

Application of spectroscopy for non-destructive 'in-bottle' measurement of wine composition and quality

Introduction

Spectroscopic techniques used or developed by the Rapid Analytical Methods team of The Australian Wine Research Institute (AWRI) include the ultraviolet (UV), visible (Vis), near infrared (NIR) and mid-infrared (MIR) wavelength regions of the electromagnetic spectrum. These have been used to develop methods for rapid analysis and quality control in the Australian wine sector, as well as by wine scientists and researchers. Key factors contributing to the development of these methods by work done at the AWRI and in the wine sector have been major advances in instrument reliability, readily available chemometrics software and improved computing power – these have enabled a paradigm shift in rapid analytical methods based on spectroscopic data.

Infrared and UV-Vis spectra can effectively represent a 'fingerprint' of the grape or wine sample being analysed and can be used to simplify and reduce analytical times for many grape

and wine analytes. Rapid sensor techniques are also increasingly being used by research scientists as cost-effective quantitative and qualitative analytical tools. They can be used for continuous and non-destructive measurements on grape and wine products and processes over time, and in many cases give an understanding of the chemical properties of in-process materials during the production of various wine products. These methods are attractive due to their inherent features of versatility, flexibility, effectiveness and richness of information.

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What is infrared spectroscopy?

Infrared spectroscopy uses radiation in the region of the electromagnetic spectrum between the visible and the microwave wavelengths. The nominal range of wavelengths for near-infrared (NIR) is between 750 and 2,500 nm ($13,400$ to $4,000$ cm^{-1}), while for the mid-infrared (MIR), the spectral range is from 2,500 to 25,000 nm ($4,000$ to 400 cm^{-1}). Solid, liquid or gaseous samples can absorb some of the incoming infrared radiation at specific wavelengths resulting in a 'fingerprint' or spectrum.

Although NIR intensities are 10-1000 times lower than for the MIR range, and the peaks concomitantly smaller, highly sensitive spectrometers can be built through several means including the use of efficient detectors and brighter light sources. This allows concentrated bulk or even aqueous materials to be scanned and analysed quickly and easily.



Whilst spectral peaks in the MIR frequencies are often sharper and better resolved than in the NIR, all the higher overtones (1st through 6th) of the OH, NH, CH, and SH bands from the MIR wavelengths are still observed in the NIR region, although much more weakly. This, in addition to the existence of combination bands (e.g. CO stretch and NH bend in protein), gives rise to a relatively crowded NIR spectrum with overlapping bands.

A major disadvantage of this characteristic overlap and complexity has been the difficulty of quantification and interpretation from NIR spectra.

On the other hand, the broad overlapping bands can diminish the need for using a large number of wavelengths in calibration and analysis routines. In recent years, new instrumentation and computer algorithms have taken advantage of this complexity and have made the technique much more powerful and simple to use.

NIR spectroscopy is characterised by low molar absorptivities and scattering, which permit nearly effortless evaluation of pure materials. The NIR region of the electromagnetic spectrum, once regarded as having little potential for analytical application, has now become an inexpensive and powerful technique with the advent of computers and better spectrophotometers (e.g. increased signal to noise ratio). NIR spectroscopy conserves time and materials in comparison to many more conventional analytical methods because:

1. analysis times under one second are possible
2. simultaneous multicomponent analysis is the norm
3. no or very little sample preparation is required for liquids, solids, or gases
4. non-invasive and non-destructive analysis is possible
5. cost per analysis is very low (no reagents are used)
6. automated correction of background and interferences can be performed in instruments using computer algorithms
7. detection limits can be low, and precision can be high
8. sample sizes ranging 'from picograms to planets' can be analysed
9. some molecular structural information can be estimated from spectra

It is well known that the NIR technique is based on the correlation between chemical properties, as determined by defined reference methods, and absorption of light at different wavelengths in the NIR region, measured by reflectance, transmittance or transreflectance (Williams, 2001). In NIR spectroscopy, radiation scattered by particles is measured and gives both physical and chemical information related with composition, allowing this technique to measure different types of samples from solids to liquids.

In-bottle measurement

An ideal method for the routine determination of chemical composition in a food or beverage manufacturing process should be non-invasive, nondestructive and rapid to ensure timely processing of the product being analysed. NIR spectroscopy offers the advantages of simplicity of sample handling and the speed of collecting the information (spectra).

