Gas Chromatographic Determination of Hydrophile-Lipophile Balance of Nonionic Emulsifiers[1]

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ABSTRACT

The determination of the hydrophile-lipophile balance (HLB) values of nonionic emulsifiers by a gas chromatographic method is described. Factors that affect this correlation have been examined. Attempts to extend this method to the determination of the required HLB values of fats and oils are presented. These data along with supplementary wet methods have been used to determine optimum conditions for emulsion stability of simple model systems (fat-water-emulsifier).

INTRODUCTION

The idea that an emulsifier molecule should contain a balance between polar and nonpolar moieties was pointed out by Clayton (1). This approach was further delineated in quantitative terms by Griffin (2,3) with the introduction of the concept of hydrophile-lipophile balance (HLB). Essentially, by a long experimental procedure, values were assigned to emulsifiers ranging from 1 (lipophilic) to 20 (hydrophilic), and suitable applications were assigned to these ranges as in Table I.

It remained for several other workers to observe correlations between HLB values of emulsifiers and various other physical properties and parameters. Griffin (4) observed an increase in water solubility with increasing emulsifier HLB values.

When the structure of the emulsifier molecule is known, the HLB can be calculated from the relation HLB = $(E + P)/5$, where E is the weight percentage of oxyethylene content and P is the weight percentage of polyhydric alcohol content.

By examining the HLB values for a large number of emulsifiers, Davies (5) was able to derive group values for the structural moieties of the molecule in the following equation: HLB = $7 + \Sigma$ (hydrophilic group numbers) - Σ (lipophilic group numbers).

Both methods work well for a series of sorbitan and polyoxyethylene (POE) sorbitan esters, etc., but suffer from the limitation that they are not applicable to nonionic surfactants containing other hydrophilic oxide units, sulfur or nitrogen-containing surfactants or ionic surfactants.

Griffin (2,3) also observed that HLB = $20(1 - S/A)$, where S is the saponification number of the ester and A is the acid number of the acid. This method appears limited in the inability to obtain good values for S.

TABLE I

HLB vs. Application[a,b]

HLB range	Applications
1-6	Water in oil emulsifier
7-9	Wetting agent
8-18	Oil in water emulsifier
13-15	Detergent
15-18	Solubilizer

[a] Reference 2.

[b] HLB, hydrophile-lipophile balance.

Several other methods measure the polar nature of the emulsifier molecule, which should account for the hydrophile-lipophile properties. Gorman and Hall (6) reasoned that the dielectric constant of the molecule should express the molecular polarity intrinsic to the HLB concept. Indeed a linear relationship was observed for the reported HLB value vs. the log of the dielectric constant for a group of 20 different emulsifiers.

Ben-Et and Tatarsky (7) measured the ratio of hydrophilic to lipophilic hydrogens by nuclear magnetic resonance and obtained a good correlation with published HLB values for a series of emulsifiers.

As another approach, Harva et al. (8) used the emulsifier as the liquid phase on a gas chromatograph (GC). A linear relationship was obtained between the partition coefficients (water and diisobutylene) and the HLB numbers of the emulsifier used as the liquid substrate on the column packing.

Huebner (9) suggested a more meaningful polarity index based on a GC determination of the "apparent carbon number" of methanol by looking at the retention times of a homologous series of hydrocarbons, again using the emulsifiers as the liquid GC packing substrate.

In an effort to further simplify the GC procedure, Becher and Birkmeier (10) injected a hexane-methanol mixture onto columns, each coated with a nonionic emulsifier. A linear relationship was obtained when HLB was plotted against ρ, the ratio of the retention time of ethanol over the retention time of hexane.

Recently Mickle et al. (11) measured emulsifier HLB by a similar GC technique injecting isoamyl alcohol onto columns, each containing a different emulsifier. A linear relation was again obtained by plotting retention time of the alcohol vs. known HLB values.

This paper is an extension of the gas chromatographic determination of HLB and will examine the factors that affect the correlation between gas chromatographic data and emulsifier HLB values including treatment of data, column temperature, column length, flow rate, liquid loading, solid supports and injection mixtures. Correlations of GC data and required HLB values of fats and oils are also presented.

EXPERIMENTAL PROCEDURES

The gas chromatograph used was a Varian Aerograph Model 90-P equipped with a thermal conductivity detector. Injector and detector temperatures were 100 C, and the helium flow rate was 40 cc/min. Columns were generally prepared by dissolving the emulsifier (used as received with no further purification) in a suitable organic solvent such as chloroform, acetone, etc., and adding this solution to the solid support, usually Johns-Manville Chromosorb W non-acid-washed 60/80 mesh. A rotary evaporator was used to remove the solvent. Warming the flask facilitated the solvent removal, although some emulsifiers tended to break down under the elevated temperatures. If this occurred, the packing was prepared again without heating. Experimental quantities of the various emulsifiers used were obtained directly from the manufacturers or their representatives.

