

## SEPARATION OF CAROTENOIDS FROM TOBACCO

At the HPLC 84 meeting Bailey and Beven of the British-American Tobacco Co. Ltd., reported an efficient analytical system for the resolution of naturally-occurring carotenoids extracted from tobacco leaf. The method was for the characterization of individual carotenoid peaks so that the user would be able to establish similarities and differences between carotenoids from different sources.

**LC Analysis.** A normal-phase, gradient elution method was developed and used to resolve a carotenoid extract into "more components than had been reported previously." A typical separation is shown in Figure 1.

Since the polarity of carotenoids ranges from polyunsaturated C<sub>40</sub> hydrocarbons through mono- and di-epoxy derivatives and from mono- to tetra-ols, the resolution of such mixtures requires both the surface adsorption features of silica and an eluent ranging from very non-polar to relatively polar.

A stepwise gradient was run at 1 ml/min (see dotted line in Figure 1) with the solvent program:

- Step A 5% Isopropanol in hexane 10 min.
- Step B 7% Isopropanol in hexane 5 min.
- Step C 11% Isopropanol in hexane 10 min.
- Step D 14% Isopropanol in hexane 10 min.
- Step E Re-equilibration between runs:  
5% Isopropanol in hexane 10 min.

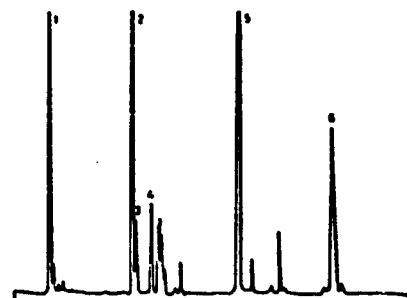
The silica which the investigators used was very similar to RESOLVE<sup>TM</sup> Silica. Therefore, one would speculate that given the advantages of the RCM-100<sup>®</sup> Radial Compression Module for gradient analysis, a faster analysis would be possible using the Radial-PAK<sup>TM</sup> cartridge.

While it was not clear from the abstract which wavelength was being monitored in Figure 1, it was surmised to be 450 nm.

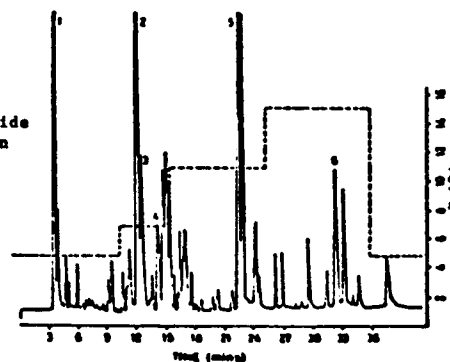
FIGURE 1

### CAROTENOIDS FROM TOBACCO LEAF EXTRACTS

#### A) NORTH AMERICAN FLUE CURED



#### B) SOUTH AMERICAN AIR CURED



1. Carotenes
2. Lutein
3. Zeaxanthin
4. Lutein epoxide
5. Violaxanthin
6. Zeoxanthin