

## Introduction

Mercury compounds are permitted by the FDA for use in eye makeup at concentrations up to 65 parts per million and are permitted only if no other effective and safe preservative is available for use. Other cosmetics may contain trace amounts of mercury—less than 1 part per million (0.0001 percent) [21 CFR 700.13]. Minnesota bans intentionally added mercury in mascaras, eyeliners and skin-lightening creams.

These requirements lead to a need for measuring trace amounts of mercury within a complex sample matrix. This poster explores a practical approach to analyzing total mercury with inexpensive techniques such as block digestion followed by quantitation with Cold Vapor Atomic Absorption (CVAA).

The method involves acid treatment of the samples on a block digester and analysis by cold vapor generation-atomic absorption (CVAA) detection. Cosmetics (~0.20 g) were subjected to digestion with 2 mL H<sub>2</sub>SO<sub>4</sub> and 2 mL HNO<sub>3</sub> and subsequently KMnO<sub>4</sub>. The solution was diluted to 50 mL with 3% HCl. An aliquot was reduced with a 10% SnCl<sub>2</sub> (w/v) in 7% HCl (v/v) solution and then passed to a gas-liquid separator where the evolving Hg<sup>0</sup> was swept in a stream of argon to the detector cell and the atomic absorbance signal was recorded.

## Experimental

- 10 samples each of 5 types of cosmetics, from widely-available “low-end” brands to very “high-end” brands.
- Modified U.S. EPA 245.5 “Mercury In Sediment” digestion.
- Method detection limit (MDL) performed with 2 batches of 7 fortified matrix blanks at 5 ppt and 2 ppt.
- The results were evaluated per 40 CFR 136, Appendix B.
- The analytical analysis consisted of 3 batches each having the quality controls of CCV, CCB, ICV, LFB and LRB immediately after the calibration. A CCV/CCB pair was analyzed after every 10 samples and at the end of each analytical batch. An additional LRB was analyzed at the midpoint of each batch.



Figure 1: Cosmetics Sample Set 2

## Instrumentation

Mercury Analyzer: CETAC QuickTrace™ M-7500 Cold Vapor Atomic Absorbance Mercury Analyzer.

- Working range from < 1 ppt to > 500 ppb.
- 4-channel 12-roller head peristaltic pump ensures consistent sample and reagent uptake with online reduction of the sample in a closed system.
- Includes CETAC ASX-520 autosampler.

