Accurate mid-infrared analysis in wine production - fact or fable?

By Eric Wilkes and Luke Warner, Interwinery Analysis Group*
*Email: chairperson@interwinery.com.au

Since the late 1990s, a number of companies have offered instruments based on mid-infrared (MIR) spectrometers to study a wide range of analytes in both grapes and wine. These differ from the more traditional near infrared (NIR) instruments (which have been widely adopted for alcohol measurement and used by some companies for colour assessment) and UV/Visible instruments (traditional colour analysis and other analytes through the use of enzymatic kits) in the region of the electromagnetic spectra they use and how the calibrations are developed. When the instruments were introduced, they promised to revolutionise wine analysis by giving a wide range of analytical results from a single sample, with almost no sample preparation or reagents, slashing analysis costs and time. Industry uptake of these instruments has, however, been relatively limited. This has not only been due to the significant upfront and ongoing maintenance costs, but also because the user experience and perceptions about the accuracy and ease of use of the instruments have varied significantly.

In an attempt to share experiences and improve the understanding and use of these instruments, a number of users from within the Australian and New Zealand wine industries, in conjunction with the Interwinery Analysis Group (IWAG), have formed an informal Wine Industry MIR User Group. As a first step, the group has reviewed the analytical results from the 2013 rounds of IWAG proficiency testing and done a survey of industry users to get an indication of analytical performance, how the instruments are being used, and the general levels of satisfaction of users within the wine industry. The goal for these activities is to ultimately try to separate MIR fact from fiction.

DIVIDED OPINIONS

The online survey garnered 28 responses, with industry members indicating instruments used in the wine sector are dominated by those produced by the FOSS Company, based in Denmark (12 model FT120s, six model FT2s and eight OenoFoss). However, there were also a number of users of the recently introduced Bruker Alpha. This does not represent all the instruments in place in the Australian and New Zealand wine industries, but it is enough to give a reasonable cross section of the industry views and attitudes.

Although often marketed as useable for juice, ferment and wine analysis, the survey found that the most popular uses for MIR instruments was in process monitoring and making production decisions. All the instruments surveyed were used for wine analysis, with a lower proportion (especially amongst the more expensive FT120 and FT2 units) used for grape analysis and only a few of the latter units used for ferment monitoring. This is probably a function of the difficulty experienced in presenting consistently homogenous samples to the instruments, considering the turbid and gaseous nature of ferments.

The OenoFoss was used at all points of production from grape analysis to packaging. This could be due to the relatively low cost of the unit and the market it was aimed at - small to medium wineries with minimal wine testing capabilities. While a number of respondents said they limited the results from this instrument (which a user cannot develop custom calibrations for) to trending of wine parameters rather than absolute values, many did use it for production decisions.

When used for juice analysis, the most popular parameters tested were Brix, pH, total acidity (TA) and yeast assimilable nitrogen (YAN). Malic acid was also routinely measured by 35% of respondents, with a further 35% of respondents not using MIR instruments for juice analysis at all. In the case of ferment monitoring, 52% of respondents do not use their MIR units at all. Alcohol, TA, pH, volatile acidity (VA) and glucose/fructose (GF) were the most commonly measured analytes amongst those that did choose to use their MIR instrument for this component of production.

Unsurprisingly, finished wine products are the most commonly analysed samples on MIR instrumentation. The most common analytes to be measured on finished products are TA, alcohol, pH, GF and VA. It was interesting to see that GF ranked so highly despite the anecdotal perception of issues in developing reliable and robust calibrations for that particular analyte at levels less than 1g/L, which are common in many Australian wines.

Just less than half of all respondents felt as though their MIR instrument met their company's needs. Those that felt it did not meet their needs cited not being able to use it to its full potential, highly inaccurate or variable results, or little or no confidence in results. Happy users cited convenience and quick analysis times as being selling points. Half of all respondents thought that the MIR instrument was worth the capital investment, with those who detailed their reasoning remarking on the return on investment in regards to not needing to hire extra staff, not needing to spend as much money on reagents such as enzyme kits, and saving

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time. Those who felt it failed to live up to expectations cited a distinct lack of knowledge in developing and maintaining calibrations, and the costs required to maintain and service the instruments.

