

# Contaminants and Degradants

The need for universal HPLC detection in analytical laboratories is widespread. While several detection technologies (e.g., low wavelength UV, refractive index, evaporative light scattering, chemiluminescent nitrogen detectors) are currently being used, there is significant room for improvement in performance characteristics such as sensitivity, dynamic range, consistency of response factors and gradient or solvent compatibility.

To help address the many challenges of universal detection, ESA has developed the Corona CAD™ detector. This novel technology offers many benefits to analytical scientists including:

- High Sensitivity - Low ng limits of detection.
- More Consistent Response Factors - Response magnitude does not significantly depend on analyte properties (e.g. molar absorptivity, proton affinity).
- Broad and Useful Dynamic Range - 4 orders of magnitude (ng - µg quantities).
- Excellent Reproducibility - Typically less than 2% RSD.
- Broad Applicability - Can be used with a wide variety of HPLC conditions to measure virtually any nonvolatile analyte including proteins, lipids, carbohydrates and small molecules.
- Ease of Use - Easy setup. Uses minimal bench space and requires only gas input pressure and signal output range to be set.

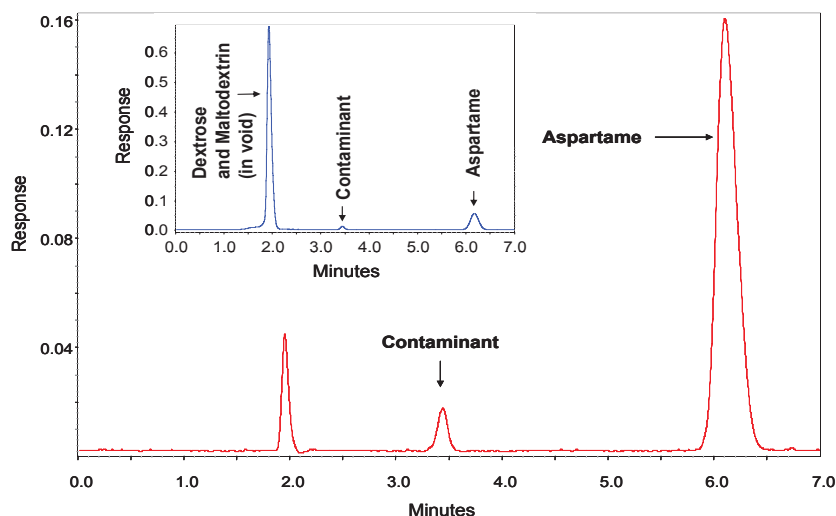


Figure 1. Detection of a Contaminant in Both the Aspartame Standard (red) and Equal® (inset).

This application note describes the use of the Corona CAD for measuring contaminants and degradants using aspartame (as both the standard and as part of the artificial sweetener, Equal®) as an example. Greater details regarding the analysis can be found in Artificial Sweeteners - 3, Equal® (70-6955).

A contaminant (eluting at ~3.5 minutes) could be found in both the aspartame standard and Equal® (Figure 1). The contaminant did not correspond to any of the known and commercially available precursors or breakdown products (Figure 2). Spiking experiments verified that the contaminant was not aspartyl-phenylalanine. After prolonged storage (2 months; freezer) a degradant formed (eluting at ~5.0 minutes) (Figure 3) but this too did not correspond to any of the analytes measured in Figure 2. This may be the diketopiperazine degradant that is known to be formed during storage, but is not commercially available.