Since the emulsifiers used in this paper are known interchangeably by their chemical and trade names, a list of both are included. The numbers before the names will be

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[1] Presented at the AOCS Meeting, Ottawa, September 1972.

referred to in various figures throughout the paper. They are as follows: (1) sorbitan monolaurate, SPAN 20; (2) sorbitan monopalmitate, SPAN 40; (3) sorbitan monostearate, SPAN 60; (4) sorbitan tristearate, SPAN 65; (5) sorbitan monooleate, SPAN 80; (6) polyoxyethylene (20) sorbitan monolaurate, i.e., POE (20) sorbitan monolaurate, TWEEN 20 or polysorbate 20; (7) POE (20) sorbitan monopalmitate, TWEEN 40 or polysorbate 40; (8) POE (20) sorbitan monostearate, TWEEN 60; (9) POE (20) sorbitan monooleate, TWEEN 80; (10) POE (4) sorbitan monostearate, TWEEN 61; (11) POE (20) sorbitan tristearate, TWEEN 65; (12) POE (5) sorbitan monooleate, TWEEN 81; (13) POE (20) sorbitan trioleate, TWEEN 85, etc.

A coiled copper column, generally 7 ft x 1/4 in., was filled with the coated packing by gravity and aspirator vacuum. The column was cured prior to use by heating in the chromatograph oven at 100 C for 1-12 hr depending on the stability of the coatings. A 50:50 v/v mixture of hexane-ethanol (analytical reagent grade or better to eliminate the possibility of interfering impurity peaks) was injected a minimum of three times at each temperature to insure reproducibility. Injections of the single components were made to establish peak identity. The column temperature was raised in 10 C increments to a maximum of 80-90 C depending on substrate stability. (Stability was tested by reducing the column temperature after the final high temperature injection, injecting the mixture again and comparing the retention times with those obtained from the initial similar temperature injection.) Emulsifier mixtures, fats and oils, when used as coatings, were prepared and evaluated in the same manner as described above.

The "required HLB values" of fats and oils were determined for correlation with GC data in the following manner: mixtures of POE sorbitan monostearate and sorbitan monostearate were prepared with resultant integral HLB values between 14.9 and 4.7 from the following relationship: $HLB_{mix} = 14.9(x) + (1\text{-}x)4.7$, where x = (% POE sorbitan monostearate)/100 and $(1 - x)$ = (% sorbitan monostearate)/100. The validity of such a relationship is discussed elsewhere in the paper. Emulsions were prepared by taking 50 g of the oil or melted fat, 50 g of water and 0.25 g (0.5% based on oil) of the emulsifier mixture. The mixture was emulsified using a blender or lab size piston homogenizer. The most stable emulsion within a series using the same oil with different emulsifier mixtures was determined in several ways: measurement of the extent of aggregation (creaming) or coalescence (oiling off), opacity of the aqueous phase, etc. In some cases destabilization of the emulsion was accomplished by centrifugation, freeze-thaw cycles, heating or some combination of these stresses.

RESULTS AND DISCUSSION

Initially the method described by Becher and Birkmeier (10) for the GC determination of HLB values of emulsifiers was followed. Columns were prepared using as the liquid column coating various sorbitan and POE sorbitan esters, and the retention times of hexane and ethanol injected on the columns at 80 C were recorded. The ratios $R_{ethanol}/R_{hexane}$ were calculated for each emulsifier and these values (ρ) plotted against the published HLB values. A linear relationship similar to that published by Becher and Birkmeier (10) for a series of sorbitan esters was obtained again with the greatest amount of scatter (±3 HLB units) appearing in the 8-11 HLB range.

Because a portion of the retention time of each component was due to physically traveling through the column (void volume), and the dead space of the injector and detector, the retention times of the hexane and ethanol were measured on a column containing just the solid support with no liquid substrate. These retention times, both ca. 1 min, were similar to the air peak retention time when the mixture was injected on all the coated columns. Accordingly, the adjusted retention times ($R_{hexane,EtOH}\text{-}R_{air}$) were used and the ρ' values calculated and plotted. Figure 1 shows the results.

