

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 as supplied
2	Chromogen	Ready to use	17 mL	Stable as supplied
3	Standard	Refer to standard preparation procedure	4 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 (≥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 SO₂. 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 SO2 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, A1, 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, A2, at precisely 10 minutes.

CALCULATIONS

- 1. Calculate the net absorbance for the blank assay: Blank absorbance, $A_{RB} = A2 - (A1 \times 1300/1800)$
- 2. Calculate the corrected absorbance for the standard assay:

Standard absorbance, $A_{STD} = A2 - (A1 \times 1300/1800)$ Corrected absorbance, $C_{standard} = A_{STD} - A_{RB}$

3. Calculated the corrected absorbance for the samples:

Sample absorbance, $A_{SAMPLE} = A2 - (A1 \times 1300/1800)$ Corrected absorbance, $C_{sample} = A_{SAMPLE} - A_{RB}$

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

Free SO₂ (mg/L) = $C_{\text{sample}} \times 50 \text{ mg/L x dilution factor}$

*A calculation spreadsheet is available for download at the following locations. Australia based users <u>https://winechek.com/calculation-worksheets/</u>

Users outside of Australia http://www.vintessential.com.au/resources/calculation-worksheets/

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