Rapid Raw Material Identification for Formulation Compounds Using Handheld Raman Technology

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Ever since the European Medicines Agency (EMA) recommended that all pharmaceutical companies test every raw material that passes through their manufacturing facilities, this practice has attracted great interest in the nutraceuticals, food manufacturing, cosmetics, and agricultural industries, pushing them to strive for more rigorous and technology-oriented quality control standards. In recent years, handheld Raman technology has gained a noticeable market in raw materials identification in various industries where traditional analytical techniques like HPLC and NIR spectroscopy have been the primary technologies. The surge of handheld Raman instrumentation is mainly due to advancements in Raman technology such as small lasers and detectors with good sensitivity and at a lower price that has led to faster, smaller, more rugged, and less expensive analytical tools that require minimum training for use. The fact that Raman technology requires no sample preparation, no direct contact with the sample, and has the capability to test a sample through transparent packing material such as glass or plastic has made it an ideal tool for rapid raw material identification.

In this study, four typical ingredients used as excipients in the food industry and for pharmaceutical and nutraceutical products were tested for raw material identification: whey, sorbitol, stearic acid, and calcium phosphate dihydrate dibasic. The data presented in this study were collected using a NanoRam® handheld Raman spectrometer from B&W Tek. With a 785nm wavelength laser excitation source, the NanoRam is a compact, handheld Raman spectrometer with an integrated computing system for material identification and verification. Using intelligent principal component analysis (PCA) model building software and on-board spectral libraries, the NanoRam allows for the rapid development of reliable methods for identification and verification of raw materials. Identification of an unknown compound is achieved in a very short timespan (typically less than 30 seconds), making it a practical choice for rapid identification and verification purposes.

The raw material identification test on the NanoRam involves firstly creating a method for the raw material to be tested. A minimum of 20 spectra are acquired for each material to establish a PCA model for the material that represents the natural variability of the sample, as multiple lots of material can be used for the 20 spectra. PCA is a multivariate technique which reduces the dimensionality of the data set by finding an alternative set of coordinates (i.e. the principal components) that explain the structured variance in the data. After the method is created for the material, the spectrum of a sample measured with the Nanoram will be compared with the method associated with the PCA model to see if it falls within the 95% confidence limit of the model space. A statistical p-value will be calculated to give a “Pass” or “Fail” result based on the level of similarity of the unknown material compared to the method on a 95% confidence level. If the p-value is greater than or equal to 0.05, a

"Pass" result will be returned. If the p-value is lower than 0.05, a "Fail" result will be given, indicating that the sample spectrum falls outside of the method limits.

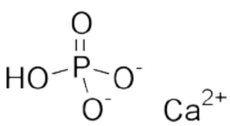
Experiment

The NanoRam with a point and shoot sampling accessory was used for testing. The samples were measured through a Whirl-Pak transparent plastic bag.

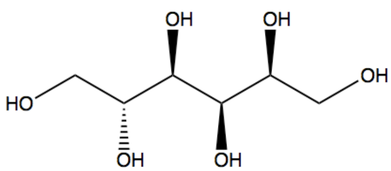
Test Results

The four raw materials included in this test are compounds used in the food industry, nutraceutical industry, cosmetic industry, as well as pharmaceutical industry: calcium phosphate dihydrate dibasic; sorbitol; stearic acid; and whey protein, which is a mixture of beta-lactoglobulin (~65%), alpha-lactalbumin (~25%), bovine serum albumin (~8%), and immunoglobulins[1].

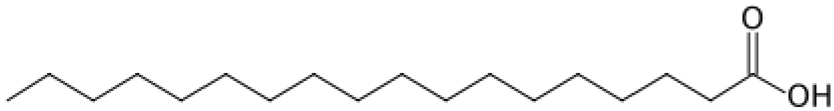
Calcium phosphate dihydrate dibasic:



Sorbitol:



Stearic acid:



The overlaid spectra of calcium phosphate dihydrate dibasic ($CaHPO_4 \cdot 2H_2O$), sorbitol ($C_6H_{14}O_6$), stearic acid ($CH_3(CH_2)_{16}CO_2H$), and whey are shown in Figure 1. The compounds all have distinct Raman spectra, though some spectral overlap can be seen.

