

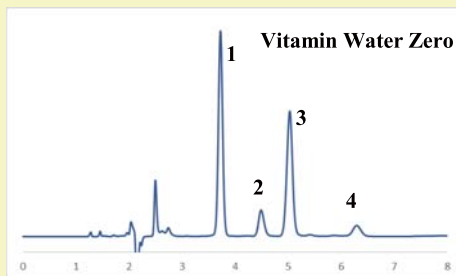
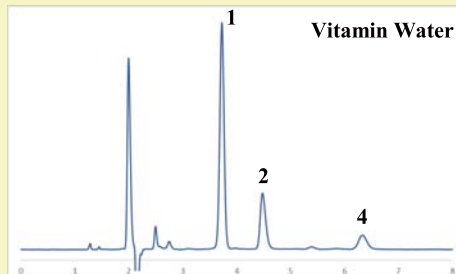
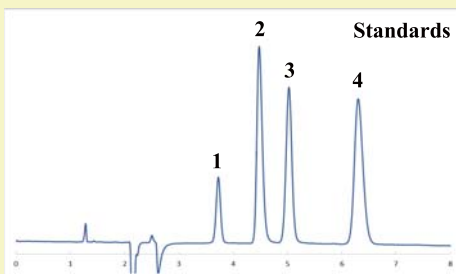
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Introduction

In recent years Functional Water Beverages have been gaining popularity among health conscious consumers. These beverages are usually enriched with Water-Soluble Vitamins (Vitamin C and B complex) and Fat-Soluble Vitamins (Vitamin A and E). Analysis of both types of vitamins poses a challenge due to the difference in solubility and wavelength absorption. We propose an easy and fast HPLC method for determining both water-soluble and fat-soluble vitamins using only 0.08% formic acid and acetonitrile with the COSMOSIL pentabromobenzyl (PBr) HPLC column. Three isocratic conditions were used for the analysis of 8 vitamins in functional water beverages using UV detection. The functional water beverages were injected directly without dilution.

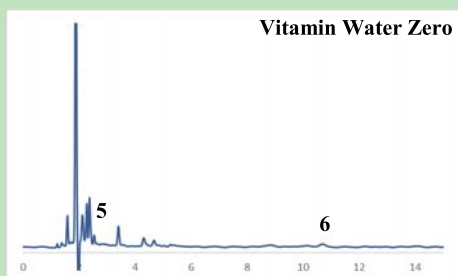
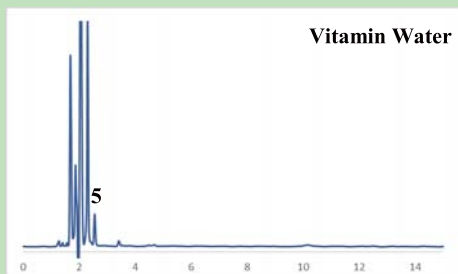
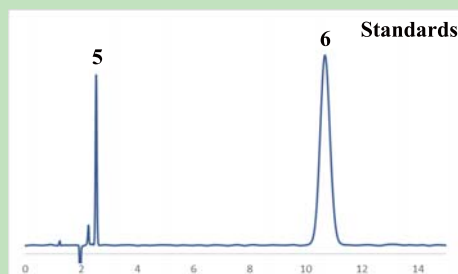
Experimental and Results

HPLC column: COSMOSIL PBr
 Column size: 4.6 x 150 mm, 5 μm
 Isocratic: 0.08% formic acid
 Flow rate: 1.0 mL/min
 Temperature: 30 °C
 Detection: UV 210 nm
 Injection Vol. 5.0 μL



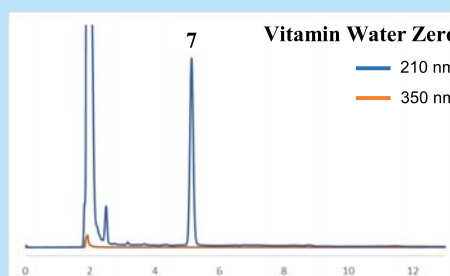
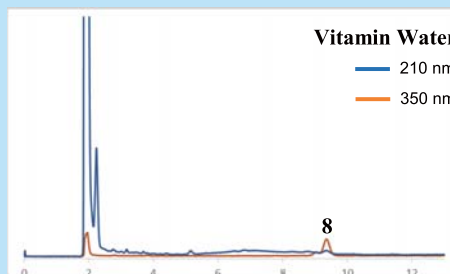
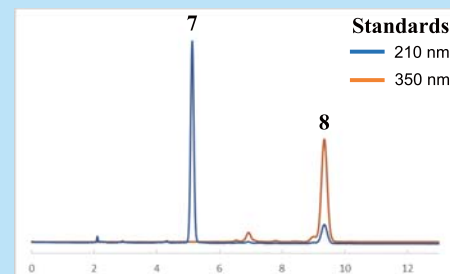
Using pure aqueous mobile phase in 0.08% formic acid, ascorbic acid, citric acid, niacinamide and pyridoxine HCL were baseline resolved under 7 minutes.

HPLC column: COSMOSIL PBr
 Column size: 4.6 x 150 mm, 5 μm
 Isocratic: 15:85 0.08% formic acid : acetonitrile
 Flow rate: 1.0 mL/min
 Temperature: 30 °C
 Detection: UV 210 nm
 Injection Vol. 5.0 μL



Vitamin B12 (cyanocobalamin) and vitamin B5 (calcium pantothenate) were separated with 15:85 0.08% formic acid : acetonitrile mobile phase.

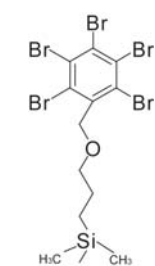
HPLC column: COSMOSIL PBr
 Column size: 4.6 x 150 mm, 5 μm
 Isocratic: Acetonitrile
 Flow rate: 1.0 mL/min
 Temperature: 30 °C
 Detection: UV 210 nm and 350 nm
 Injection Vol. 5.0 μL



Vitamin A palmitate and α-tocopheryl acetate (vitamin E) were well separated with 100 % acetonitrile under 10 minutes.

8-Component Vitamin Mix Standard

- | | |
|--------------------------------------|-------------|
| 1. Ascorbic acid (vitamin C) | (4 μg/ml) |
| 2. Citric acid | (350 μg/ml) |
| 3. Niacinamide (vitamin B3) | (4 μg/ml) |
| 4. Pyridoxine HCl (vitamin B6) | (5 μg/ml) |
| 5. Calcium pantothenate (vitamin B5) | (275 μg/ml) |
| 6. Cyanocobalamin (vitamin B12) | (158 μg/ml) |
| 7. α-tocopheryl acetate (vitamin E) | (16 μg/ml) |
| 8. Vitamin A palmitate | (33 μg/ml) |



COSMOSIL PBr

Conclusion

- COSMOSIL PBr column was used to successfully identify all 8 components of the mixture, using UV detection and 3 isocratic conditions.
- Vitamin Water was found to contain ascorbic acid, citric acid, pyridoxine HCl, calcium pantothenate, and vitamin A palmitate. Cyanocobalamin was on the product label, but was not detected, possibly due to degradation and/or low amount.
- Vitamin Water Zero was found to contain ascorbic acid, citric acid, niacinamide, pyridoxine HCl, calcium pantothenate, cyanocobalamin, and α-tocopheryl acetate.