During the last 20 years NIR spectroscopy has assumed immense practical and economic importance as a rapid, integrated method for testing of a wide range of grown or manufactured products (McClure 2003). At short wavelengths (700 – 1600 nm), the spectrum has less intense peaks and can tolerate longer path lengths (5 up to 30 mm) but yields poor spectral resolution, while peaks in longer wavelength regions (> 1500 nm) are more intense and require short optical path length but provide

better spectral resolutions (Murray 1993; Williams 2001).

Glass is transparent to NIR wavelengths, allowing the use of NIR radiation to analyse liquids in glass containers (Kamat et al. 1999). Several authors report the use of glass vials of different sizes and/or from different suppliers in the pharmaceutical industry. This work demonstrated the use of NIR spectroscopy to determine moisture in lyophilized sucrose through intact glass vials.

The use of NIR spectroscopy was reported by other authors for analysis of foods and beverages such as sufu, beer and whisky, non-destructively, in their point of sale containers (Lu and Han 2005; Iñon et al. 2005; Nordon et al. 2005).

The Vis-NIR spectroscopy method has been used by the wine industry in order to assess the quality of grape spirit, and to measure composition in both grapes and wines (Damberg et al. 2002 and 2003). Recently, Skouroumounis and co-workers at the AWRI demonstrated that single wavelengths in the Vis region could be used to assess oxidation (browning) of white wines in either coloured or non-coloured bottles (Skouroumounis et al. 2003).

Therefore, it was considered possible to extend the analysis of wine in the bottle to the NIR region, because weak absorption bands, observed for wine at shorter wavelengths in the NIR region (750 to 1100 nm), may allow the use of the longer path lengths associated with the bottles themselves. An innovative technique utilising Vis-NIR spectroscopy (Australian Innovation Patent AU 2005100565 A4) has been developed for the assessment of wine composition and quality directly and non-destructively in the bottle.

Applications of in-bottle spectroscopy for the wine sector

Determination of wine composition

A range of commercially available bottled white and red wines (n= 226) were scanned in transmittance mode (400 – 1100 nm) using a scanning monochromator (FOSS NIRSystems6500 (FOSS NIRSystems, Silver Spring, MD, USA)).

Apart from the plastic box for mounting the bottles, no modifications (hardware) to the original configuration of the instrument were made (Figures 1A and B). No temperature control of the sample was used. Wine samples were analysed for alcohol content, pH, and the concentrations of free and total sulfur dioxide (SO₂) using standard laboratory methods at the AWRI Analytical Service.

Partial least squares regression (PLS) between spectra and chemical compositional data were developed using full cross validation and the optimum number of terms (latent variables) in the PLS calibration models were determined by the PRESS function (Naes et al. 2002).

Statistics calculated for the calibrations included the coefficient of correlation in calibration (R^2_{cal}) and the standard error of cross validation (SECV). The residual predictive deviation (RPD), defined as the ratio between the standard deviation of the population and the SECV, was used to test the accuracy of the calibration models developed. If the RPD is greater than three, calibration models are considered as acceptable for analytical purposes. Table 1 shows the results for the NIR calibrations for alcohol, free and total SO₂ and pH using in-bottle spectroscopic analysis.



Figure 1A: Presentation of a wine bottle to a NIR spectrophotometer to measure composition.



Figure 1B: Bottle sample holder box used as attachment to the instrument.

Table 1: Near infrared calibration statistics for wine composition measured in the bottle

	N	SD	R^2_{cal}	SECV	RPD
Ethanol (% v/v)	96	0.85	0.67	0.48	1.8
Free SO ₂ (mg/L)	53	8.75	0.83	4.01	2.2
Total SO ₂ (mg/L)	111	47.4	0.70	28.6	1.7
pH	55	0.18	0.50	0.15	1.2

N: number of samples used to develop the cross validation model

R^2_{cal} : coefficient of correlation in calibration

SECV: standard error of cross validation

RPD: SD/SECV

Wine quality

Several case studies have been conducted at the AWRI in order to assess the ability of this Vis-NIR method to monitor relative wine quality in respect of compositional changes related to oxidation in batches of bottled white wines. Preliminary studies showed that the bottle to bottle variation, due to the glass in such batches did not appear to have a large effect on the ability of the method to develop useful calibrations from the NIR scans.