Greater than half of all respondents did not rate the supplier or manufacturer support highly. The trend appeared to be that support from the supplier immediately upon purchasing a unit was great, but trailed off after six to 12 months. However, when users did deal with the supplier or manufacturer, support received was always excellent. Many respondents stated they would like to see better calibration support from the supplier or manufacturer.

Interestingly, 12.5% of users utilise manufacturers' calibrations directly and do not adjust them prior to routine use. This is despite the fact that the majority of these calibrations are developed in Europe with only a limited amount of data from Australian wines. Also, these 'out of the box' calibrations do not take into account the effects of an individual laboratory's biases. It is these differences between the individual laboratory's results and the generic result that drives much of the perceived need for adjustments with these (and many other) techniques. Greater than 50% of users adjust calibration slope and bias at least once before being used, while 33% develop their own unique calibrations. Nearly all respondents felt they understood the concepts behind calibrations of MIR instruments.

Table 1. Comparative 2013 red wine results for MIR and the most popular method based on IWAG submissions. Results considered significant different based on a 95% confidence level are highlighted in red.

	MIR		Reference		
analyte	mean	Standard Deviation	mean	Standard Deviation	Reference method
pH	3.50	0.05	3.53	0.05	pH meter
TA	6.28	0.71	6.53	0.24	autotitrator
Alcohol	13.61	0.16	13.60	0.10	NIR
Glucose/fructose	3.27	1.03	2.40	0.25	enzymatic
Acetic acid	0.42	0.06	0.42	0.05	enzymatic
Malic acid	0.17	0.15	0.10	0.16	enzymatic
Specific gravity	0.9950	0.0008	0.9950	0.0010	Density meter
Dissolved CO_2	0.21	0.09	0.21	0.04	Thermal cond.

Table 2. Comparative 2013 white wine results for MIR and the most popular method based on IWAG submissions. Results considered significant different based on a 95% confidence level are highlighted in red.

	MIR		Reference		
analyte	mean	Standard Deviation	mean	Standard Deviation	Reference method
pH	3.31	0.05	3.35	0.05	pH meter
TA	6.53	0.78	6.73	0.28	autotitrator
Alcohol	12.73	0.18	12.77	0.10	NIR
Glucose/fructose	5.13	0.84	5.13	0.83	enzymatic
Acetic acid	0.25	0.08	0.18	0.05	enzymatic
Malic acid	3.02	0.71	2.95	0.50	enzymatic
Specific gravity	0.9931	0.0006	0.9933	0.0011	Density meter
Dissolved CO_2	1.02	0.18	1.00	0.03	MVE

When asked how calibration performance is monitored, responses from members were varied. Most wineries utilised good laboratory practice standards when monitoring performance. Another popular method was to crosscheck results from MIR instruments with reference method results. Some crosschecked their results with reference methods from other, often NATA accredited laboratories. Many respondents did not indicate they check instrument carryover between samples (for FT120 and FT2) or repeatability between measurements.

When asked if they felt they received adequate internal senior management support with regards to the time it takes to develop and maintain robust calibrations, only 50% of respondents replied positively. For those who do not get adequate support, they felt that management do not realise the time commitment required to calibrate and maintain MIR instrumentation. Some do not even bother calibrating instruments due to time constraints.

There was overwhelming support for the concept of a user group, with all respondents indicating they would find an MIR user's group particularly beneficial to their company and would consider becoming members. Many would welcome the opportunity to share information with, and seek support from other members.

