

64 and 65 series spectrophotometers Protocol: P09-007A

Using water test analysis kits with the 64 and 65 series spectrophotometers

■ Introduction

The Jenway water analysis test kits are colorimetric kits for the determination of various ions and compounds in water. The Jenway Aquanova spectrophotometer is pre-programmed with parameters for each of the tests and is fitted with a cell holder to fit the various vials or cuvettes used for the tests. However it is possible to use these test kits on other Jenway spectrophotometers.

First it is necessary to decide whether, if using test kits, the tests are to be performed in the specified vials (either 16mm or 24mm diameter for Jenway kits). If so then it will be necessary to purchase the Aquanova cell holder, part code 637 071. If using a 64 series spectrophotometer with a single cell holder fitted, this can simply be exchanged for the Aquanova cell holder. If a 65 series is to be used or a 64 series fitted with an 8-cell changer, it will also be necessary to purchase a base-plate kit, part code 630-005 to which the Aquanova cell holder can be fixed; the 8-cell changer will have to be completely removed. The other alternative is to perform the test in 10mm square cuvettes for which no modifications to the instruments will be required; a portion of the sample can simply be transferred from the vial to a cuvette.

Since the 64 and 65 series spectrophotometers are not pre-programmed with the test kit data, it will be necessary to determine the appropriate factors using standard solutions to construct a calibration curve. The simplest mode to use (and the only mode if a single cell holder is fitted) is Photometrics. As an example, a step by step method is given below for using the Jenway Nitrate test kit, part code 025 325, with the model 6405 in Photometrics mode using a single cell holder (Aquanova vial or 10mm square cuvette).

■ Method

Determination of Nitrate in the Jenway model 6405 using the test kit 025 325

When running the test for the first time it is necessary to determine whether the test gives a linear standard curve over the concentration range. This can be done by measuring the absorbance of a series of standards and plotting a calibration curve. If the curve is linear, the derived factor (gradient) can subsequently be entered in the concentration set up and the concentration mode will give a direct readout of the unknown sample concentration. Alternatively, each time a new test is performed, a single calibration standard can be used to calibrate the instrument. If the curve is not linear then the absorbance of unknowns will need to be converted to concentration by calculation from the calibration graph.¹

1. Turn on the 6405 (first making sure there are no samples in the chamber) and allow to warm up for about 30min.
2. From the Main Menu, select Photometrics mode.
3. Select SETUP using the arrow keys and press the Enter key.
4. Scroll down to MEASURE MODE and select ABSORBANCE.
5. Scroll to EXIT and press the Enter key.
6. Select the required wavelength (410nm for the Nitrate kit) using the up/down keys, or by pressing the Goto key and entering the required value using the numeric keypad. Press Enter to confirm.

¹ If a multi-cell changer for 10mm cuvettes is fitted in the instrument, then it is possible to set up the standard curve in Quantitation mode and this will calculate the results from non-linear curves as well as linear. Follow the instructions given in the manual.

Preparation of standards and samples

For the calibration curve a nitrate standard solution is required. The range of the Jenway Nitrate kit is 1-30mg/l N which is equivalent to 4.43 to 133mg/l NO_3^- . The conversion factor is as follows: $\text{mg/l NO}_3^- = \text{mg/l N} \times 4.3$

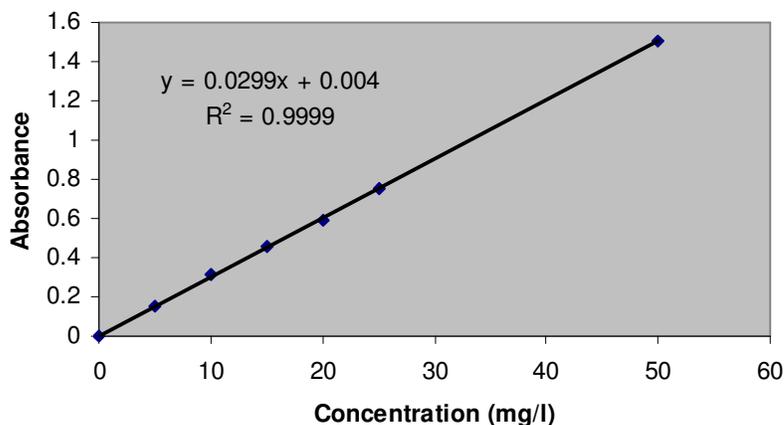
Standards can either be prepared from a potassium nitrate stock solution or from a purchased nitrate standard. Jenway offers 1000ppm, 100ppm and 10ppm (1000, 100 and 10 mg/l respectively) nitrate standard solutions part codes 025 067, 025 152 and 025 151 respectively. To prepare a nitrate stock, prepare a suitable portion of potassium nitrate, KNO_3 , by drying at 105°C for 3 hours until a constant weight is obtained. Dissolve 7.2182g of the dried potassium nitrate in distilled water and make up to 1000ml. This solution is a 1g/l nitrogen standard (equivalent to 4.43g/l NO_3^-).