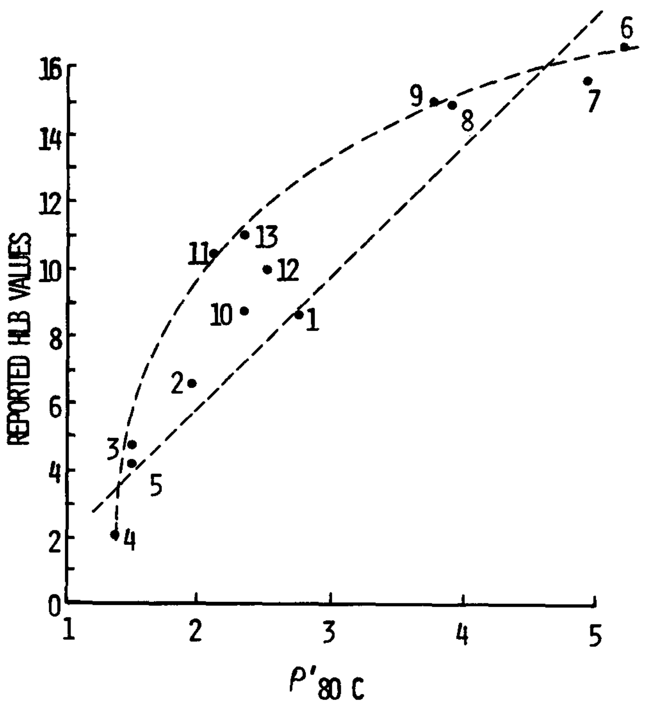

FIG. 1. Plot of hydrophile-lipophile balance vs. $\rho'_{80\ C}$ for a series of sorbitan and POE sorbitan esters. $\rho'_{80\ C} = (R_{EtOH}\text{-}R_{air})/(R_{hexane}\text{-}R_{air})$. Refer to experimental section for emulsifier numbering.

A curvilinear relationship such as $ln\ \rho'$ vs. HLB, also shown, appeared to lessen the scatter. Replotting the data as shown in Figure 2 illustrates this.

Regression analysis bears out the qualitative observations just mentioned. For a plot of published HLB values against

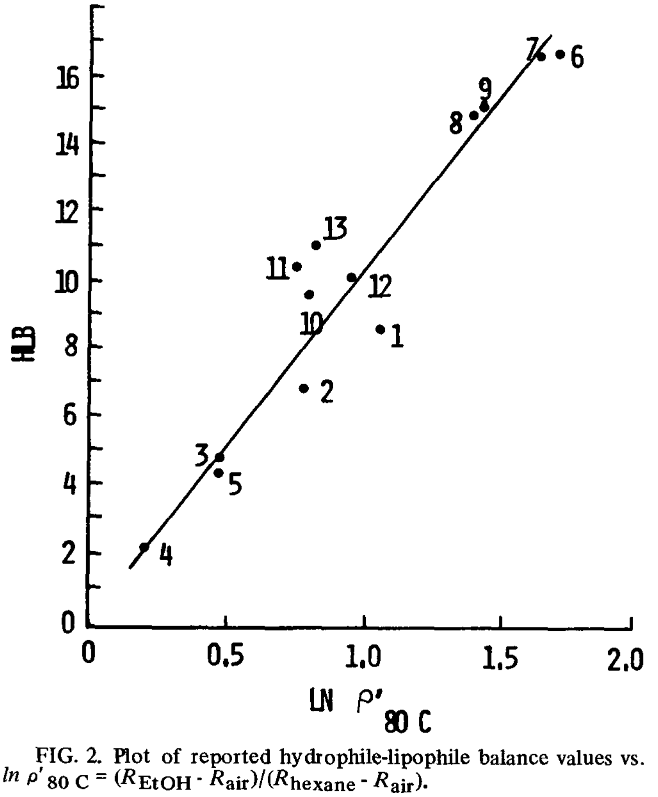

FIG. 2. Plot of reported hydrophile-lipophile balance values vs. $ln\ \rho'_{80\ C} = (R_{EtOH} - R_{air})/(R_{hexane} - R_{air})$.