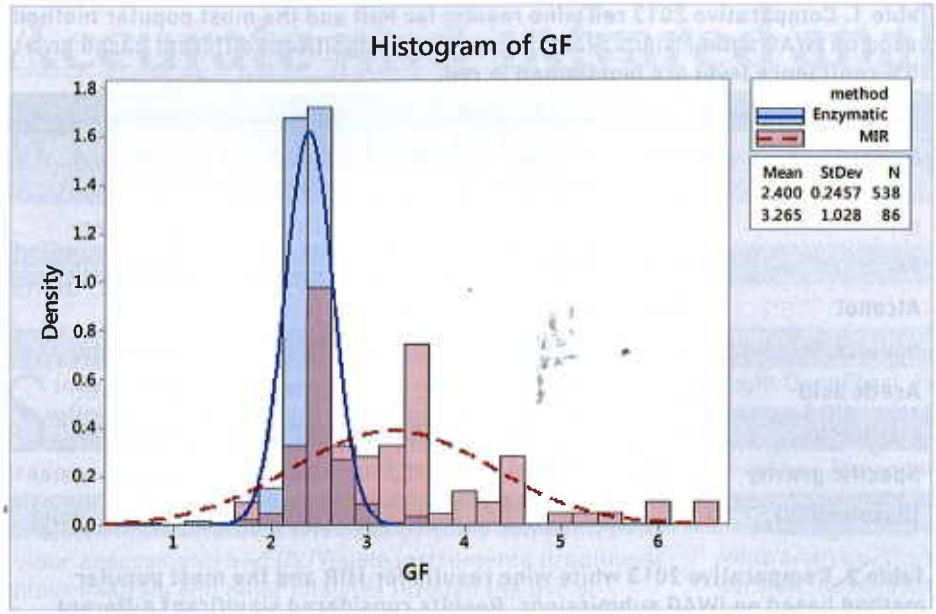

Figure 1. Comparison of glucose/fructose results for MIR and enzymatic methods in red wine showing the bi-modal distribution MIR results.

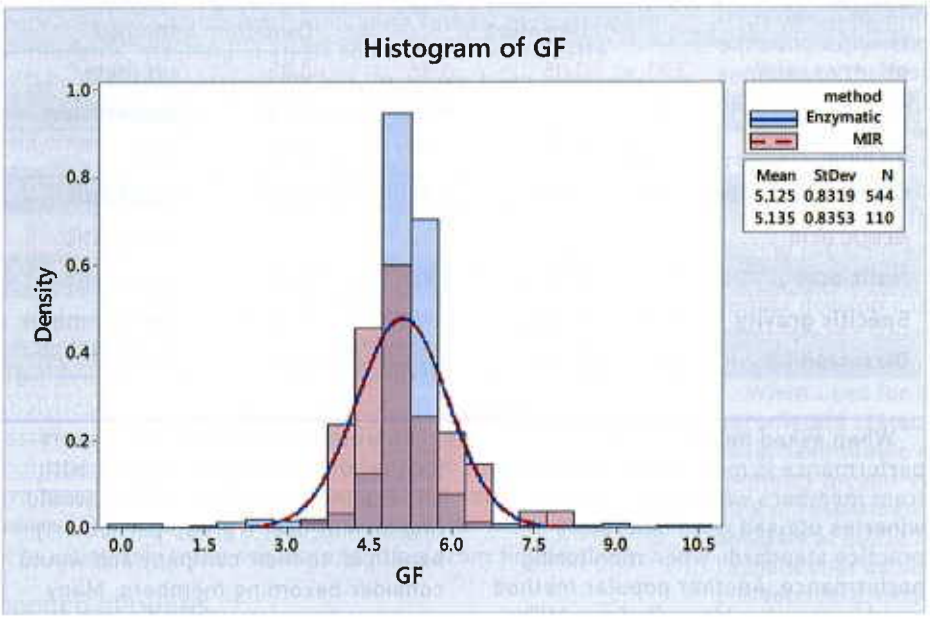

Figure 2. Comparison of glucose/fructose results for MIR and enzymatic methods in white wine showing the high similarity in results.

BUT HOW DO THE NUMBERS STACK UP?

