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Raman Spectroscopy

Quality control of hand sanitizer gels and 70% alcohol products using Raman spectroscopy

Application Note

Pharmaceuticals RA87

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Abstract: The use of hand sanitizer gels has become much more prevalent since the outbreak of the COVID-19 pandemic, and more generally, 70% alcohol products have had widespread use for centuries in medical treatments such as wound disinfection and antiseptics. To remain effective, the alcohol concentration in these products must not be below a defined threshold and needs to be controlled. This application note demonstrates that Raman spectroscopy is an efficient solution to evaluate alcohol concentrations in quality control processes.

Keywords: Raman spectroscopy, hydroalcoholic gels, 70% alcohol, MVAplus multivariate analysis, MultiWell application.

Introduction

This application note focusses on the analysis of two common pharmaceutical products that we all have in our own medicine cabinets: local antiseptics containing 70% alcohol and hand sanitizer gels that have grown in use in a time of global pandemics. The World Health Organization (WHO) recommends that a product formulation for hand disinfection contains either 80% v/v ethanol or 75% v/v isopropanol. Products for this use are no longer effective below 60% v/v alcohol, making it clear that this alcohol concentration has major consequences for public health and must be closely monitored.

Here, we present how Raman spectroscopy can be used to determine alcohol concentrations of solutions using the LabSpec 6 MVAPlus multivariate analysis platform. Combining this with the MultiWell module from HORIBA Scientific, we show how this can be performed in an automated analytical workflow.

Raman spectroscopy is based on the inelastic scattering that occurs during the interaction of light with the chemical bonds within a material. The Raman effect allows for the non-destructive chemical analysis of solids, powders, liquids, and gases, thereby allowing further analysis of the samples by other analytical techniques.

The reference technique for alcohol quantification is GC-FID (gas chromatography with flame-ionization detection) which requires a gas supply, and where data acquisition/retention times for ethanol is on the order of several minutes. The Raman measurements of the two alcohols studied here (ethanol and isopropanol) were acquired over several seconds. Moreover, HORIBA Raman systems can be used, not only to control the alcohol content of hand sanitizer gels, but also to carry out a more in-depth study of the material by, for example, analyzing the possible presence of microplastics inside the sample (1).

Materials and methods

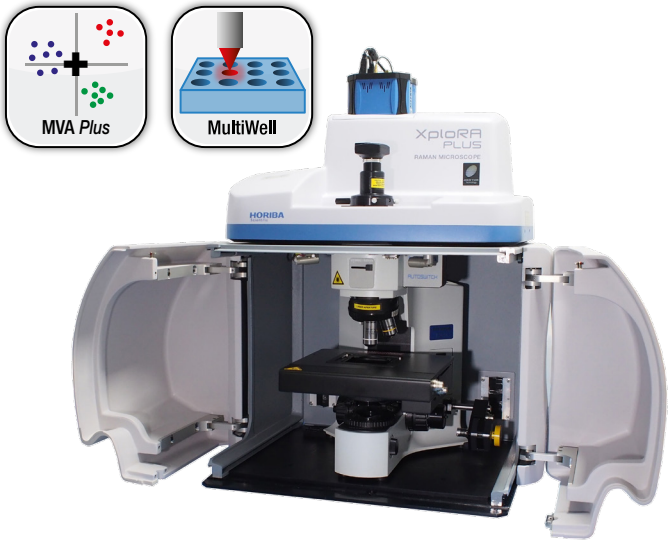

Figure 1: XploRA™ PLUS system and LabSpec 6 applications used (MVAPlus and MultiWell)

This study was conducted using a XploRA™ PLUS equipped with a 532 nm laser and a 600 g/mm grating, combining the full functionality of a microscope with high-performance Raman spectroscopy. The easy-to-use XploRA™ PLUS system is ideal for research and analytical laboratories. The XploRA™ PLUS was integrated with a 96-microwell plate in combination with the LabSpec 6 MultiWell acquisition module which enables automated measurements thanks to automated movement of the large sample stage and the inbuilt autofocus system. Each well of the microplate was filled with a sample volume of 200 µL allowing Raman measurements

with a low volume of sample. Standard alcohol solutions of ethanol and isopropanol were prepared at seven different concentrations by serial dilution from a stock solution at 100% v/v (0, 12.5, 25, 37.5, 50, 62.5, 75 and 100% v/v). Two commercial hand sanitizer gels and a 70% v/v alcohol were analyzed as supplied. They were measured with and without addition of ethanol standard at 20% v/v for result accuracy and all samples were measured in triplicate to ensure repeatability. All data were acquired by the LabSpec6 software and the integrated MVAPlus application was used for the data analysis. MVAPlus is a powerful multivariate analysis tool: up to 4,000,000 of spectra can be analyzed thanks to multiple algorithms. Using MVAPlus, a reliable distribution of the major compounds of a sample is obtained.