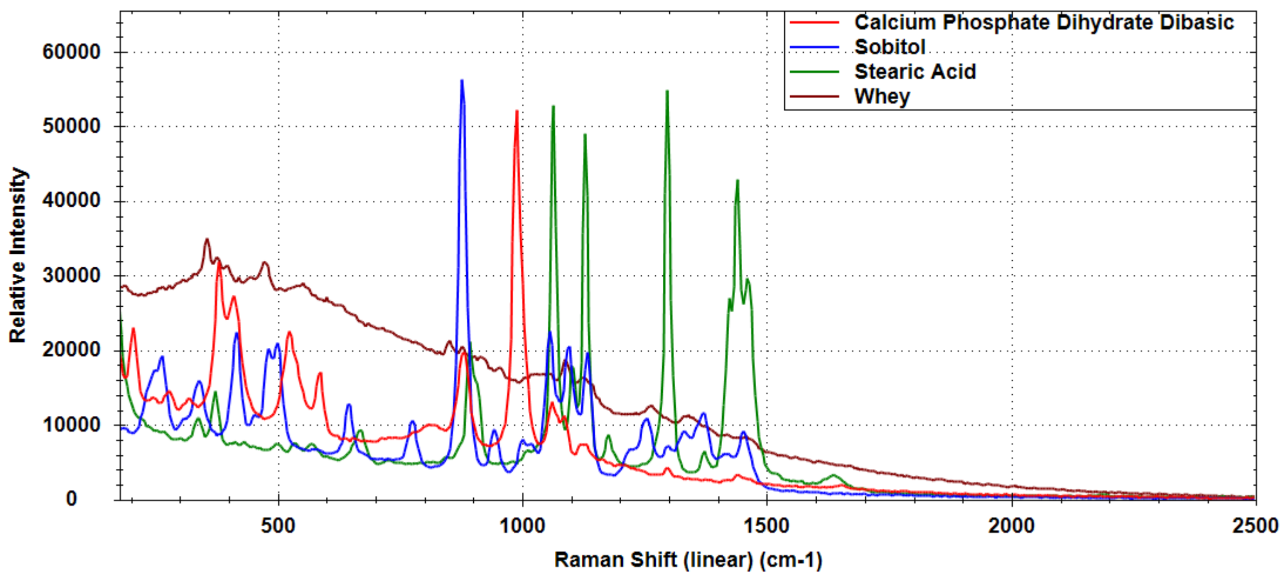


Figure 1. Raman spectra of calcium phosphate dihydrate dibasic, sorbitol, stearic acid, and whey

The methods "Whey", "Calcium Phosphate Dihydrate Dibasic", "Sorbitol", and "Stearic Acid" were created on the NanoRam. The significance level for passing is set at 0.05 for all methods.

All four materials passed against their own methods, which is demonstrated in the diagonal line shown in Table 1. All materials failed the methods for the other materials, respectively, providing a clear differentiation to separate the four types of materials.

Method / Sample	Whey	Calcium Phosphate Dihydrate Dibasic	Stearic Acid	Sorbitol
Whey	PASS (p=1)	FAIL (p=0)	FAIL (p=0)	FAIL (p=0)
Calcium Phosphate Dihydrate Dibasic	FAIL (p=4.292E-08)	PASS (p=0.9919)	FAIL (p=0)	FAIL (p=0)
Stearic Acid	FAIL (p=2.819E-07)	FAIL (p=0)	PASS (p=0.5351)	FAIL (p=0)
Sorbitol	FAIL (p=3.01E-09)	FAIL (p=1.321E-14)	FAIL (p=0)	PASS (p=0.9998)

Table 1.

The "Pass" test result and corresponding p-value is shown in Figure 2 for whey; Figure 3 for calcium phosphate dihydrate dibasic; Figure 4 for stearic acid; and Figure 5 for sorbitol.