7. Dilute the standard stock solution to prepare at least 5 standards covering the concentration range of the test samples.
8. Add 1ml of standard or sample to each vial (16mm diameter). Prepare a blank by adding 1ml of deionised water to a vial.
9. Add one Nitrate Chromotropic powder pack straight from the foil to each vial and cap tightly. Invert 10 times to mix. Some powder may not dissolve. Incubate for 5 min.
10. If using 10mm cuvettes transfer the samples to cuvettes (e.g. part code 060 087).
11. Place the blank sample in the 6405 and press the Cal key. The display will momentarily show 'Calibrating Dark Level' and then 'Calibrating Light Level'. The display will then update to show 0.000Abs.
12. Remove the blank sample. Place the first standard sample in the chamber and record the absorbance. Repeat for the remaining standards.
13. Plot a calibration graph, concentration vs. absorbance and check for linearity.

If the standard curve is linear over the range to be tested, then a factor can be entered in the CONCENTRATION SETUP to allow direct readout of concentrations from the unknown samples.

Example standard curve:

Standard curve



The factor is the value of m defined as the slope of the straight line in $Y = mX + C$, where Y = the concentration value, m = the slope factor, X = the sample absorbance and c = the blank absorbance. Here the factor is 0.0229. Note this is an example only and not a real test.

14. From the Photometrics mode select SETUP using the arrow keys and press the Enter key.
15. Scroll down to CONCENTRATION SETUP and at FACTOR, enter the derived factor from the standard curve.

16. Scroll down to UNITS and select mg/l.
17. Exit back to MEASURE MODE and select ABSORBANCE.
18. Scroll to EXIT and press the Enter key.
19. Place the blank sample in the 6405 and press the Cal key. The display will momentarily show 'Calibrating Dark Level' and then 'Calibrating Light Level'. The display will then update to show 0.000Abs.
20. Select SETUP using the arrow keys and press the Enter key.
21. Scroll down to MEASURE MODE and select CONCENTRATION.
22. Scroll to EXIT and press the Enter key.
23. Place the first sample in the 6405 and the reading will show the concentration of the sample calculated from the factor entered during setup.

The next time the test is done, the same factor can be used without having to run the complete standard curve. However for improved accuracy and validation, each time the test is performed, the instrument can be calibrated with a single standard and the factor will be modified accordingly. This will account for any changes which may occur for instance in batches of the test kits.

24. Prepare a blank and a standard using the highest concentration required for the test.
25. From the Photometrics mode select SETUP using the arrow keys and press the Enter key.
26. Scroll down to CONCENTRATION SETUP and at CAL STANDARD, enter the concentration of the standard sample.
27. Scroll down to UNITS and select mg/l.
28. Exit back to MEASURE MODE and select ABSORBANCE.
29. Scroll to EXIT and press the Enter key.
30. Place the blank sample in the 6405 and press the Cal key. The display will momentarily show 'Calibrating Dark Level' and then 'Calibrating Light Level'. The display will then update to show 0.000Abs.
31. Select SETUP using the arrow keys and press the Enter key.
32. Scroll down to MEASURE MODE and select CONCENTRATION.
33. Scroll to EXIT and press the Enter key.
34. Place the standard in the 6405 and press the Cal key. The instrument will now display the concentration of the calibration standard and re-calculate the factor.
35. Place the first sample in the 6405 and the reading will show the concentration of the sample calculated from the factor based on the standard used for calibration.
36. The new factor can be viewed by going back into CONCENTRATION SETUP.

When using the model 65xx, the instructions are essentially the same, only the software is in a different format.