Therefore, it appears that for ease of use in a commercial setting, attention to bottle orientation to the detector is not required.

A sample set of white wines, sourced from another project at the AWRI, that were being assessed by a sensory panel of experts for oxidised characters were also scanned in the bottle to evaluate the potential of this method to measure wine quality, such as oxidation (Skouroumounis et al. 2003). This sample set comprised replicate bottles of a single wine bottled several months before analysis and differing only in degree of development after bottling (Skouroumounis et al. 2003).

The principal component analysis (PCA) plot of the spectra (Figure 2) shows that the wine samples clustered into two groups according to the level of oxidation (low and either medium or high) as scored by the tasters. This result suggests that a PCA analysis of wine spectra may allow monitoring of the extent of oxidation in wine bottles. The PCA loadings showed features around 700 and 900 nm. The first principal component (PC1) explains 96% of the variation, with a valley around 930 nm and a peak at 980 nm both associated with O-H and C-H absorption bands, due to water and ethanol.

PC2 explains 3% of the variation, with loadings at 670 nm, 770 nm, 838 nm, 918 nm, 954 nm and 990 nm due to O-H and C-H stretch third overtone vibrations, associated with water and ethanol. The results of the PCA suggest that the monitoring of bottled wine for relative degree of oxidation is possible.

Summary

The results of the research described here demonstrate the potential application of Vis-NIR to determine wine composition in the bottle, although the accuracy and precision suggests that it is not sufficient for analytical purposes. Nevertheless, the possibility of monitoring wine quality parameters through the bottle non-intrusively gives a new dimension to the analysis of wine quality. New commercially available spectrophotometers in the market (e.g. diode array instruments) with simple sample presentation extend this potential for at/on-line applications. Although these results are promising, the authors believe that this application of spectroscopy can be used only as an indicative rather than a quantitative analytical tool because of limitations in analytical accuracy. It is expected

that future development of this application will provide the wine industry with a very fast and non-destructive method to monitor composition or changes and to detect unwanted problems in bottled wine prior to retail sale (e.g. oxidation), and provide a rapid means of qualitative analysis.

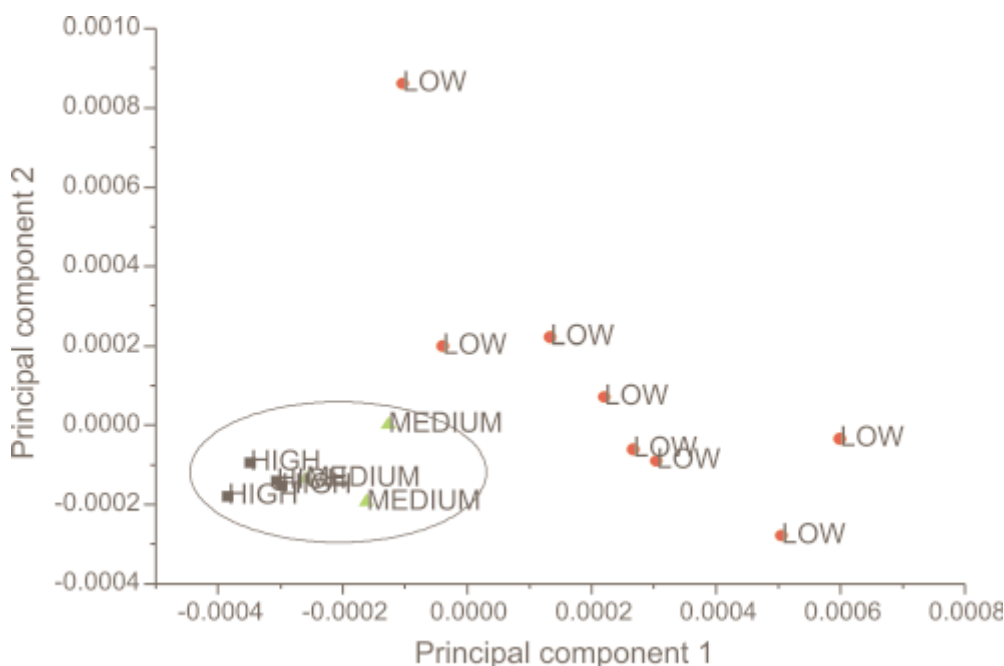


Figure 2: Principal component scores of wines analysed in the bottle labeled by degree of oxidation

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