



TEST KIT FOR THE DETERMINATION OF FREE SULFUR DIOXIDE

PRODUCT

Product no. 4A190, for 30 tests.

CONTENTS

The kit includes the following reagents:

Reagent No.	Reagent	Preparation	Quantity	Stability
1	Buffer	Ready to use	33 mL	Stable
2	Chromogen	Ready to use	17 mL	Stable
3	Standard	Refer to standard preparation procedure	2 x 74 mg	Stable as supplied

Standard concentration is 50 mg/L when prepared as instructed.

SAFETY

- Please read the Safety Data Sheets (SDS) before use
- Take the necessary precautions for the use of laboratory reagents

PROCEDURE

Operating Parameters

Wavelength

340 nm

Cuvettes

Semi-micro with 1cm path length

Temperature

20 – 25°C

Final volume in cuvette

1.80 mL

Zero

against air with no cuvette in light path

STANDARD PREPARATION

Quantitatively transfer the contents of one bottle (74 mg) of the supplied sodium metabisulfite powder ($\geq 99\%$ purity) to a 100 mL volumetric flask. Add a little distilled water to the bottle and transfer this to the volumetric flask to ensure all of the powder has been transferred. Make to volume with distilled water. Immediately cap and mix until completely dissolved. Store this concentrated solution in the refrigerator for up to one week. To prepare the 50 mg/L working standard, dilute 1 mL of the concentrated sulphite solution with 9 mL of distilled water, i.e. prepare a 1 in 10 dilution. Discard after use. Fresh working standard can be prepared from the concentrated stock solution as needed.

Please note that the Standard in this assay is used as a calibration factor (for calculation purposes only) and will not give a mg/L result. Expected A1 Standard absorbance is approximately 0.1, expected A2 Standard absorbance is approximately 1.2 – 1.4.

SAMPLE PREPARATION

DO NOT decolourise with either PVPP or activated charcoal, as both fining agents have been demonstrated to remove sulfite from the sample. Turbid samples may be filtered or centrifuged.

All red wine samples must be diluted 1 in 5 with distilled water.

DO NOT dilute white wines, ciders or spirits unless the final A_2 absorbance reading is greater than 1.5 absorbance units, or the sample contains more than 50 mg/L of free SO_2 . If dilution is needed, the best results are achieved with the least dilution possible, for example dilute 1 in 2 with distilled water.

It is recommended to run a sample with known free SO_2 concentration (such as a cask wine previously tested) as a control with each assay.

SAMPLE ANALYSIS

a. Pipette the following volumes of reagents into the cuvettes:

Reagent	Blank assay	Standard assay	Sample assays
1. Buffer	1.00 mL (1000 μL)	1.00 mL (1000 μL)	1.00 mL (1000 μL)
Sample or Standard		0.30 mL (300 μL)	0.30 mL (300 μL)
Distilled water	0.30mL (300 μL)		

b. Mix well by gentle inversion and read absorbances, A_1 , after 3 minutes.

c. Pipette the following reagent into the cuvettes:

2. Chromogen	0.50 mL (500 μL)	0.50 mL (500 μL)	0.50 mL (500 μL)
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d. Mix well by gentle inversion and read absorbances, A_2 , at **precisely 10 minutes**.

CALCULATIONS

1. Calculate the net absorbance for the blank assay:

$$\text{Blank absorbance, } A_{\text{RB}} = A_2 - (A_1 \times 1300/1800)$$

2. Calculate the corrected absorbance for the standard assay:

$$\text{Standard absorbance, } A_{\text{STD}} = A_2 - (A_1 \times 1300/1800)$$

$$\text{Corrected absorbance, } C_{\text{standard}} = A_{\text{STD}} - A_{\text{RB}}$$

3. Calculated the corrected absorbance for the samples:

$$\text{Sample absorbance, } A_{\text{SAMPLE}} = A_2 - (A_1 \times 1300/1800)$$

$$\text{Corrected absorbance, } C_{\text{sample}} = A_{\text{SAMPLE}} - A_{\text{RB}}$$

4. Since the concentration of the standard is 50 mg/L, calculate the free sulfur dioxide content of the samples as follows:

$$\text{Free } \text{SO}_2 \text{ (mg/L)} = \frac{C_{\text{sample}}}{C_{\text{standard}}} \times 50 \text{ mg/L} \times \text{dilution factor}$$

A calculation spreadsheet is available for download at:

<http://www.vintessential.com.au/certification/calculation-worksheets/>

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