



Application Note AN-K-075

Moisture analysis of rebaudioside M by Karl Fischer titration

Simplified rebaudioside M moisture determination via volumetric Karl Fischer titration paired with an oven system

Natural sweeteners like stevia and its derivatives are an increasingly important part of the global sweetener market as more consumers substitute sugar with healthier alternatives. Stevia's derivatives are significantly sweeter than table sugar. These compounds are estimated to be 50 to 300 times sweeter in comparison. This sweet taste coupled with the fact that the human body cannot metabolize steviol glycosides (i.e., they contribute zero calories to

one's diet), makes them a very attractive sugar replacement.

The moisture content of stevia materials is a crucial factor when discussing their shelf life and stability. Measuring the moisture content of a stevia derivative, rebaudioside M, using an oven system coupled with volumetric Karl Fischer (KF) titration provided a repeatable approach for quantifying the moisture content of this material.

SAMPLE AND SAMPLE PREPARATION

Rebaudioside M powder was used for this study. No

sample preparation is required.

EXPERIMENTAL

The analysis was carried out with a Double Pt ring electrode for volumetry (6.00346.100). Testing was executed on the 874 Oven Sample Processor (2.874.0120) coupled with an 852 Titrando (2.852.0060).

Approximately 0.2 g of sample was weighed into an

874 Oven Sample Processor 6 mL vial and placed onto the rack. The oven was set to a temperature of 100 °C, and the evaporated moisture was carried to the volumetric titration vessel by nitrogen gas flow at 70 mL/min. The moisture from the sample was titrated with Hydranal Comp 5 reagent.

RESULTS

Results are recorded in Figure 1, Figure 2, and Table 1.

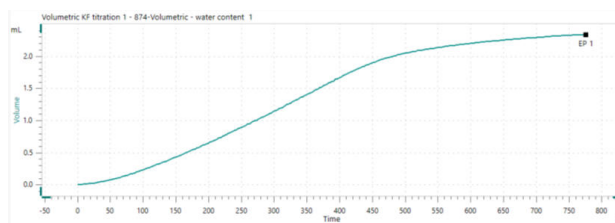


Figure 1. Example volumetric KF titration curve for rebaudioside M.

A temperature gradient was performed and demonstrated that the ideal temperature conditions for the rebaudioside M sample were in a range of ~80–210 °C (Figure 2). A temperature of 100 °C was selected to ensure that moisture was carried from the sample to the titration vessel while minimizing potential sample impact from the temperature. Samples were analyzed in triplicate to exhibit robustness of the method.

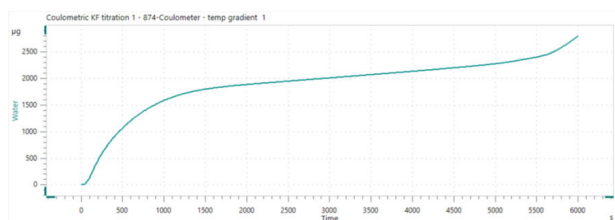


Figure 2. Temperature gradient for rebaudioside M powder demonstrating the optimal operating temperature range.

Table 1. Results for all rebaudioside M samples (n = 3) evaluated for moisture content using the 874 Oven Sample Processor coupled with volumetric KF.

Result 1 (%w/w)	Result 2 (%w/w)	Result 3 (%w/w)	Mean (%w/w)	%RSD
4.42	4.61	4.60	4.54	2.32
4.31	4.44	4.55	4.43	2.66
9.11	9.32	8.96	9.13	1.96

CONCLUSION

Natural low-calorie sweeteners like stevia derivatives are growing in popularity as people choose healthier alternatives to sugar. Working to develop standardized, specific methodology can help support sweetener industry development, effective quality assessment, and the evaluation of rebaudioside M and other steviol glycosides' stability.

This Application Note provides a foundation for the

analysis of stevia powder by Karl Fischer titration. An oven system paired with volumetric Karl Fischer analysis provides an accurate, repeatable method to evaluate stevia powder and its derivatives. This method's compatibility with the 874 Oven Sample Processor allows increased sample throughput through automation, providing operators even more time to complete other important tasks in parallel.

CONTACT

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CONFIGURATION



874 Oven Sample Processor

The 874 Oven Sample Processor is used for automatic thermal sample preparation in Karl Fischer titration. The oven method is particularly suitable for samples that do not release their water until higher temperatures have been reached, for sparingly soluble samples, or those that react with the KF reagent.



852 Titrando with Touch Control and generator electrode with diaphragm

Coulometer including generator electrode with diaphragm, Touch Control, 801 Magnetic Stirrer, and complete volumetric titration cell.

Coulometry is the ideal method for water content determination in liquids, solids, and gases when it comes to water content determination in the trace range (10 µg to 10 mg absolute water). In addition, coulometry is an absolute method and thus no titer determination is necessary.

Recommended coulometry measuring range: 10 µg - 200 mg absolute water

The **852 Titrando** controls not only coulometric, but also volumetric Karl Fischer titration.

For use with OMNIS Software, *tiamo* software, or Touch Control unit. Compliance with GMP/GLP and FDA regulations such as 21 CFR Part 11, if required.



Double Pt ring electrode for volumetry

Indicator electrode for Karl Fischer titration which, thanks to its two fused platinum rings, is more robust and easier to clean than the conventional platinum pin electrode (6.0338.100).