Results and discussion

Standard samples of alcohols at different concentrations

The two alcohols studied, ethanol and isopropanol, were prepared at eight concentrations and deposited in triplicate in a standard 96-well microplate. Spectra obtained for the 48 samples were acquired automatically and sequentially using MultiWell acquisition module of LabSpec 6 software.

The obtained spectra were then analyzed using multivariate analysis based on a classical least squares (CLS) method. The CLS method uses a supervized algorithm: reference spectra (Figure 2 B) with ethanol in red, isopropanol in green and water in blue) are known by the user. Identification of reference spectra was confirmed by the HORIBA KnowItAll® database provided with LabSpec 6 software and powered by Wiley Science Solutions. CLS fitting results are represented in Figure 2 A) where each spot corresponds to a single well and the colour corresponds to the component distribution depending on the alcohol concentration, where an increasing red component corresponds to ethanol and green to isopropanol. Results of the CLS fitting are named scores and are expressed in percentages of the reference component contribution. The scores obtained for the measured alcohol concentration were correlated to the expected concentrations for each solution (Figure 2 C) and D) respectively) leading to range curves. The scores values used are mean values of the three replicates. R2 coefficients obtained for the calibration curves were of 0.9938 and 0.9925 for ethanol and isopropanol respectively. This shows a high correlation was obtained for the two alcohols demonstrating the efficiency of CLS algorithm.