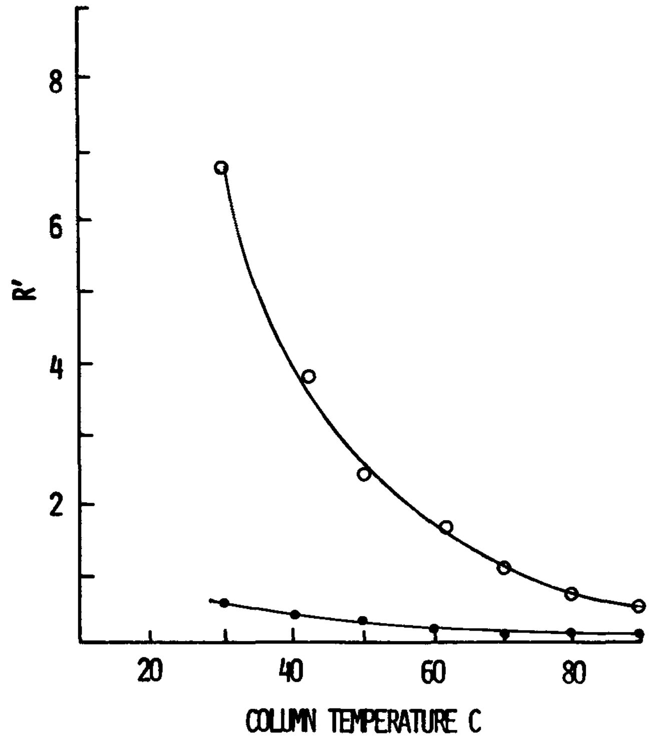

FIG. 3. Plot of $R'(R_{EtOH}$ - R_{air}[○]; R_{hexane} - R_{air}[●]) vs. column temperature for 5% POE sorbitan monolaurate on 7 ft 60/80 Chromosorb W column.

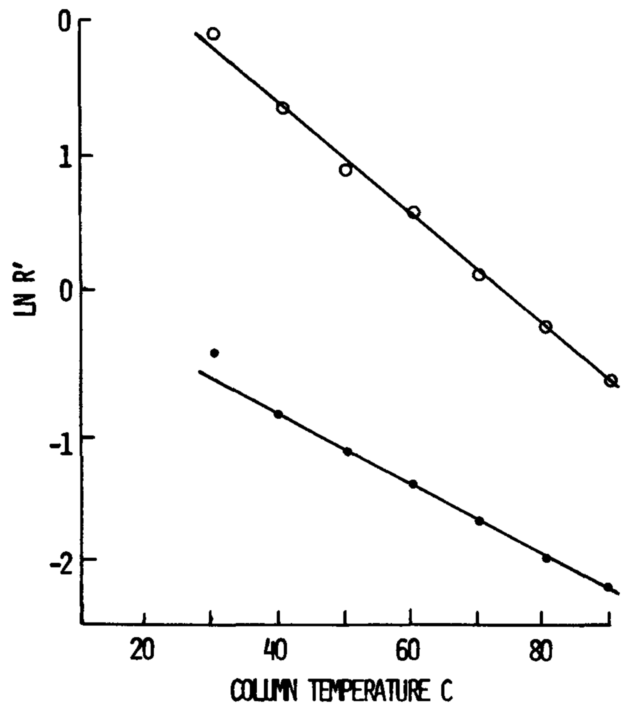

FIG. 4. Plot of *ln* R' for ethanol (○) and hexane (●) vs. column temperature for a column of 7 ft 5% POE sorbitan monolaurate.

ρ (R_{EtOH}/R_{hexane}), the standard error of estimate is 1.76 HLB units and the linear equation that best fits the data is HLB = 31.2 ρ - 30.7 (compared with Becher's equation HLB = 8.55 ρ - 6.36 [10]). When the air peak is subtracted out as in Figure 1, which plots HLB value against ρ' ([R_{EtOH}-R_{air}]/[R_{hexane}-R_{air}]), the resultant linear equation that best fits the points is HLB = 3.37 ρ' + 0.58 with the standard error of estimate increased to 1.93 HLB units. However, if the data is fitted to a logarithmic curve, the equation for the best fit becomes HLB = 10.2 *ln* ρ' + 0.45 with a resultant standard error of estimate of 1.40 HLB units, lower than either of the previous treatments. The greatest amount of scatter in Figure 2 occurs in the region of HLB 8-11. The same scatter pattern was observed in a plot of published HLB values vs. log of dielectric constant of some nonionic emulsifiers (6).

The fact that the relationship between HLB and ρ' appears to be logarithmic agrees well with the previously reported linear correlation of the log of the dielectric constant vs. HLB by Gorman and Hall (6). In GC, linear relationships are frequently obtained when plotting the log of retention volumes vs. the number of structural units such as in homologous series of alcohols, sulfur compounds, ethers, etc. Although the emulsifiers used are not, strictly speaking, a homologous series, they do exhibit increasing polarity with increasing HLB and as such might be expected to exhibit similar behavior.

Mickle et al. (11) were able to explain some of their emulsion stability results more fully when HLB values derived from GC data were used, rather than published values which differed in some cases.

