

# Data Table

## Part 1. Isocratic Separation

	Red Dye			Blue Dye		
	Run #1	Run #2	Run #3	Run #1	Run #2	Run #3
Start of Band (mL)						
End of Band (mL)						
$W$ (mL)						
$V_{Rave}$ (mL)						
$k'$						

### Calculated Values

 $\alpha$  \_\_\_\_\_

 $R$  \_\_\_\_\_

## Part 2. Step Gradient Separation

Beaker	Eluant	Observations—Eluted Fractions
1	H <sub>2</sub> O	
2	5% isopropyl alcohol	
3	28% isopropyl alcohol	
4	70% isopropyl alcohol	

## Calculations *(Use a separate sheet)*

Determine the following values and show calculations. Refer to question six in the Pre-Lab Questions. Enter results in the Part 1 data table.

1. Bandwidth,  $W$ , for each dye.
2. Average Retention Volume,  $V_{\text{Rave}}$ , for each dye.
3. Capacity Factor,  $k'$ , for each dye.
4. Selectivity,  $\alpha$ , for the two dyes with this isocratic separation.
5. Resolution,  $R$ , for the two dyes with this isocratic separation.

## Post-Lab Questions

1. What is meant by polarity of molecules? What causes differences in polarity?
2. In discussing solubility, the rule “like dissolves like” is frequently used. What does this mean?
3. Draw the structural formula of isopropyl alcohol. Explain how it differs in polarity from water.
4. For good separation of the dyes, the resolution should be greater than one. What was the value you calculated? Did the two dyes overlap as they emerged from the column, or was the separation a good one?
5. In the step gradient separation, four separate fractions were collected. How were these related to the polarities of the column and of the eluting solvent?