The IWAG proficiency testing program involves six rounds of testing per year - three white rounds and three red rounds - with two bottles of the same wine tested each round. Comparing the performance of the wineries that submitted MIR results against those using other analytical methods gives us some useful insights into the overall performance in industry of these instruments for the analysis of a red and a white wine over the period of a year. Tables 1 and 2, see page 65, set out the means and standard deviations for MIR versus the next most popular methodology for analyses where sample numbers made comparison statistically viable.

The results for both red and white wines for alcohol, specific gravity and dissolved carbon dioxide show no significant difference between MIR methods and the next most popular method based on a 95% confidence level. Also, the standard deviations for these analytes were comparable, and in some cases better, suggesting the variability between laboratories is similar to more traditional methods.

For the red wine, acetic acid measurement was also not significantly different; however, the values for malic acid and glucose/fructose do show significant differences for the MIR methods. In the case of malic acid, the results from both methods were approaching the limit of detection for both MIR and enzymatic methods, with the standard deviation approaching the actual analytical values. While statistically significant in practical terms, there is little real difference (0.07g/L). This is supported by the white wine result where no significant difference was found at levels around 3g/L. The red wine result for glucose/fructose was more concerning with 0.87g/L difference between results. Interestingly, the MIR results histogram (see Figure 1, page 65) shows a roughly bi-modal distribution with one peak located in the same location as the peak for the enzymatic results. This may suggest either a difference in response for different instrument models or variability in the calibrations used. In comparison, the white wine glucose/fructose results were almost identical (Figure 2, page 65), reinforcing the conclusion already made in this article, or perhaps inferring an interferent specific to the red wine used in the proficiency program.

As already stated, the malic acid and glucose/fructose results were not significantly different for the white wine compared with those obtained using traditional methods. The white wine acetic acid values did show a statistically significant result, however, the 0.07g/L difference between the results is unlikely to be of any practical significance at this level.

In red and white wines, both pH and TA showed differences that could be considered statistically significant. In the case of pH (differences of 0.03 and 0.04) the histograms show (see Figure 3 for the red example) that the distributions are very similar and that the problems could be addressed with simple bias corrections. The differences in the TA (0.25 and 0.20g/L, respectively) are due to some significant outlier values (Figure 4), either as the result of a poor calibration or difference of a small group of instruments. Removal of the outliers from this data removes any significant difference between the results.

In short, while the results for the MIR instruments used by wineries in the IWAG proficiency testing rounds do not perfectly match the results

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for the most commonly used method for all analytes, the differences are either of little practical concern or could be addressed with improved calibrations and procedures; at least for the two wines used in this study. To get a more robust understanding of the analytical accuracy of MIR instruments, analysis of a wider range of wines by a large group of wineries is required. It should also be noted that although not shown here, a number of the other traditional methods also used to analyse the same wine components performed more poorly than the MIR methods.

SO, WHAT IS NEXT?

While the analytical results do look promising, there continues to be significantly differing views as to the effectiveness of MIR instruments in the wine industry. It is apparent that the successful implementation of MIR instruments involves new protocols and skills to those traditionally implemented for wine analysis. Now that the user group has been formed and an initial understanding of the range and role of instruments in industry has been collated (as well as an assessment of their performance in the industry standard proficiency testing program), the next step planned will be to do a set of round robin testing involving upwards of 20 wines to get a detailed understanding of the issues with the current calibrations and any repeatability issues that may exist. This information will be used to:

- develop information on the best way to use the instruments
- identify any inherent weakness in certain models
- decide and act upon the best strategies to develop uniform and sustainable calibrations suitable for the Australian and New Zealand production environment.