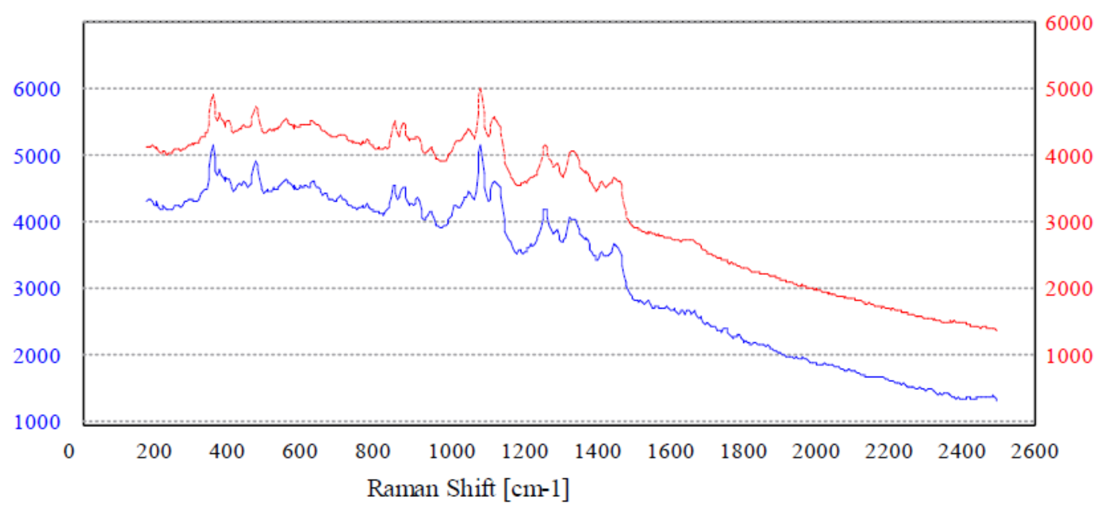


Figure 2. Material whey passed the method "Whey"

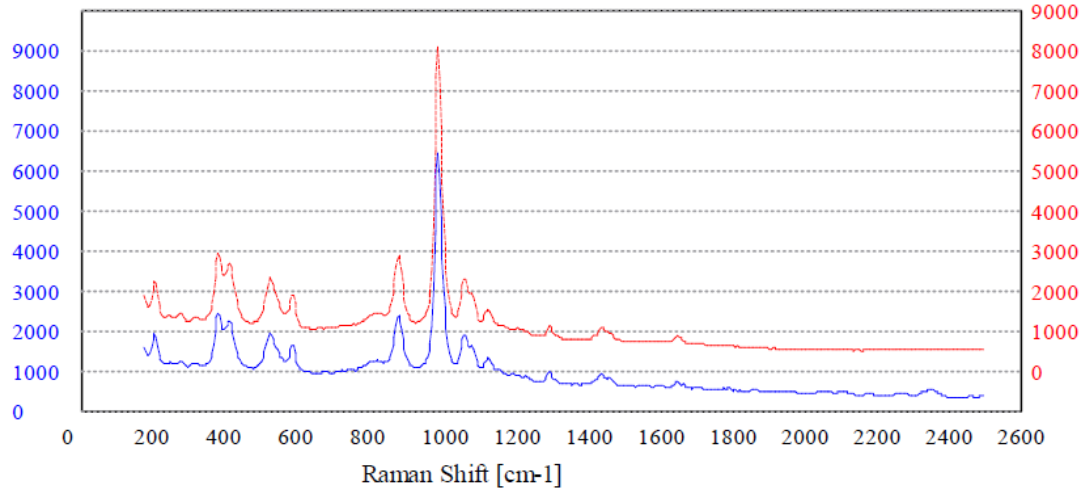


Figure 3. Material calcium phosphate dihydrate dibasic passed the method "Calcium Phosphate Dihydrate Dibasic"

