue Color) Reaction When Exposed to Diphenylamine

| | 0 - 2,500 Safe | 2,500 - 6,000 Mod. Safe | 6,000 - 10,000 Potentially Toxic | 10,000 - 15,000 Very Toxic | > 15,000 |
|--|-------------------|----------------------------|-------------------------------------|-------------------------------|----------|
| Percentage of samples one or two turned blue | 46 | 67 | 86 | 95 | 100 |
| Percentage of samples both turned blue | 24 | 44 | 71 | 86 | 93 |

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