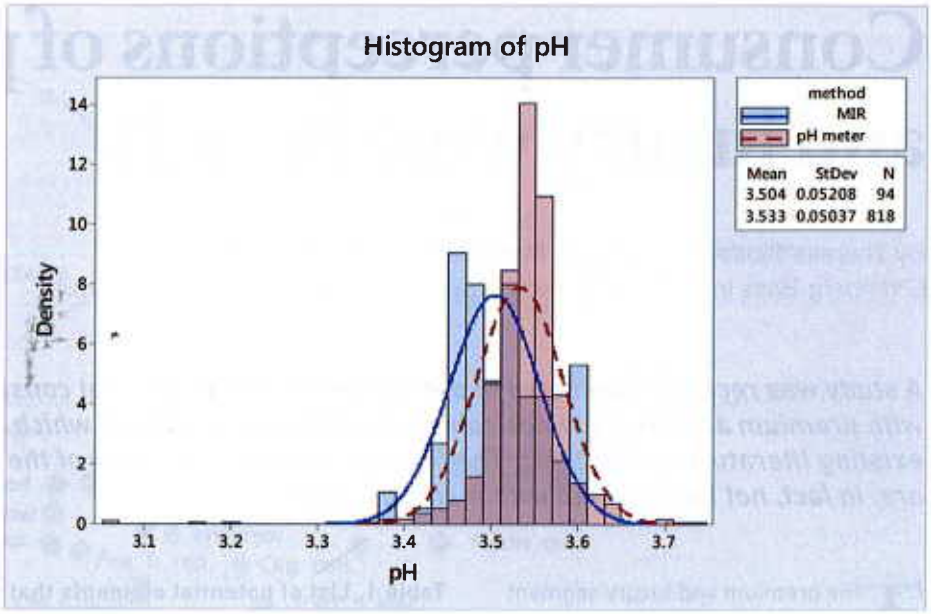

Figure 3. Comparison of pH results for MIR and enzymatic methods in red wine showing the simple bias difference.

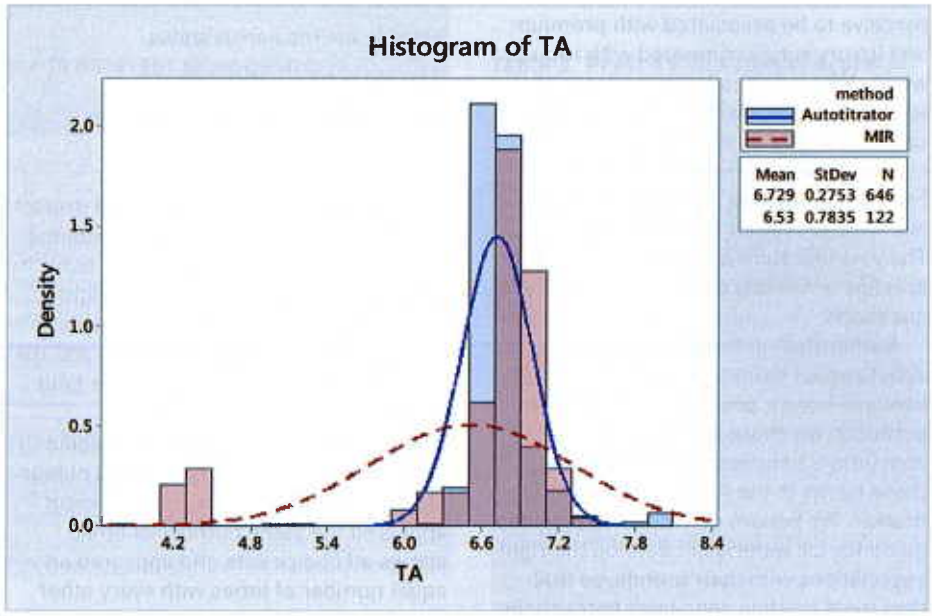

Figure 4. Comparison of TA results for MIR and enzymatic methods in white wine showing the impact of a limited number of outliers.

The wine industry MIR user group welcomes all interested participants. For further information contact Eric Wilkes (eric.wilkes@awri.com.au) or Luke Warner (chairperson@interwinery.com.au)

The Interwinery Analysis Group (IWAG) is a proficiency testing provider that supports wineries and wine testing facilities in Australia and internationally. IWAG's proficiency program is a quality management tool that gives technicians, winemakers and customers confidence in laboratory results. The program involves members concurrently analysing duplicate wine samples, allowing laboratories to compare their analytical results with those submitted by other laboratories and identify questionable results.

For further information visit www.interwinery.com.au