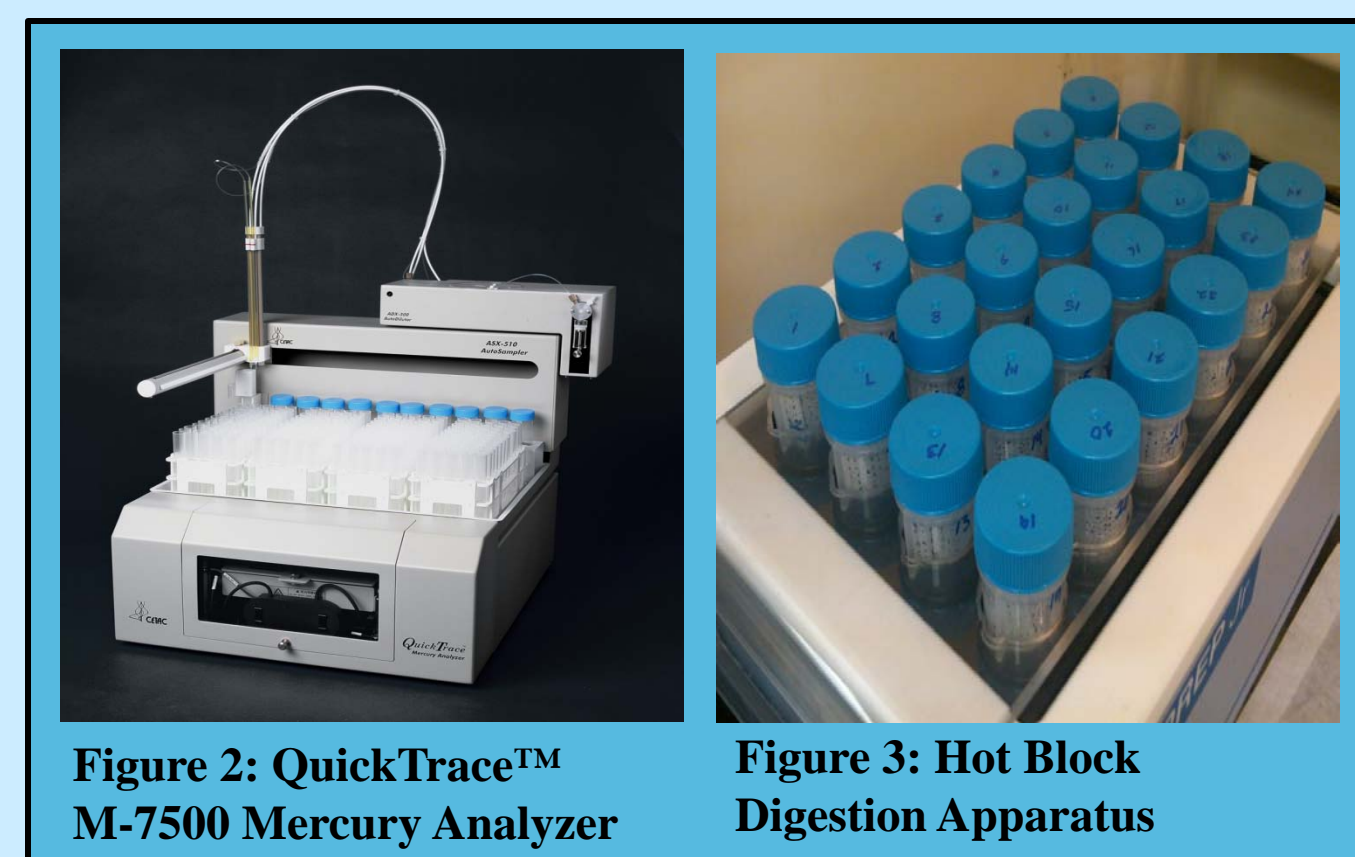


Figure 2: QuickTrace™ M-7500 Mercury Analyzer

Figure 3: Hot Block Digestion Apparatus

The reduced sample flows into the non-foaming gas-liquid separator (GLS) where ultra-high purity argon is purged through the sample and the liberated elemental mercury enters the system. Excess water vapor is removed before the sample stream passes through the sample cell where its response is measured at a wavelength of 253.652 nm.

Digestion: SCP Science DigiPREP JR which is a 24 position carbon fiber hot block digestion system.

## Reagents

- The modified EPA method 245.5 used direct additions of concentrated trace metal H<sub>2</sub>SO<sub>4</sub> and HNO<sub>3</sub>.
- 5% solution of potassium permanganate, 5g KMnO<sub>4</sub> w/v per 100 mL of DI water.
- Hydroxylamine hydrochloride solution, 12g of hydroxylamine hydrochloride per 100 mL of DI water.
- Reducing reagent, 10% stannous chloride dihydrate w/v in 7% v/v trace metal grade HCl.

## Digestion Procedure

Samples were weighed (~0.2g) into tared 50 mL certified mercury-free polypropylene digestion tubes. The samples were pre-digested with 2 mL of H<sub>2</sub>SO<sub>4</sub> and 2 mL HNO<sub>3</sub>. The concentrated acids were added to the digestion tubes, the tubes were loosely capped and the samples were predigested for 1.5 hours at 80 °C. After the samples were

