PROCEDURE 1

DETERMINATION OF ADEQUATE CHROMATOGRAPHIC PEAK RESOLUTION

In this method of dealing with resolution, the extent to which one chromatograph peak overlaps another is determined.

For convenience, consider the range of the elution curve of each compound as running from -2σ to $+2\sigma$. This range is used in other resolution criteria, and it contains 95.45 percent of the area of a normal curve. If two peaks are separated by a known distance, b, one can determine the fraction of the area of one curve that lies within the range of the other. The extent to which the elution curve of a contaminant compound overlaps the curve of a compound that is under analysis is found by integrating the contaminant curve over the limits b $-2\sigma_s$ to b $+2\sigma_s$ where σ_s is the standard deviation of the sample curve.

This calculation can be simplified in several ways. Overlap can be determined for curves of unit area; then actual areas can be introduced. Desired integration can be resolved into two integrals of the normal distribution function for which there are convenient calculation programs and tables. An example would be Program 15 in Texas Instruments Program manual ST1, 1975, Texas Instruments, Inc., Dallas, Texas 75222.

In judging the suitability of alternate GC columns or the effects of altering chromatographic conditions, one can employ the area overlap as the resolution parameter with a specific maximum permissible value.

The use of Gaussian functions to describe chromatographic elution curves is widespread. However, some elution curves are highly asymmetric. In cases where the sample peak is followed by a contaminant that has a leading edge that rises sharply but the curve tails off, it may be possible to define an effective width for t_c as "twice the distance from the leading edge, measured along a perpendicular bisection of that line."

$$\frac{1}{\sqrt{2\pi}\sigma_c}\int_{b-2\sigma_s}^{b+2\sigma_s} e^{\left(\frac{-t_c^2}{2\sigma_c}\right)}dt = \frac{1}{\sqrt{2\pi}}\int_{\frac{b-2\sigma_s}{\sigma_c}}^{\infty} e^{\left(\frac{-x^2}{2}\right)}dx - \frac{1}{\sqrt{2\pi}}\int_{\frac{b+2\sigma_s}{\sigma_c}}^{\infty} e^{\left(\frac{-x^2}{2}\right)}dx$$

The following calculation steps are required:*

1. $2\sigma_s = t_s/\sqrt{2 \ln 2}$

2. $\sigma_c = t_c/2\sqrt{2 \ln 2}$

3. $x_1 = (b-2\sigma_s)/\sigma_c$

4. $x_2 = (b+2\sigma_s)/\sigma_c$

5. $$Q(x_1) = \frac{1}{\sqrt{2\pi}} \int_{x_1}^{\infty} e^{\left(\frac{-x^2}{2}\right)} dx$$

6. $$Q(x_2) = \frac{1}{\sqrt{2\pi}} \int_{x_2}^{\infty} e^{\left(\frac{-x^2}{2}\right)} dx$$

7. $$I_o = Q(x_1) - Q(x_2)$$

8. $$A_o = I_o A_c / A_s$$

9. $$Percentage\ overlap = A_o \times 100$$

where:

A_s	=	Area of the sample peak of interest determined by electronic integration or by the formula $A_s = h_s t_s$.
A_c	=	Area of the contaminant peak, determined in the same manner as A_s.
b	=	Distance on the chromatographic chart that separates the maxima of the two peaks.
H_s	=	Peak height of the sample compound of interest, measured from the average value of the baseline to the maximum of the curve.
t_s	=	Width of sample peak of interest at ½ peak height.
t_c	=	Width of the contaminant peak at ½ of peak height.
σ_s	=	Standard deviation of the sample compound of interest elution curve.
σ_c	=	Standard deviation of the contaminant elution curve.
$Q(x_1)$	=	Integral of the normal distribution function from x_1 to infinity.
$Q(x_2)$	=	Integral of the normal distribution function from x_2 to infinity.
I_o	=	Overlap Integral.
A_o	=	Area overlap fraction.

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* In most instances, $Q(x_2)$ is very small and may be neglected.