The effect of temperature on the retention times of hexane and ethanol was investigated next. Becher and Birkmeier (10) observed some changes in ρ at different temperatures, but the implication of this was left an open question.

Retention values were obtained at temperatures ranging from 30 to 90 C for the emulsifiers already presented. The results for a typical liquid emulsifier, POE sorbitan monolaurate, are shown in Figure 3.

The curves appear to be a logarithmic function of the retention times, and when replotted as *ln* R' Figure 4 results. That the correlation of retention time vs. temperature should be logarithmic is implicit from an Antoine equation log $V_g = A + (B/[t + C])$ where A, B and C are constants, V_g = net retention volume and t = temperature.

As shown in Figure 4, the two lines, representing the *ln* value of the relative retention times (R-R_{air}), are convergent with increasing temperature. Such a phenomenon was observed with all the emulsifiers because the slope of *ln* R'_{EtOH} (reflecting the polar interactions) was always steeper than *ln* R'_{hexane} (reflecting the nonpolar interactions). This convergence would lead to smaller *ln* ρ' values as the temperature increased. In fact, after *ln* ρ' values were determined for all the emulsifiers over the range 30-90 C, an interesting trend was observed: the higher the HLB, the faster the convergence, i.e., the more rapidly the *ln* ρ' values would decrease. Since the polarity of the emulsifier increases with increasing HLB, the resultant greater ethanol interaction would tend to explain the trend.

The difference in ρ with changes in temperature was observed by Becher and Birkmeier (10), who speculated on the possible change of HLB with temperature. The change of polarity of liquid substrates with temperature changes was the subject of a paper by Petsev (12), who observed that the polarities of liquid substrates may increase, decrease or remain constant with increasing temperatures depending on structure.

Considering these facts, it can be seen that the same temperature must be used when measuring *ln* ρ' for different emulsifiers. When the retention data for *ln* ρ' values of various emulsifiers are plotted at different temperatures, good linear correlations still exist between *ln* ρ' and published HLB values. The slopes of the lines increase with increasing temperature. Thus theoretically any temperature could be used, provided the standard *ln* ρ' vs. known HLB plot was available at that temperature. Regression analysis of the data at other temperatures yielded the following equations and standard errors of estimate: $HLB_{40\ C} = 8.08\ ln\ \rho'_{40\ C} - 0.36$, 1.16 HLB units; $HLB_{60\ C} = 8.78\ ln\ \rho'_{60\ C} + 0.24$, 1.17 HLB units; $HLB_{80\ C} = 10.2\ ln\ \rho'_{80\ C} + 0.45$, 1.40 HLB units.

One practical consideration regarding the column temperature to be used is the state of the emulsifier on the

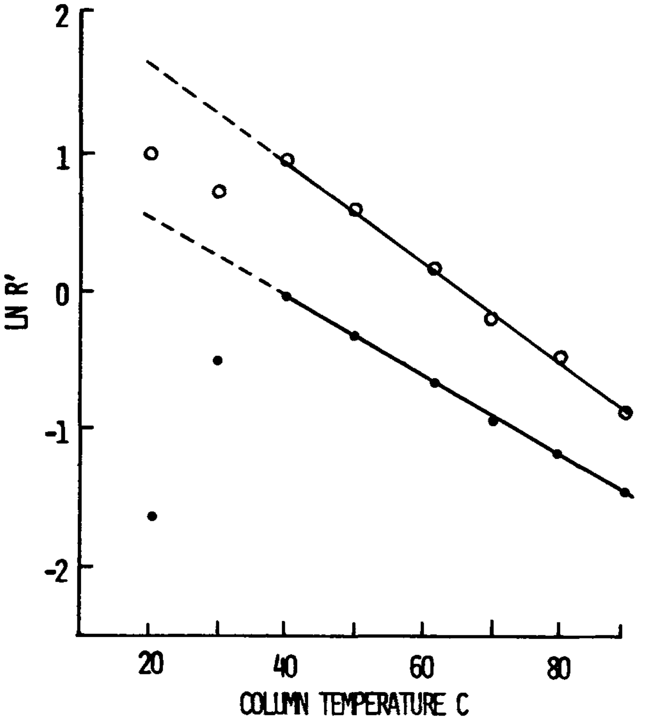

FIG. 5. Plot of $\ln R'$ for hexane (●) and ethanol (○) for sorbitan monopalmitate on 7 ft 5% Chromosorb W vs. column temperature.

column. When it is a liquid at room temperature, a good linear plot is obtained as in Figure 4. However, when the emulsifier is a solid such as sorbitan monopalmitate, pour point 48 C (Fig. 5), the linearity falls off at temperatures below its melting point, due to the fact that gas-solid interactions are much less than gas-liquid interactions. Thus for data as in Figure 5, where values below the mp of a particular emulsifier are needed, the straight line achieved at the higher temperatures has been extrapolated to the desired temperature, and this value is used. Use of the actual values rather than the extrapolated values will result in much greater scatter for the emulsifiers, which are solid at the temperature being considered.