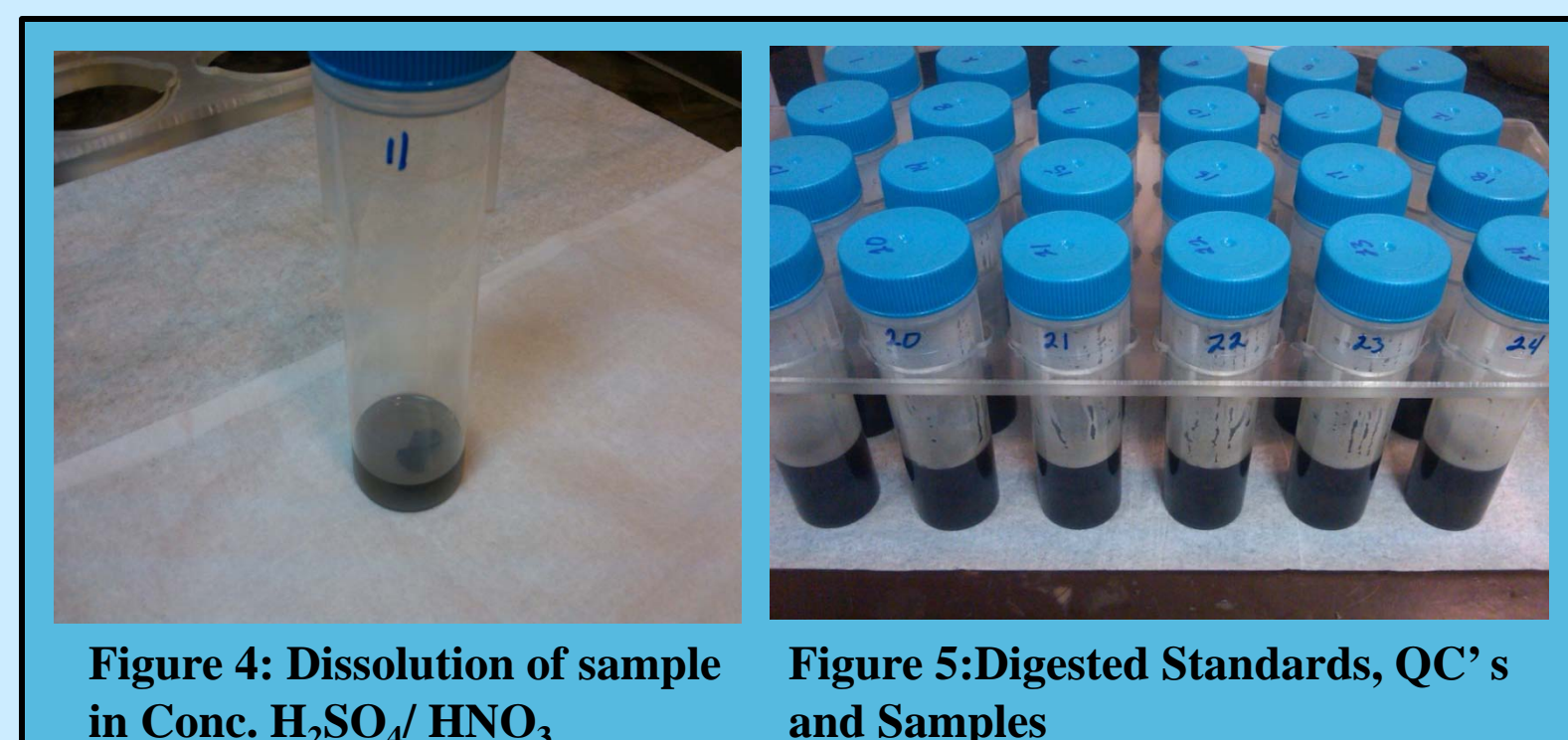


Figure 4: Dissolution of sample in Conc. H<sub>2</sub>SO<sub>4</sub>/HNO<sub>3</sub>

Figure 5: Digested Standards, QC's and Samples

dissolved and allowed to cool to room temperature, 7.0 mL of KMnO<sub>4</sub> and 5 mL 3% HCl was added to each digestion tube. The tubes were loosely capped, swirled then heated at 95 °C for 2 hours.

After again cooling to room temperature, 3.0 mL of the hydroxylamine solution was added to reduce the KMnO<sub>4</sub>. The digestion tubes were swirled to assure complete reaction. They were then brought up to a final volume of 50 mL with 3% HCl with thorough mixing prior to CVAA analysis of the samples.

## CVAA Instrument Method Settings

- Argon flow: 40 mL/minute
- Sample uptake time: 70s
- Pump speed: 100%
- Sample utilization: 11 mL per digest
- Baseline correction: 2-point, width = 10s
- Peak height : 4 replicates of 4 seconds each at each peak's maximum response level.
- Timing: Sample uptake – 70s, Rinse – 120s, Total – 2m12s

