



Application Note AN-RS-014

Trace Detection of Rhodamine B in Cayenne Powder

Protecting consumer safety with Misa

The addition of dyes to provide uniform coloration and enhance visual appeal in food products is a common practice. Rhodamine B is a dye utilized extensively in biotechnology and industrial applications and is one of several colorants banned for use as food additives in Europe and North America. The most common analytical methods for detection of illicit dyes in food products, GC/MS and HPLC, are laboratory-based instrumental methods that require

specialized training. With Misa (Metrohm Instant SERS Analyzer), detection of trace amounts of Rhodamine B in ground cayenne pepper is quick and easy after a facile extraction procedure with minimal material consumption. Rhodamine B can be detected in cayenne powder at a concentration of 50 $\mu\text{g/g}$. However, a simple concentration step improves that limit to 10 $\mu\text{g/g}$.

INTRODUCTION

Ground cayenne pepper bought commercially was doped with Rhodamine B (RhB) and tested

with Misa to simulate a realistic food screening scenario.

REFERENCE MATERIAL AND LIBRARY CREATION

To establish a reference spectrum, a pure RhB standard (50 $\mu\text{g/g}$ in ultrapure water) was analyzed using gold nanoparticles (Au NPs). The unique SERS spectrum shown in **Figure 1** can be used to create a library entry for RhB.

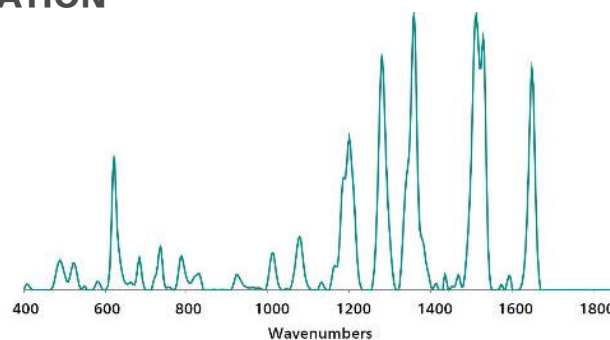


Figure 1. Gold NP SERS standard reference spectrum of Rhodamine B.

EXPERIMENT

A stock solution of RhB in methanol was prepared. Purchased cayenne powder was treated with serial dilutions of the stock (3 mL stock to 1 g cayenne) to yield samples with 1000, 500, 100, 50, 10, 5, and 1 $\mu\text{g/g}$ of RhB. Samples were thoroughly mixed and air-dried. To prepare extracts, 0.1 g of each spiked sample was added to a vial with 400 μL of methanol, shaken to mix, and left to settle for 10 minutes. To prepare test samples, 50 μL of the methanol extract was pipetted into a vial with 400 μL of Au NP solution and 50 μL of 0.5 mol/L salt solution. The vial was shaken to mix, and then placed into the vial attachment on Misa for testing.



Table 1. Experimental Parameters

Instrument		Acquisition	
Firmware	0.9.33	Laser Power	5
Software	Misa Cal V1.0.15	Int. Time	10 s
Misa Vial Attachment	6.07505.040	Averages	10
ID Kit - Au NP	6.07506.440	Raster	ON

RESULTS

In **Figure 2**, overlaid spectra of RhB indicate detection down to 50 $\mu\text{g/g}$. For each concentration tested, the baseline spectrum

from unadulterated cayenne was subtracted from the average of baseline-corrected, replicate measurements.

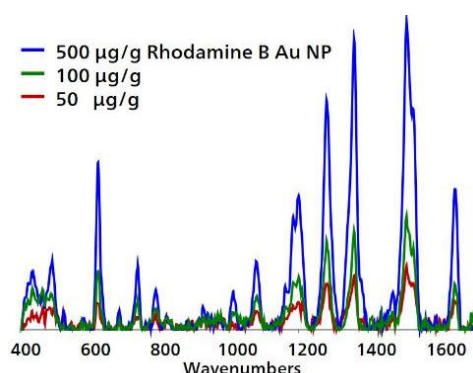


Figure 2. Gold NP SERS concentration profile of RhB extracted from adulterated cayenne powder. Spectra are baselined, with Au NP and control subtracted.

To improve trace detection and spectral signal-to-noise, a very simple concentration method was applied to each extract. All extracts were fully air-dried, then resuspended in methanol to

yield a 5x increase in concentration. The spectra in **Figure 3** demonstrate detection of RhB down to 10 $\mu\text{g/g}$.

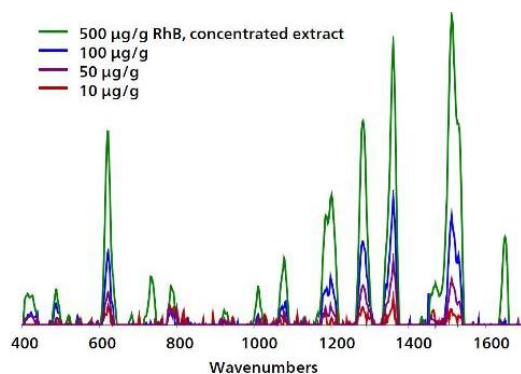


Figure 3. RhB profile after 5x concentration.

FIELD TEST PROTOCOL

Detection of Rhodamine B in the field

Using the large end of the scoop, add 3–4 scoops of sample to a 2 mL vial. Add methanol to the vial until ~1/3 full. Cap and shake the vial gently to mix, then let sample rest for 2 minutes. Fill a clean vial halfway full with Au NPs. Using

pipettes, add 1 drop each of sample solution and NaCl solution to the Au NPs, then cap and shake the vial gently to mix. Insert into vial attachment on Misa for measurement.

Table 2. Requirements for Field Test Protocol

ID Kit - Au NP	6.07506.440
includes:	Gold nanoparticles (Au NP)
	Scoop
	Disposable pipettes
	2 mL glass vials
Reagents	
Methanol	
NaCl solution	3 g NaCl in 100 mL water
Test settings	Use ID Kit OP on MISA

CONCLUSION

Trace levels of detection, ease of sample preparation, and rapid assay times collectively recommend Misa as a reliable, cost-effective

solution for high-throughput, on-site identification of adulterated food products.

CONTACT

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CONFIGURATION



MISA Advanced

Metrohm Instant SERS Analyzer (MISA) 是一款高性能、便携式分析系,可快速/定非法物、食品添加和微量食品染料。MISA 的特点是配了 Metrohm 的道光栅描 (ORS) 技的光。其空需求小和并且池寿命有所延,用于或移室用。MISA 提供各 1 激光附件,可活取。分析可通 BlueTooth 或 USB 接行。

MISA Advanced 套件是一个完整套件,其作用是用能用 Metrohms 米粒溶液和 P-SERS 条行 SERS 分析。MISA Advanced 套件包含了一个 MISA 小管附件、一个 P-SERS-附件、一个 ASTM 校正准件、一个 USB 迷、一个 USB 供元和用于行 MISA 器的 MISA Cal 件。随供了一个用来安全保管器和附件的固保箱。



ID – Au NP

ID 套件 - Au NP 包含了 Mira/Misa 用使用体金溶液行 SERS 分析所需的件。套件包含了一个一次性抹刀、一个移液管、品小瓶和一个含金体的瓶子。