Departures from linearity have also been observed at the other end of the range (>80 C) due possibly to thermal instability of the liquid substrate (emulsifier) at the high operating temperatures. However, with retention values taken over the entire temperature range, these deviations become readily apparent when plotted.

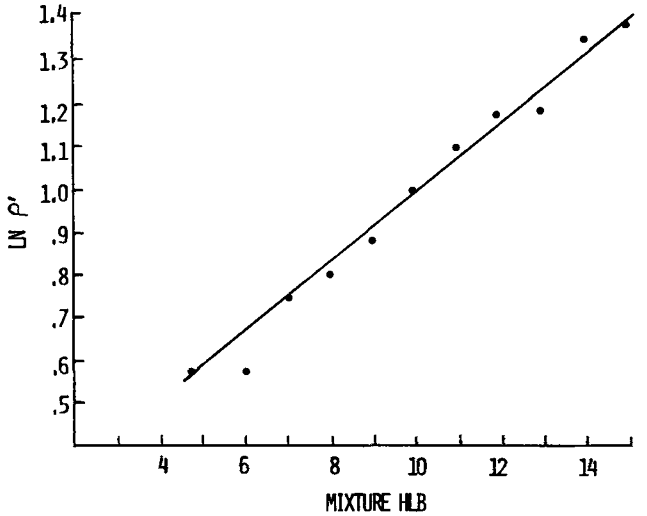

FIG. 6. Plot of $\ln \rho'$ observed for mixtures of POE sorbitan monostearate and sorbitan monostearate, for which composition of mixture was determined by relationship hydrophile-lipophile balance$_{\text{mixture}}$ = 14.9 (% POE sorbitan monostearate) + 4.7 (% sorbitan monostearate).

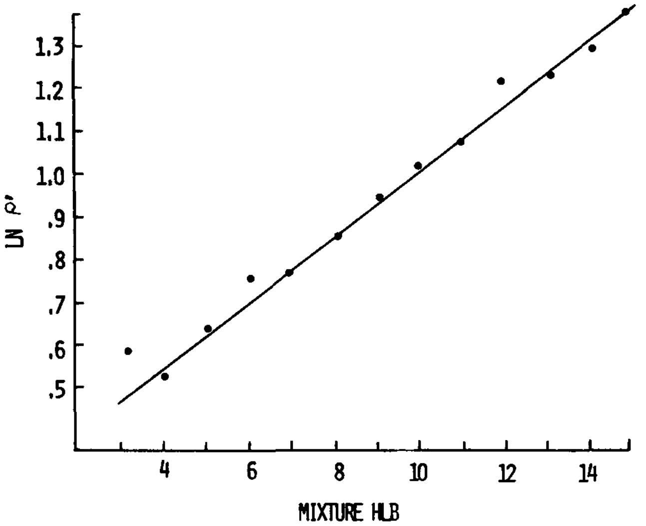

FIG. 7. Plot of $\ln \rho'_{70\ C}$ observed for mixtures of POE sorbitan monostearate and monodiglycerides, for which hydrophile-lipophile balance was determined by relationship hydrophile-lipophile balance$_{\text{mixture}}$ = 14.9 (% POE sorbitan monostearate)+ 3.2 (% monodiglycerides).

Having thus established the temperature relationship and the method of data treatment, other operational parameters needed to be considered: column length, flow rate, substrate loading, solid support and binary injection mixture composition. Using the liquid substrate POE (20) sorbitan monopalmitate, the effect of different column lengths (3, 5 or 7 ft) was shown to be negligible with respect to $\ln \rho'$ values (at 80 C $\ln \rho'$ was 1.66, 1.58 and 1.60 for 3, 5 and 7 ft columns, respectively). Obviously the retention values for hexane ($\ln R'_{\text{hexane}}$) and ethanol ($\ln R'_{\text{ethanol}}$) varied, but the difference ($\ln \rho' = \ln[R'_{\text{ethanol}}/R'_{\text{hexane}}] = \ln R'_{\text{ethanol}} - \ln R'_{\text{hexane}}$) remained constant within experimental error.