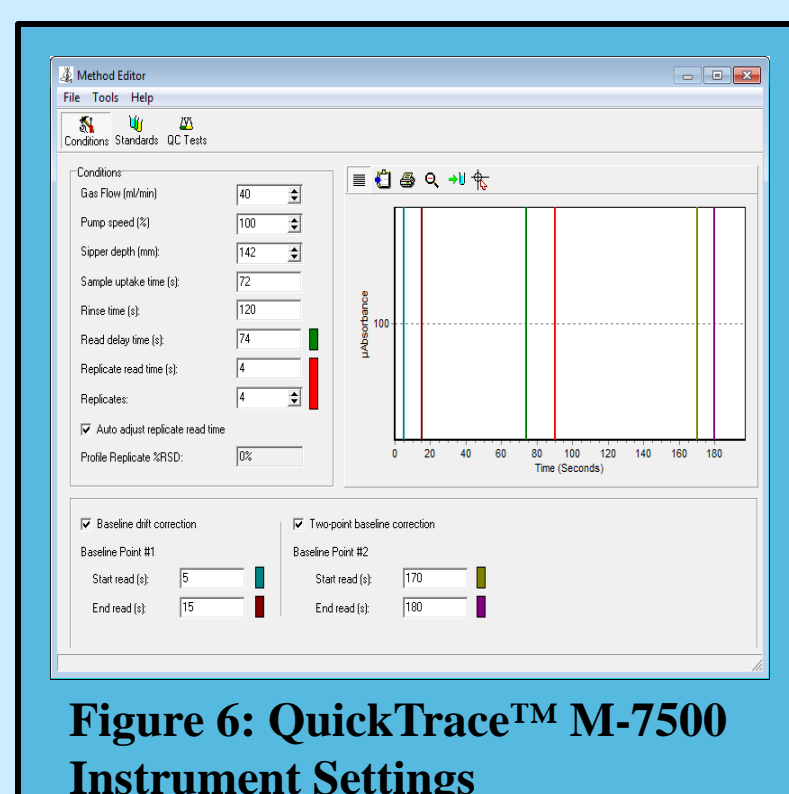


Figure 6: QuickTrace™ M-7500 Instrument Settings

## Calibration

Standards: serial dilutions of a 1000 ppm Hg stock standard to 1 ppb. 0.25, 0.50, 1.25, and 2.50 mL aliquots were transferred into certified mercury-free polypropylene digestion tubes containing the same 2 mL concentrated H<sub>2</sub>SO<sub>4</sub> and 2 mL HNO<sub>3</sub> used for sample digestion. The standards were treated like samples and digested along with the samples/QC's to eliminate any standard bias. The final calibration concentrations were 0, 5, 10, 25 and 50 ppt.

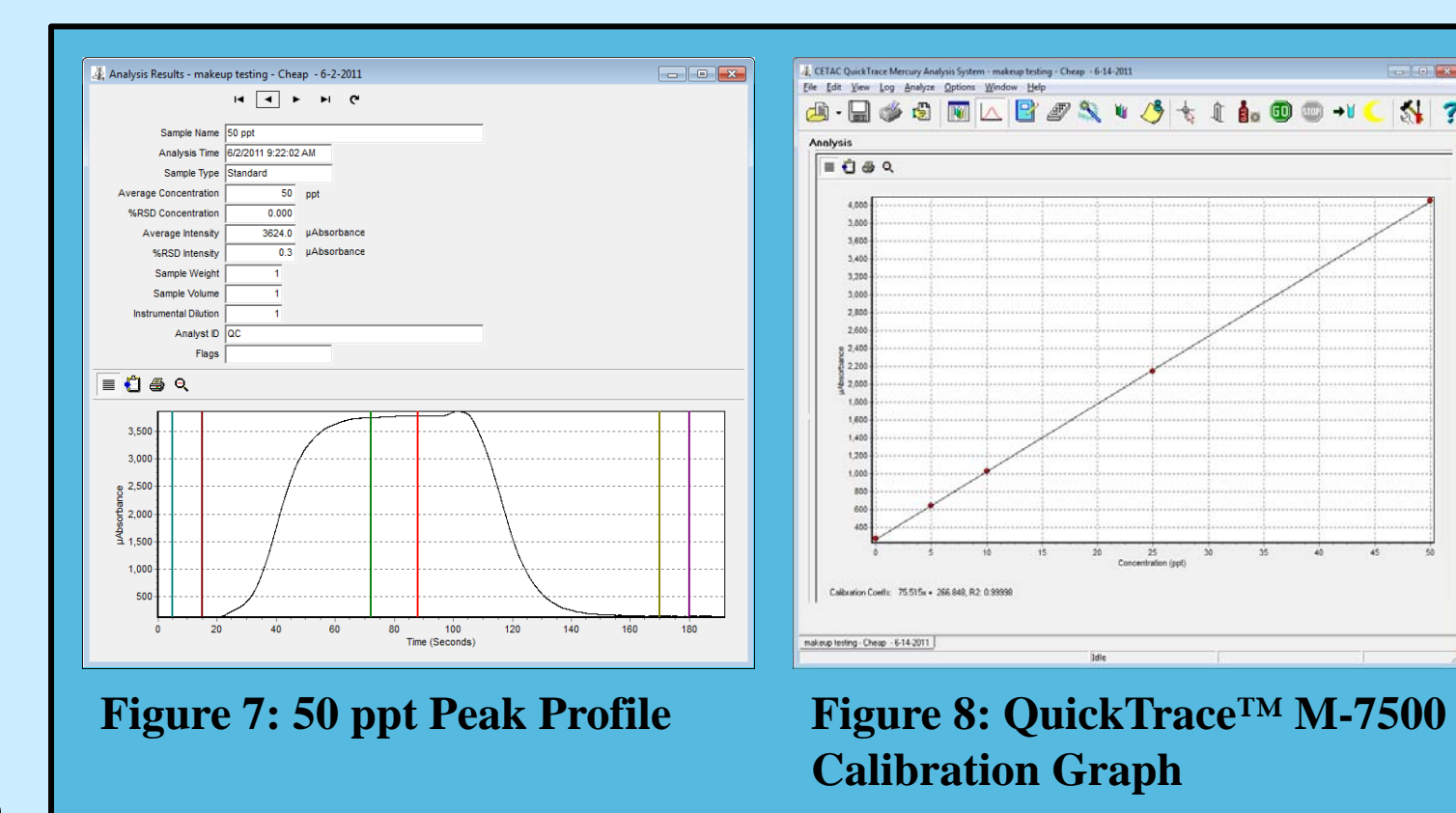


Figure 7: 50 ppt Peak Profile

Figure 8: QuickTrace™ M-7500 Calibration Graph

## Sample Analysis

Predigest spikes were performed at 50 - 150% of the sample's concentration. A sample for each cosmetics type was spiked in duplicate. Sequence included a 5 point calibration, CCV, CCB, ICV, LRB, LFB and samples. The oxidized mercury was reduced with on-line addition of 10% stannous chloride w/v in 7% hydrochloric acid v/v.