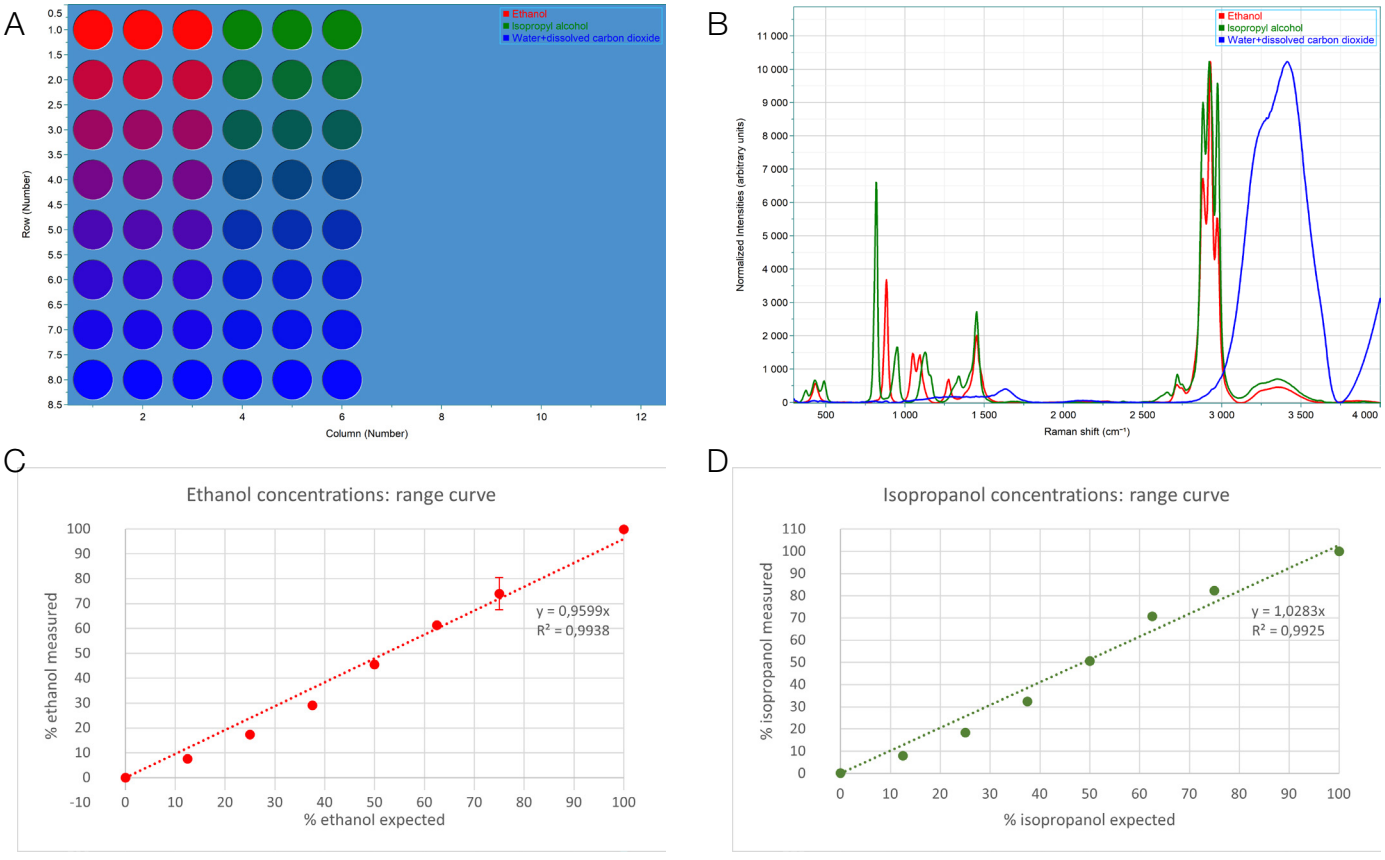

*Figure 2: A) MultiWell display results using multivariate analysis; B) Reference spectra for multivariate analysis (CLS method); C) Range curve for ethanol and D) Range curve for isopropanol. **Red:** ethanol, **Green:** isopropanol, **Blue:** water.*

Applications to common pharmaceutical products: hydroalcoholic gels and 70% alcohol

Since the large increase in hand sanitizer gel production during the COVID-19 epidemic crisis, the Food and Drug Administration (FDA) now recommends the following composition, which has been approved by the World Health Organization:

- 80% v/v ethanol or 75% v/v isopropanol.
- 1.45% v/v glycerine.
- 0.125% v/v hydrogen peroxide.
- q.s. sterile or distilled water.

In any case, the alcohol content must be at least of 60% v/v regardless of the type of alcohol to have real effectiveness.

To monitor this in commercial products, two different hydroalcoholic gels were analyzed by Raman spectroscopy with a further 70% alcohol pharmaceutical product added to this analysis. Each sample was measured in triplicate. Figure 3 shows obtained spectra and the measured data were analyzed using CLS multivariate analysis.

For the 9 spectra, contribution scores obtained for ethanol were 70% compared to those obtained for isopropanol, which were below 5% meaning that the three products can be identified as being formulated from ethanol. CLS contribution scores below 5% are not considered as significant. Contribution scores of glycerin were also below 5%. As the concentration of glycerin is very low in the preparation of hand sanitizer gels, it could not be evaluated using the CLS Raman analysis described here. Contribution scores obtained for ethanol and water, the main components, of the three pharmaceutical products are indicated in Table 1.