Using the 7 ft column of 5% POE (20) sorbitan monopalmitate at 80 C, variation of the helium flow rate of 20, 30 and 40 cc/min yielded $\ln \rho'_{80\ C}$ values of 1.56, 1.60 and 1.60, respectively; thus the effect is negligible.

The liquid loading of POE (20) sorbitan monopalmitate was investigated at levels of 2.5, 5, 10 and 25%, and the results again showed no deviation within experimental error of this parameter on the $\ln \rho'_{80\ C}$ values (1.48, 1.64, 1.55 and 1.60, respectively).

TABLE II

Properties of Solid Supports and Gas Chromatographic Behavior[a]

Solid support	Density, g/cc	Surface area, m^2/g	$\ln \rho'_{80\ C}$
Chromosorb W	0.24	1.0	1.60
Chromosorb P	0.47	4.0	1.32

[a] $\ln \rho'_{80\ C}$ values obtained using 7 ft column of 5% POE sorbitan monopalmitate.

TABLE III

Solvent Properties

Solvent	Boiling point, C	Dielectric constant
Water	100	78.5
Ethanol	78	24.3
Hexane	69	1.9
Methanol	64.7	33
3-Methyl-pentane	64	~2

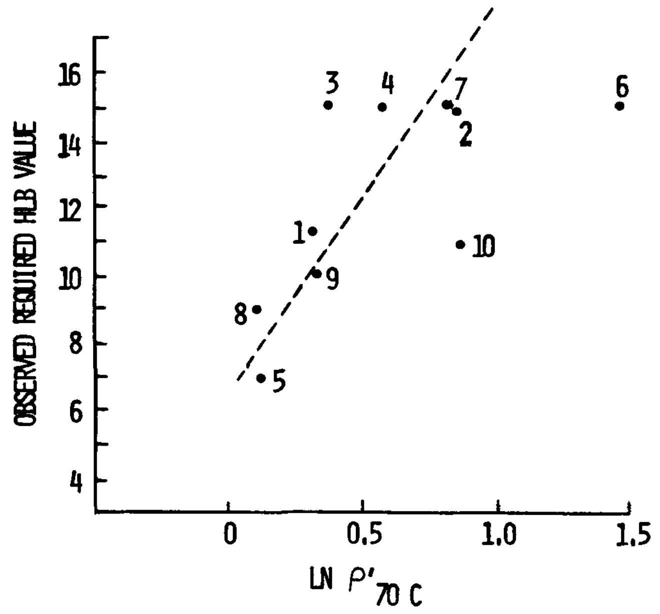

FIG. 8. Plot of observed required hydrophile-lipophile balance (HLB) vs. $\ln \rho'$ of various oils and fats.

Oil or fat	$\ln \rho'_{70}$	Observed required HLB	Published (15) required HLB
1. Isopropyl palmitate	.30	10	10
2. Castor oil	.83	14	14
3. Oleic acid	.35	14	16
4. Lauric acid	.56	14	16
5. Cottonseed oil	.11	6	6
6. Paraffin	1.49	14	10
7. Petroleum jelly	.80	14	7-8
8. Coconut oil	.10	8	
9. Corn oil	.33	9	
10. Soybean oil	.85	10	

From these studies, the need to use corrected values for retention times (that is R_{EtOH}, R_{hexane}-R_{air}) becomes apparent. If simple retention values were used (R_{hexane}, R_{ethanol}), the consistency of the data to variations of column length, flow rate and substrate loading was not observed. Therefore, by treatment of the data in the manner described, interlaboratory variations in $\ln \rho'$ values should be minimized.

The effect of solid support was investigated by the use of 5% POE (20) sorbitan monopalmitate on Chromosorb P in addition to Chromosorb W, which was generally used. The values obtained and some pertinent physical characteristics of the solid supports are shown in Table II. In this case, a difference in $\ln \rho'_{80\ \mathrm{C}}$ was obtained that was greater than experimental error; thus variations in solid support cannot be tolerated. The differences were probably due to the physical properties shown; however they were not pursued further.

The choice of binary injection mixture provided some latitude for experimentation. Ideally the mixture should contain a polar and nonpolar component. Water, being the most polar, presented problems in that its behavior on most columns is poor: broad unsymmetrical peaks with tailing, long retention times and immiscibility with the nonpolar liquids make injection of mixtures difficult. In the mixture used, hexane and ethanol were miscible and exhibited polarity differences that produced the observed results. Other mixtures that had been used previously were butane-butadiene (13) and benzene-cyclohexane (12).