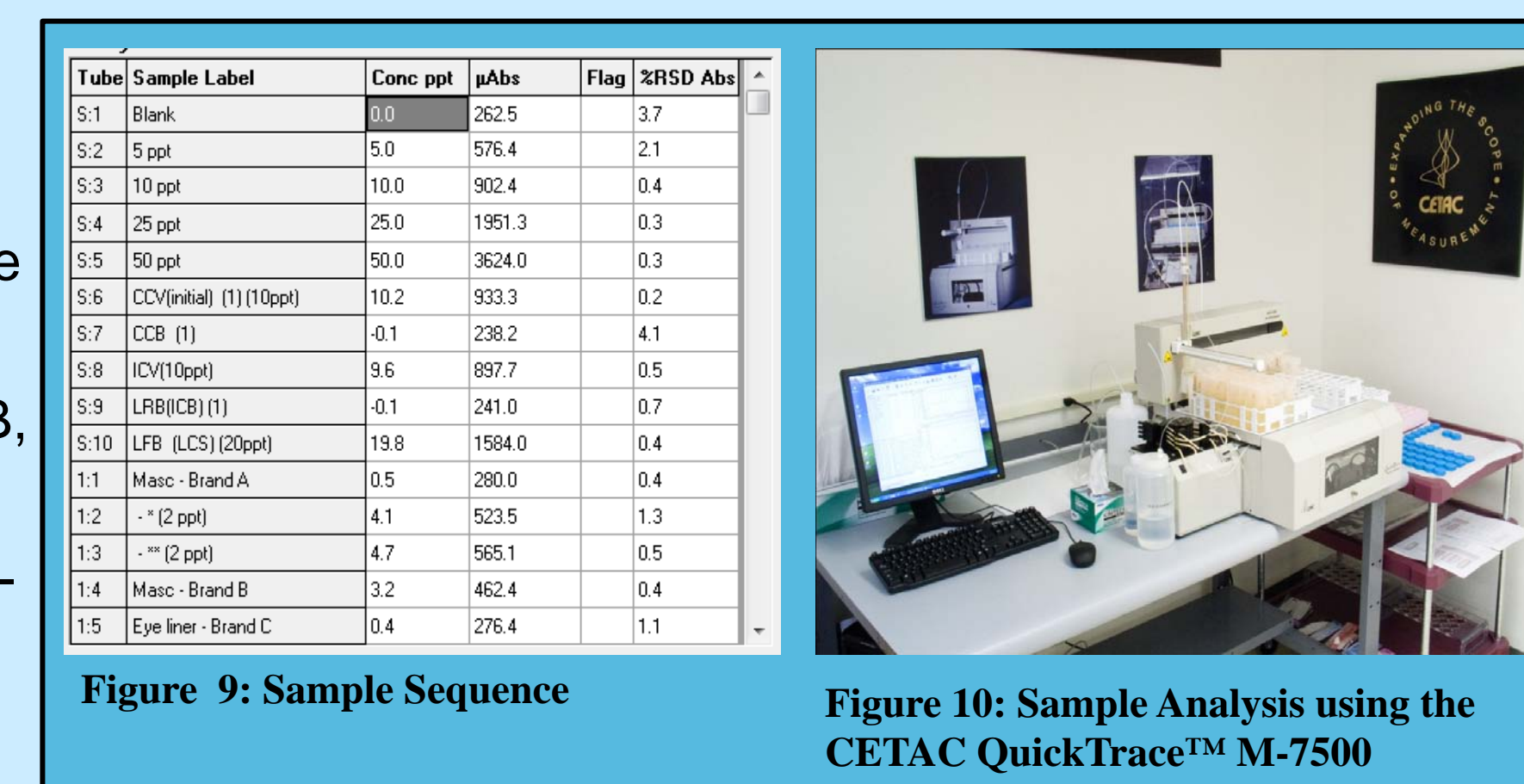


Figure 9: Sample Sequence

Figure 10: Sample Analysis using the CETAC QuickTrace™ M-7500

Purging of the hydroxylamine reagent for 30 minutes prior to use reduced the mercury background which ensures lower responses for blanks and reduces bias. The continuous flow rinse solution was 5% hydrochloric acid/ 5% nitric acid.