## Corona parameters

Gas: 35psi via nitrogen generator  
Filter: none  
Range: 100pA

HPLC Parameters:  
Mobile Phase: Aqueous acetonitrile (15% v/v) with 0.05% TFA  
Flow Rate: 1.0mL/min  
Column: Shiseido C18 SG300; 4.6 x 150mm; 5µm  
Column Temperature: 30°C  
Injection Volume: 10µL

# The Corona™ Charged Aerosol Detector

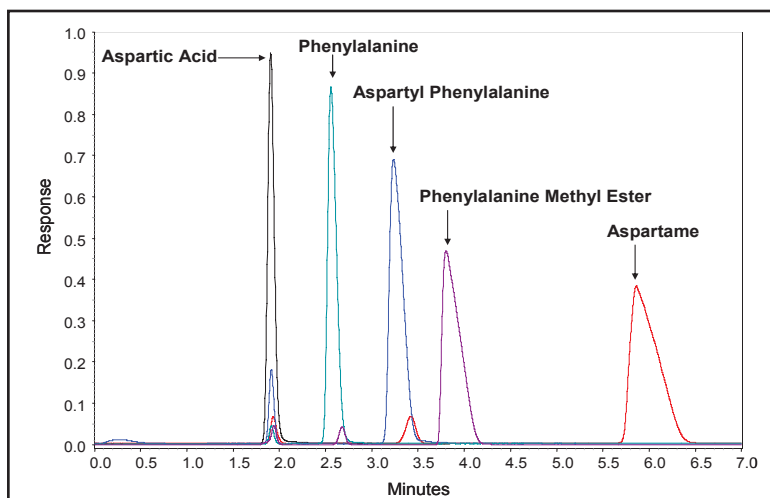


Figure 2. Aspartame and Several Possible Precursors and Degradants (10µg each, on column). Note That Many of the Standards Showed Some Degree of Contamination.

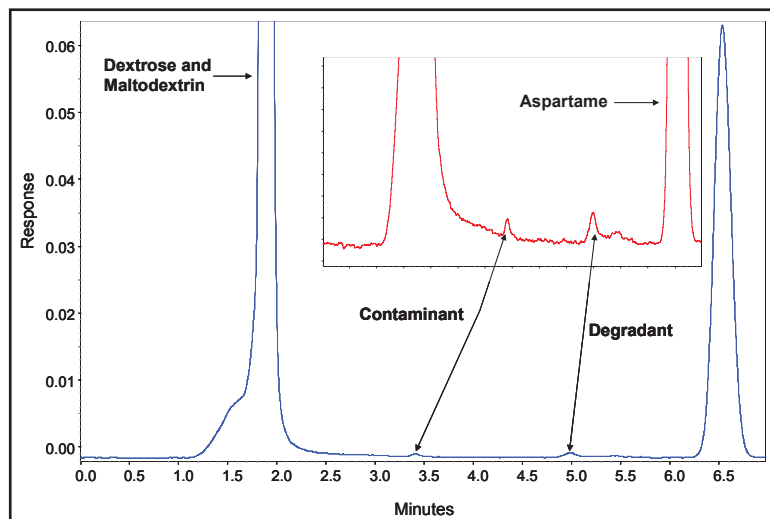


Figure 3. Detection of Contaminants and Degradants (10µg Equal® on column). Inset - at ~20X Sensitivity.

## Sample preparation

Equal® (one packet) or standards (from Supelco, Sigma and Aldrich) were dissolved in water (1mg/mL). Further dilutions were made with water.

## Conclusions

The Corona CAD provides universal detection of nonvolatile analytes with response independent of chemical properties, a wide dynamic response range, high sensitivity and good precision. These characteristics, along with reliability and simple operation, make this a superior detector for a wide range of HPLC analyses.

For more information about this application, the Corona CAD, or charged aerosol detection visit [www.coronacad.com](http://www.coronacad.com). We are interested in your opinions and are available to answer any questions you may have: please contact a technical representative at 978.250.7082, or if e-mail is more convenient, send your questions to [coronacad@esainc.com](mailto:coronacad@esainc.com).

## Ordering information

Description  
Corona

Part Number  
70-6350  
(100/120V)  
70-6351  
(230/240V)  
70-5499TA  
70-6003  
70-4050  
70-4152  
70-5073  
88-12522

Thermal Organizer Module  
Nitrogen generator  
Pump, model 582  
Autosampler, model 542  
Elite software including PC  
Column, Shiseido C18 SG300



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