Another mixture that appeared promising was methanol and 3-methyl-pentane, which have similar boiling points and diverse dielectric constants (polarity). These factors could result in greater retention time differences due to polarity and less effect due to boiling point differences.

When this injection mixture was used with the same previously used emulsifier columns, the results plotted as in Figure 3 were essentially the same as the hexane-ethanol results, with the individual points displaced just slightly resulting in no better or slightly worse correlation.

The subject of emulsifier mixtures has not progressed from an art to a well defined science as rapidly as the subject of emulsifiers themselves. The desirability and even necessity for a mixture of emulsifiers rather than a single one appears based on early observations by Schulman and Cockbain (14). They observed that a mixed interfacial film formed by simultaneous adsorption of a water soluble and oil soluble emulsifier resulted in an emulsion of greater stability, than one formed by either component separately. Such an idea was reinforced through the years by published (15,16) and probably unpublished experiences of those "familiar with the art." It has been generally assumed that the HLB of a mixture of emulsifiers is the algebraic sum of the HLB values of the individual emulsifiers. Becher and Birkmeier (10), however, showed a curvature in what should have been a linear correlation of emulsifier mixture ρ values (as determined by GC data) vs. calculated mixture HLB values. In a study of the $\ln \rho'$ values of mixtures of POE sorbitan monostearate (HLB 14.9) and sorbitan monostearate (HLB 4.7) determined by GC vs. calculated HLB values, a good linear correlation was obtained. These were chosen because of the wide range in their reported HLB values. Figure 6 shows the results.

Because such ideal behavior might not be expected with chemically dissimilar mixtures, emulsifier combinations of POE sorbitan monostearate and Atmos-150 (a mixture of mono- and diglycerides, HLB 3.2) were evaluated with the results given in Figure 7. Thus, in mixtures of chemically similar or dissimilar emulsifiers, the resultant HLB values can be determined by the algebraic summation method. From Figures 6 and 7 it can be seen that the scatter present in the HLB range 8-11, as seen in Figure 2, is absent. This lack of scatter would indicate that, when the HLB value of the emulsifier (or mixture) is accurately known or controlled, the GC determination is equally precise. It is possible that the originally determined HLB values in this midrange are slightly in error, due possibly to solubility partitioning. The data of Gorman and Hall (6) likewise exhibited a similar midrange scatter when the log of the dielectric constant was plotted against published HLB values for a series of emulsifiers.

In general, the approach of determining the HLB value of an emulsifier or mixture of emulsifiers by GC is limited in scope by the following constraint: only emulsifiers that are liquid in the range 30-90 C can be examined, thus excluding the general class of higher melting ionic emulsifiers. Still to be determined is the validity of the assumption that HLB values obtained from determinations at elevated GC temperatures can be used as a guide for formulating emulsions at lower temperatures. Other apparent shortcomings stem from the limitations of the concept of HLB: no measure of the amount of emulsifier necessary to achieve emulsion stability can be gleaned, nor can it be determined which of the myriad number of single emulsifiers or mixtures of emulsifiers all possessing the same HLB value will yield optimum emulsion stability in each individual instance.

The question of different emulsifier interactions in emulsions has been touched upon by Boyd et al. (17). Titus and Mickle (18) observed no difference in emulsion stability of milk fat-water emulsions when using one or a mixture of emulsifiers both possessing the same HLB value. The area remains open to further investigation. Approaches toward finding emulsifiers most effective in stabilizing emulsions against agglomeration or coalescence and freeze-thaw or heat stresses still have to be made on an individual basis.

A logical extension of the GC approach would be its utilization in determining the emulsifier HLB value that would best stabilize a particular fat or oil, or its "required HLB value" (3). Initial published attempts to correlate GC data of fats and oils with required HLB values determined by wet methods failed (19). Attempts to correlate required HLB values with a logarithmic function of the dielectric constant of fats and oils met with mixed results (6,19). Our attempts to correlate $ln\ \rho'$ values obtained from fats and oils with published required HLB values were largely unsuccessful. The wide variation in the properties of some of the fats and oils among different batches and brands made a wet determination of the required HLB values desirable. The results are shown in Figure 8.

However several problems still remain: The gas chromatographic behavior of the fats and oils was not as good as with the emulsifiers. Reproducibility was sometimes a problem, as was separation of the two peaks on the chromatogram. Retention times, especially of the ethanol, was sometimes dependent on injection amounts. Sample purity from batch to batch varied, which could explain the variation in observed and published required HLB values. Nonetheless, with improvements in the methodology of measuring required HLB values by normal methods and refinements of GC techniques, this may prove a potentially useful tool to aid in determining conditions for maximum emulsion stability.

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