All ICV and CCV QC's were within 10% of calculated value and the initial CCV QC's were within 5% of calculated values.

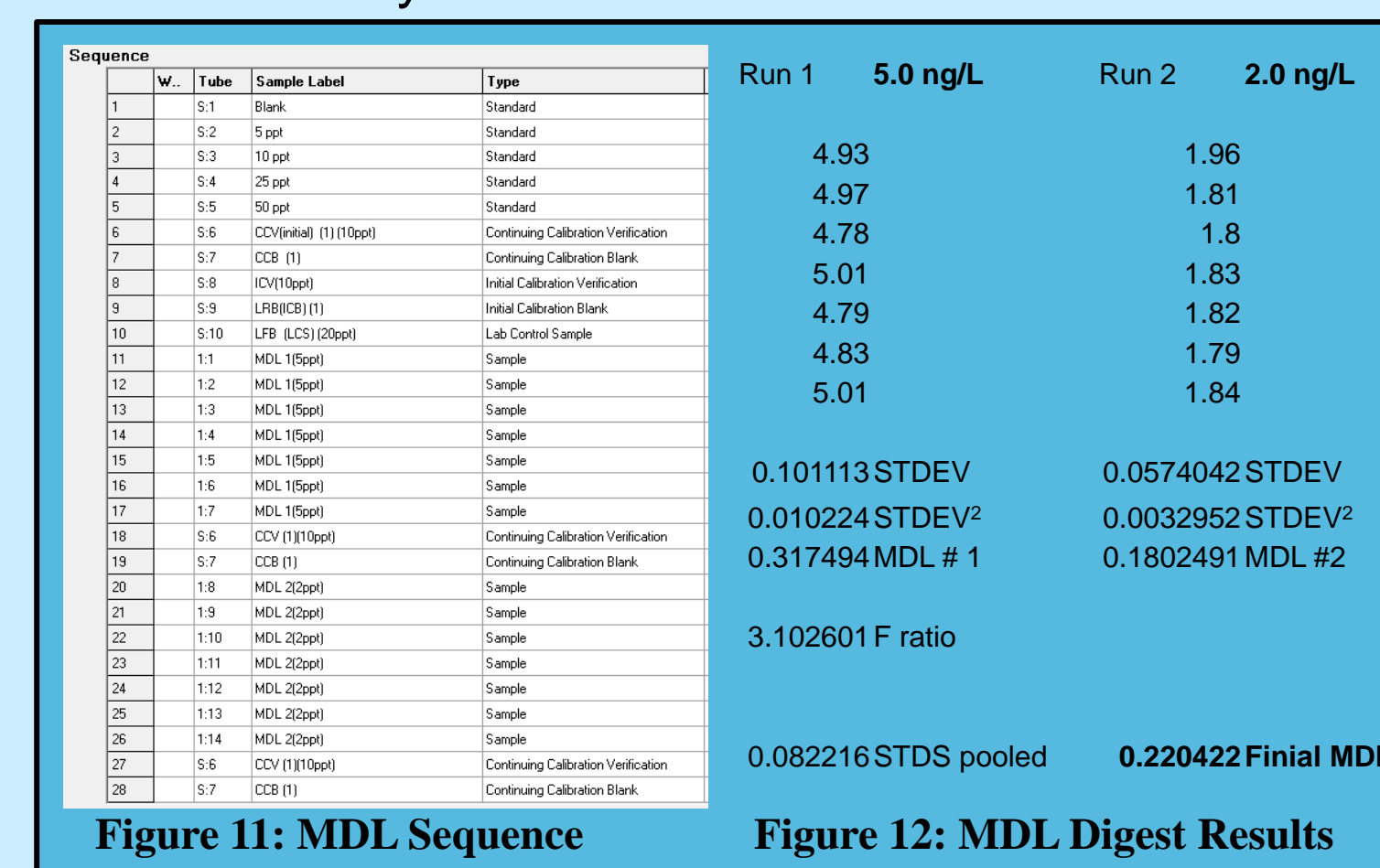


Figure 11: MDL Sequence

Figure 12: MDL Digest Results

## Results

Method detection limit = 0.22 ppt (Figure 11 and Figure 12)

“In-product” Hg = (reading/weight) \* 50 mL

The final results for all samples are given in parts per trillion (ppt). (Figure 13)