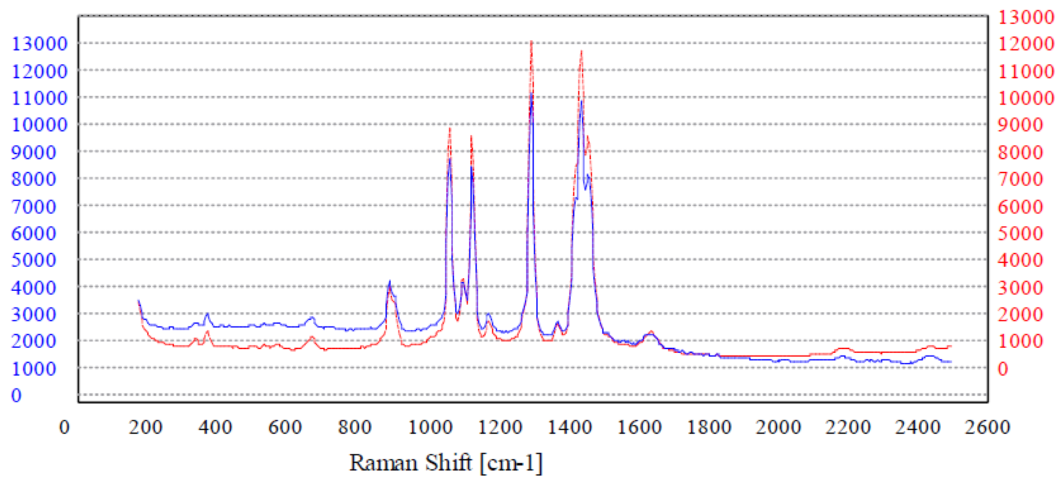


Figure 4. Material stearic acid passed the method "Stearic Acid"

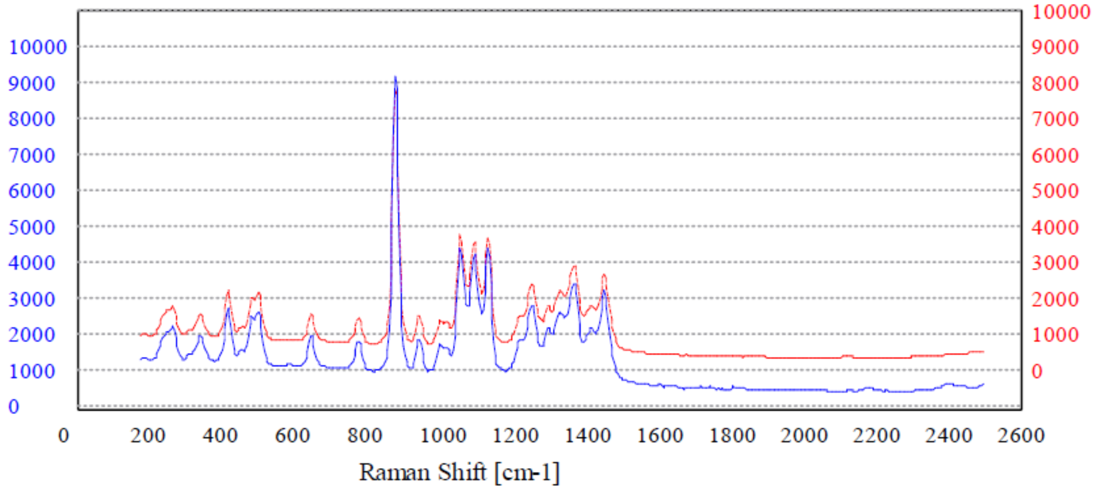


Figure 5. Material sorbitol passed the method "Sorbitol"

Conclusions

The raw materials whey, sorbitol, stearic acid, and calcium phosphate dihydrate dibasic all show very distinctive, unique Raman signatures, which indicates that Raman spectroscopy is the ideal technology for identification of these materials. The PCA model-based method provides reliable specificity to successfully identify these materials using the methods built on the NanoRam handheld spectrometer.

References

1. [1]. Haug A, Høstmark AT, Harstad OM, A; Høstmark, AT; Harstad, OM (25 September 2007). "Bovine milk in human nutrition – a review". Lipids Health Dis 6: 25. doi:10.1186/1476-511X-6-25. PMC 2039733. PMID 17894873.