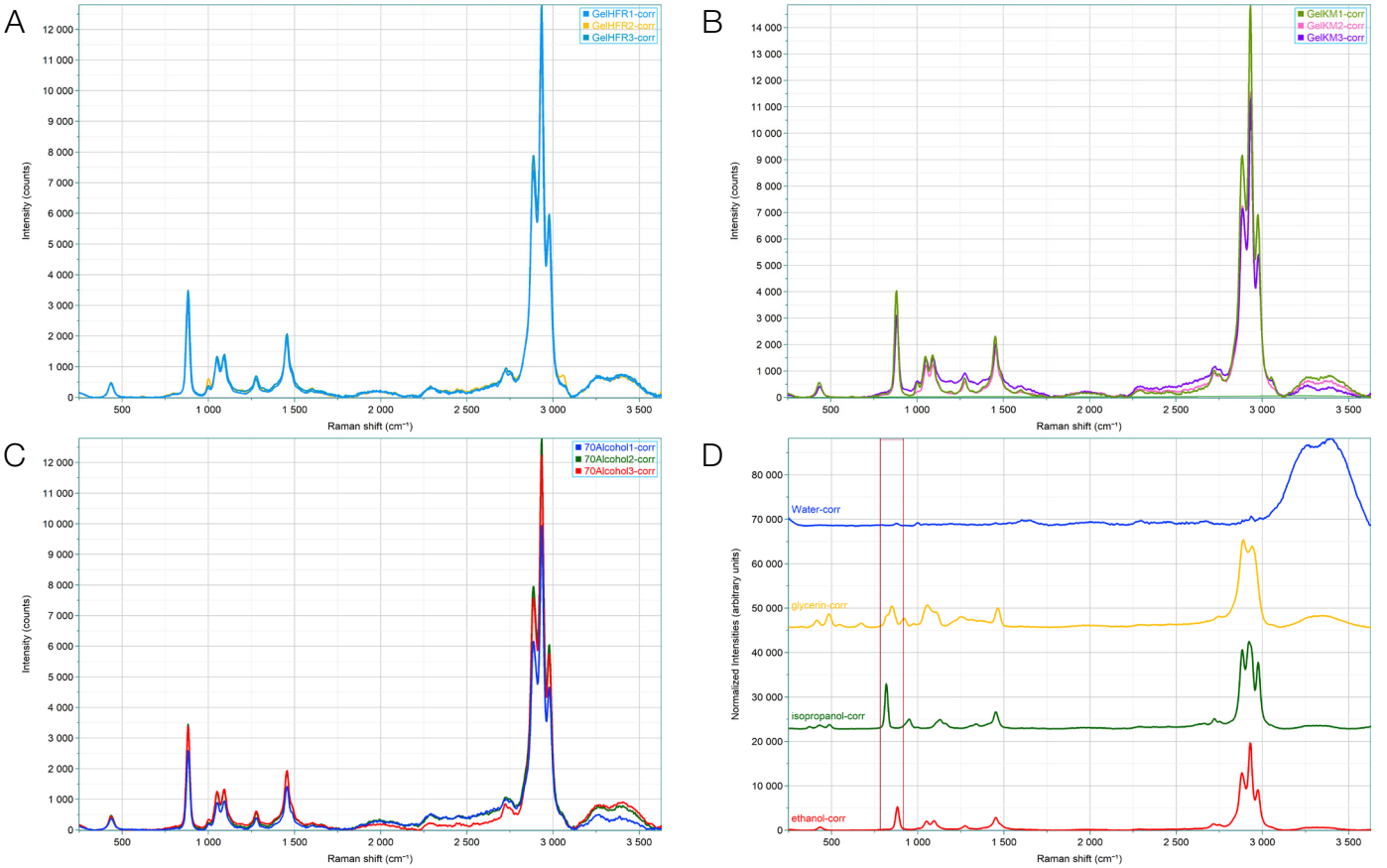

*Figure 3: Processed and superimposed Raman spectra of the 3 replicates for: A) hydroalcoholic gel 1; B) hydroalcoholic gel 2 and C) 70% alcohol. D) Reference spectra used for multivariate analysis (CLS method) with ethanol in **red**, isopropanol in **green**, glycerin in **gold** and water in **blue**.*

Table 1: Contribution scores (%) of ethanol and water for three pharmaceutical products using CLS multivariate analysis

Hydroalcoholic gel 1					
	Replicate 1	Replicate 2	Replicate 3	Mean	SD
Ethanol	89.42	85.77	86.52	87.24	1.93
Water	9.98	11.87	10.39	10.75	0.99

Hydroalcoholic gel 2					
	Replicate 1	Replicate 2	Replicate 3	Mean	SD
Ethanol	91.93	87.44	79.93	86.43	6.06
Water	9.79	7.56	2.88	6.74	3.53

70% alcohol					
	Replicate 1	Replicate 2	Replicate 3	Mean	SD
Ethanol	83.99	84.07	85.88	84.65	1.07
Water	5.40	9.65	13.73	9.59	4.17

Conclusion

Using Raman spectroscopy, two different types of pharmaceutical products, hand sanitizer gels and 70% alcohol disinfectant, were tested. This was possible by determining their alcohol concentrations, thanks to the powerful MVAPlus multivariate analysis tool by HORIBA Scientific. Quality control of these pharmaceutical products using a Raman-based technique is very simple: samples require no preparation and there is no need for a molecular labelling.

HORIBA Raman systems are compatible with automated measurements allowing high throughput experiments such as industrial quality control processes. Standard microwell plates can be used with MultiWell module of the LabSpec 6 software to acquire spectra from hundreds of samples sequentially. As an example, with 96-microwell plates, spectra acquisition of almost one hundred solutions samples is easy using a low sample volume of only 200 µL.

References

1. Application Note RA86: “Analysis of microplastics in hand sanitizers using Particle Finder™” Hajar ELAZRI, Thibault BRULE, Massimiliano ROCCHIA

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