Masc - Brand A	674.8	Eye liner - Brand C	80.6	Eye shadow - Brand C	8750.0	Liq Found. - Brand E	0.0	Powder Found. - Brand F	263.2
Masc - Brand B	738.6	Eye liner - Brand B	2170.5	Eye shadow - Brand D	559.7	Liq Found. - Brand B	0.0	Powder Found. - Brand G	0.0
Masc - Brand AA	177.3	Eye liner - Brand AA	3429.8	Eye shadow - Brand AA	84.7	Liq Found. - Brand AA	286.9	Powder Found. - Brand AA	67.1
Masc - Brand BB	0.0	Eye liner - Brand BB	177.3	Eye shadow - Brand BB	33527.1	Liq Found. - Brand BB	78.1	Powder Found. - Brand BB	2878.8
Masc - Brand H	366.7	Eye liner - Brand H	279.3	Eye shadow - Brand H	640.2	Liq Found. - Brand H	119.0	Powder Found. - Brand H	1037.2
Masc - Brand D	2160.5	Eye liner - Brand D	961.5	Eye shadow - Brand F	593.2	Liq Found. - Brand D	1714.3	Powder Found. - Brand D	160.4
Masc - Brand F	3911.8	Eye liner - Brand F	0.0	Eye shadow - Brand A	601.1	Liq Found. - Brand F	49.8	Powder Found. - Brand A	1205.1
Masc - Brand C	1011.2	Eye liner - Brand A	392.7	Eye shadow - Brand K	3443.9	Liq Found. - Brand C	858.9	Powder Found. - Brand C	309.3
Masc - Brand I	0.0	Eye liner - Brand I	2307.7	Eye shadow - Brand G	3384.6	Liq Found. - Brand C	22.6	Powder Found. - Brand B	4162.2
Masc - Brand K	28.9	Eye liner - Brand J	119.0	Eye shadow - Brand B	1460.7	Liq Found. - Brand G	760.4	Powder Found. - Brand L	927.8

Figure 13: All Sample Results (ppt)

## Discussion

Microwaves are traditionally used to digest difficult sample for the detection of the heavy metals present. However, in this study it was shown that acceptable results can be obtained by a more conventional digestion method such as block digestion.

All of the cosmetic samples tested in this limited experiment were well below the 1 ppm limit set by the FDA. Separate comparisons by type and brand did not show statistically significant differences.

### Statistical Inference on Cosmetics by Type

Hg concentration from each sample type was organized and tested to see whether cosmetic type showed any significant differences. Testing for normality and equal variances showed that the data did not follow the normality assumption, but did follow the equal variance assumption.

