

Wine Phenolics & Color

Product: Phen&Color_{ABS} F150 (75 Tests)

Uv-vis Absorbance method for red wine assessment

INTENDED USES

This reagent is intended for assessing red wine Color & Anthocyanin **uv-vis** ABS Profiles (**Red wine color** and **Polymeric, Total & Ionized Anthocyanins**) specifically for the uv-vis assessment of Wine Color & Anthocyanin Profiles.

REAGENTS

| Kit Contents | 75-Tests |
|-------------------------|----------|
| Wine Color Diluent | 120mL |
| SO ₂ Reagent | 10mL |
| ACE Reagent | 6 X 2mL |
| Ionizing Reagent | 150mL |

REAGENT DESCRIPTION

1. Red wine color

Wine Color Diluent standardizes 420, 520, 620nm Absorbance measurements by controlling pH; wine ABS is pH dependant. These tests permitting calculation of Wine color intensity, color Density, and Hue.

2. Polymeric Anthocyanin - sulfite bleaching¹

SO₂ Reagent is mixed with Wine samples to allow small chain Anthocyanin pigments to be 'bleached.' Polymeric Anthocyanins content is proportional to the remaining pigments are measured at 520nm. (**Monomeric Anthocianin** content is proportional to the change ABS during SO₂ bleaching.)

3. Total Anthocyanins

ACE Reagent – when acetaldehyde, which strongly binds sulfite, is mixed with wine samples, any bleaching influence is removed. The 520nm ABS is a measure of total Anthocyanins.

The above measurements permit calculation of co-pigmented and monomeric anthocyanins:

- o **Co-Pigmented Antho** = Total Antho – Color 520
- o **Monomeric Antho** = Color 520 - Polymeric Antho

4. Ionized Anthocyanins

Ionizing Reagent shifts causes a shift of red wine pigments toward 520nm ABS (i.e. to the Red spectrum.)

PREPARATION & STORAGE

Reagents are ready to use and stable through the labeled expiration date when stored at 2 to 8° C. After opening, protect from direct light and keep tightly closed.

Instrument Parameters: Spectrophotometer, 1cm light path, 20-37C. Read ABS values at wavelengths specified in each procedure; if available on spec, use the 750nm differential wavelength setting. ABS results account for the dilution factors, thus maintaining proportionality for these wine measurements.

PROCEDURES (Manual)

For 1cm lightpath ABS (multiply by 0.1 for 1mm lightpath)

1. Red wine color Procedure: Prepare Blank and sample cuvettes and incubate as shown.

| Pipette into Cuvettes | Reagent Blank | Sample cuvettes |
|--|---------------|-----------------|
| Sample | | 500µL |
| DI water | 500µL | |
| Wine Color Diluent | 1500µL | 1500µL |
| Mix, wait 2 minutes | | |
| Read Abs _{420, 520 & 620nm} | | |

With instrument set at 420nm, zero with Reagent Blank and measure the ABS of each sample; repeat zeroing on blank and reading each sample at 520nm, and again at 620nm ABS.

For above, the sample **dilution factor is 4**. If any Abs reading exceeds your spectrophotometer range, repeat using 250uL Sample and 1750uL DI Water, mix and re-read (this sample **dilution factor** would be **8**.)

Correct for the dilution factor *f*:

Abs (420, 520 & 620) X *f* (dilution factor)

Calculate Standardized Wine Color:

- o **Color Intensity:** Abs 420 + Abs 520 + 620nm
- o **Color Density:** Abs 420 + Abs 520
- o **Color Hue:** Abs 420 / Abs 520
- o **Contribution % by Wavelength**

$$\% 420 = \frac{\text{Abs } 420}{\text{Abs } 420 + \text{Abs } 520}$$

$$\% 520 = \frac{\text{Abs } 520}{\text{Abs } 420 + \text{Abs } 520}$$

2. Polymeric Anthocyanins Procedure:

Prepare Blank and sample cuvettes and incubate as shown. Zero your instrument with Reagent Blank; measure and record 520nm ABS for each cuvette.

| Pipette into Cuvettes | Reagent Blank | Sample Cuvettes |
|---|---------------|-----------------|
| Sample | | 667µL |
| SO ₂ Reagent | 100 µL | 100 µL |
| DI water | 1900µL | 1233µL |
| Mix, wait 2 minutes at room temperature | | |
| Read Abs _{520nm} | | |

Polymeric Anthocyanins are proportional to **Abs 520nm X 3** where 3 is the dilution factor.

3. Total Anthocyanins Procedure:

Prepare Blank and sample cuvettes and incubate as shown. Refer to Examples below for alternate dilutions.

| Pipette into Cuvettes | Reagent Blank | Sample cuvettes |
|---|---------------|-----------------|
| Sample | | 667µL |
| ACE Reagent | 133µL | 133µL |
| DI water | 1867µL | 1200µL |
| Mix, wait 2 minutes at room temperature | | |
| Read Abs _{520nm} | | |

Zero your instrument with Reagent Blank; measure and record ABS for each sample. **Total Anthocyanins** are proportional to **Abs 520nm X 3** where 3 is the dilution factor.

4. Ionized Anthocyanins Procedure:

Prepare Blank and sample cuvettes and incubate as shown. Zero your instrument with Reagent Blank; measure and record 520nm ABS for each cuvette.

| Pipette into Cuvettes | Reagent Blank | Sample cuvettes |
|---|---------------|-----------------|
| Sample | | 200µL |
| DI Water | 200µL | |
| Ionizing Reagent | 1800µL | 1800µL |
| Mix, wait 2 minutes at room temperature | | |
| Read Abs _{520nm} | | |

Ionized Anthocyanins are proportional to **Abs 520nm X 5** where 5 is the dilution factor.

Additional calculated phenolic results:

- **Co-Pigmented Antho** = Total Antho* – Color 520**
- **Monomeric Antho** = Color 520** - Polymeric Antho*

* Values calculated per Procedures 3 and 2 above, respectively

** Values calculated per Procedure 1 above

PROCEDURES (Automation)

Contact Unitech Scientific for the [ChemWell for Wine™](#) automated test procedures, indexes and technical support.

QUALITY CONTROL

Each laboratory should establish stable 'check wines', and a Quality Control procedure if controls do not recover within the acceptable tolerances.

NOTES:

¹ The 520nm ABS of single (and dimeric) phenols decreases in the presence of SO₂, due to the formation of anthocyanin-sulfite complexes. This decrease red color, referred to as **SO₂ bleaching**, is a measure of Monomeric anthocyanin content. **Polymeric anthocyanin** color intensity is not affected by the presence of SO₂, and is measured at 520nm following the addition of SO₂ to wine.

Contact Unitech Scientific for information about our additional wine phenolic reagent kits

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