

## An improved method for collecting gas-chromatographically separated estrogens\*

In recent years, the gas chromatographic separation of synthetic mixtures of estrogens as well as their isolation from biological material have been reported<sup>1-6</sup>. The further characterization and verification of the fractions obtained, in many instances, would be extremely helpful.

A technique, employed in our laboratory, for collecting the effluent fractions at the exit port of the gas chromatograph has facilitated the collection of the fractions, representing the individual estrogens, for further characterization.

### *Experimental*

The estrogens are separated employing a Barber-Colman model 10 gas chromatograph equipped with a <sup>90</sup>Sr ionization detector<sup>3,6</sup>.

The effluent fractions, representing the individual compounds, are collected at

TABLE I  
RECOVERY EFFICIENCY OF COLLECTION METHOD  
FOLLOWING INJECTION OF STANDARD

<i>Injected (<math>\mu</math>g)</i>	<i>Collected (<math>\mu</math>g)</i>	<i>Collected (%)</i>
<i>17<math>\beta</math>-Estradiol</i>		
3	2.7	91
3	2.5	83
3	2.7	91
6	5.0	83
6	5.5	91
6	4.6	76
	Average	86
<i>Estrone</i>		
3	2.5	83
3	2.7	90
3	2.4	85
6	4.8	80
6	5.6	93
6	5.2	87
	Average	86
<i>Estriol</i>		
5	4.7	94
5	4.6	91
5	4.7	94
10	8.7	87
10	8.4	84
10	9.1	91
	Average	90

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the exit port of the gas chromatograph by heating the exit line with an electric heating coil. Temperature of the line is controlled employing a 7.5 A variable transformer (powerstat). The fraction is collected by immersing the exit line (a 2.5 cm area not covered by the heating coil) directly into a 7 cm long, conical bottom pyrex glass tube containing 1.0 ml of redistilled dioxan. Each collection tube was cooled in a refrigerator at about 7°C for 10–15 min before collecting the samples. The resulting dioxan solution is then used to further characterize the compounds.

### *Result and discussion*

In Table I may be observed the recovery efficiency of the collecting method following the injection into the gas chromatograph of a synthetic mixture of estrone, 17 $\beta$ -estradiol and estriol. The recoveries are based on the observed maxima upon determining the ultraviolet absorption spectra of the compounds prior to and following passage through the gas chromatograph.

Previously published methods of collection have employed electrical precipitation<sup>7</sup>, Volman traps<sup>8</sup>, or traps packed with defatted cotton, glass wool, glass beads, or silica. None of these methods were found to be adaptable to the microgram concentrations of estrogen employed in our experiments or known to be present in biological material.

Employing the method described in this communication, will provide a means of collecting gas chromatographically separated fractions for further characterization.

The efficiency of the method permits recovery of the fractions, after passage through the column, in sufficient concentration so that microgram quantities of material may be conveniently handled. It may be possible to adapt this method to the collection of compounds other than estrogens.

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