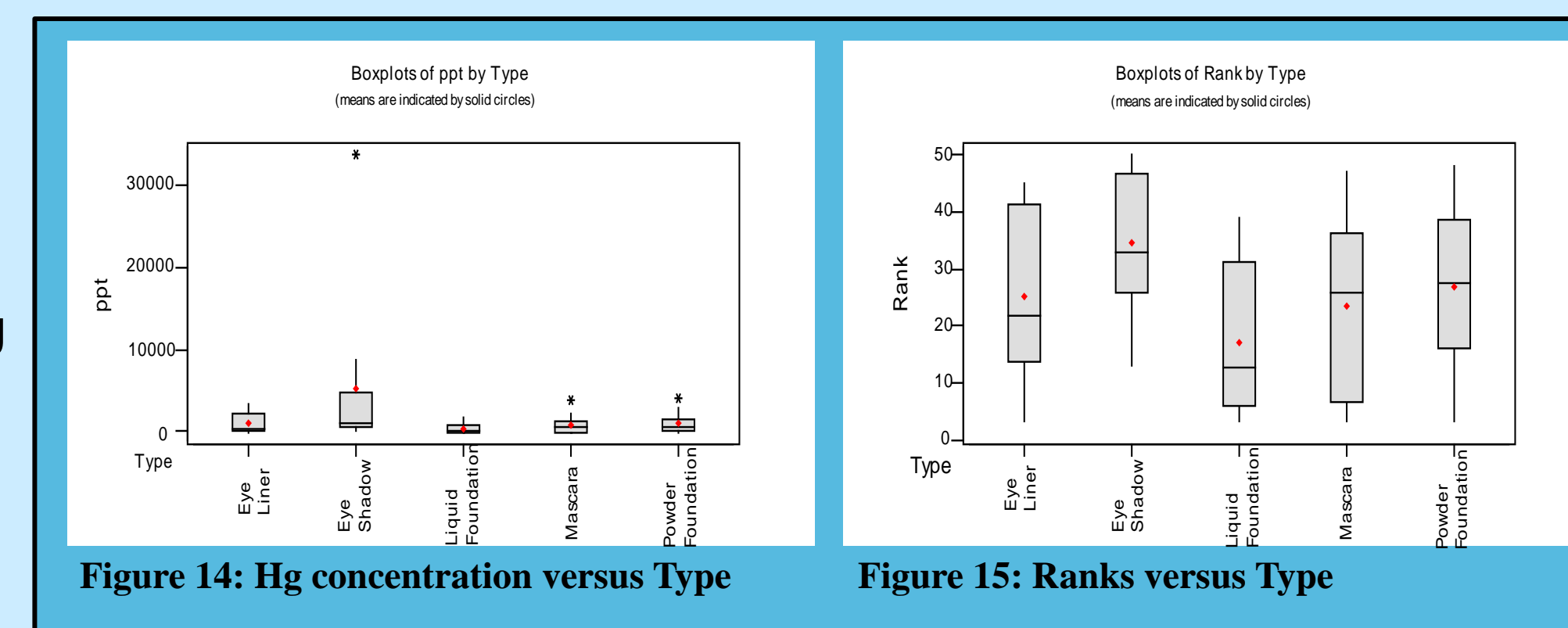


Figure 14: Hg concentration versus Type

Figure 15: Ranks versus Type

Therefore a non-parametric F-test was used. Figure 14 displays the Hg concentration versus type and Figure 15 displays ranks versus type.

The non-parametric F-test revealed p = 0.107. Although this p value represents no significant difference (α = 0.05), Mann-Whitney pairwise comparisons were carried out, and none of the pairwise comparisons were significant.

### Statistical Inference on Cosmetics by Brand

Figure 16 displays the Hg concentration versus brand and Figure 17 displays ranks versus brand.

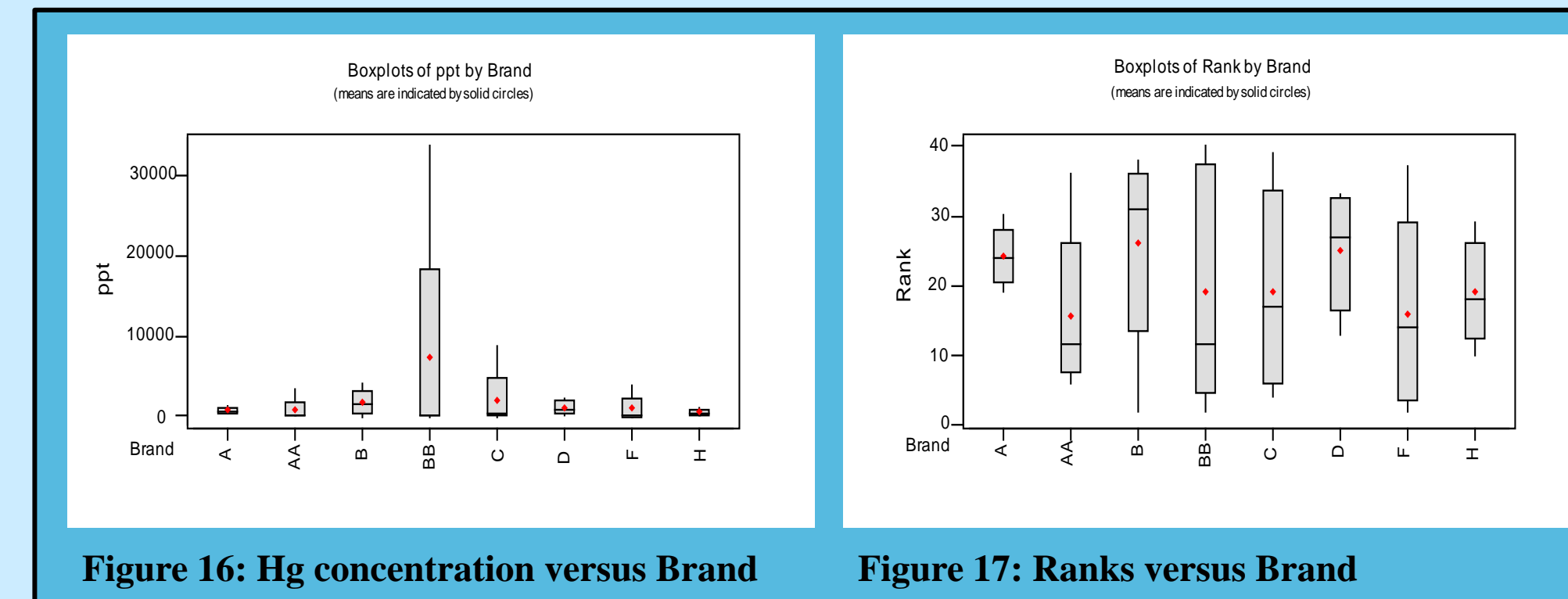


Figure 16: Hg concentration versus Brand

Figure 17: Ranks versus Brand

The non-parametric F-test revealed p = 0.787.

## References

- US EPA. Method 245.5, Mercury in Sediment (Manual Cold Vapor Technique), 1974.
- Please refer to the CETAC Application Note for additional references.