[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, CENTRAL STATE COLLEGE]

# *p*-PHENYLAZOBENZOYL CHLORIDE FOR IDENTIFICATION AND CHROMATOGRAPHIC SEPARATION OF COLORLESS COMPOUNDS. ALCOHOLS[1]

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Most reagents commonly used for the identification of alcohols (acid chlorides and isocyanates) are affected, in varying degrees, by moisture, and in some instances decompose on standing. We have found that *p*-phenylazobenzoyl chloride (m.p. 92–94°) is quite stable at room temperature for long periods of time. In the reaction with alcohols to form *p*-phenylazobenzoate esters, no special precautions need be taken to secure anhydrous materials or to exclude moisture from the reaction mixtures. The orange-to-red colored derivatives form easily by gently refluxing *p*-phenylazobenzoyl chloride with the alcohol in the presence of pyridine. The crystalline esters give substantially constant melting points over a range of less than two degrees after one recrystallization. A trace of *p*-phenylazobenzoic acid, which can not be completely removed by several recrystallizations, does not appreciably affect the melting points of the derivatives. Any acid mixed with the derivatives is very strongly adsorbed at the top of a column of activated alumina and therefore, is easily separable from the derivatives. The melting points and analyses in table I are from compounds which have been chromatographed and recrystallized from ethanol-water mixtures. The methyl ester is formed in a 46 % yield on refluxing for one hour a mixture of methanol and water (equal volumes) and the acid chloride.

Fig. 1 shows the melting points of homologous series of *p*-phenylazobenzoates plotted against the number of carbon atoms in the aliphatic part of the molecule. For the normal alkyl esters, except for the first four members, the melting points of the esters of an even number of carbon atoms in the aliphatic portion were higher than those of adjacent odd carbon atoms. A similar alternating effect has been observed for the 3,5-dinitrobenzoates (1) of normal alcohols. The *iso*-alkyl esters showed the same type of curve for the first four members. For the normal-2-alkyl esters, those of the three- and five-carbon atom alcohols were higher melting than that of the four carbon atom alcohol. Beyond this point the esters of alcohols in the $C_6$–$C_8$ range were oils at room temperature and the esters of the $C_9$–$C_{10}$ alcohols exhibited a similar melting point pattern as observed in the first three members of this series—esters of an odd number of carbon atoms in the aliphatic portion had higher melting points than those of adjacent even carbon atoms. The melting points of the derivatives of isomeric alcohols, *n*-butyl—isobutyl, *n*-amyl—isoamyl, and *n*-hexyl—isohexyl were very close. However,

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TABLE I

MELTING POINTS AND ANALYSES OF THE ESTERS OF *p*-PHENYLAZOBENZOIC ACID

| Alcohol Used | M.P., °C. Corrected | Yield, % | Formula | Nitrogen[a] | |
|---|---|---|---|---|---|
| | | | | Calc'd | Found |
| Methanol | 126.0–126.8 | 90 | $C_{14}H_{12}N_2O_2$ | 11.66 | 11.73 |
| Ethanol | 84.8– 86.2 | 93 | $C_{15}H_{14}N_2O_2$ | 11.01 | 10.83 |
| 1-Propanol | 65.0– 65.8 | 74 | $C_{16}H_{16}N_2O_2$ | 10.44 | 10.36 |
| 1-Butanol | 64.6– 65.4 | 76 | $C_{17}H_{18}N_2O_2$ | 9.92 | 10.18 |
| 1-Pentanol | 54.8– 55.8 | 52 | $C_{18}H_{20}N_2O_2$ | 9.46 | 9.61 |
| 1-Hexanol | 61.0– 61.8 | 83 | $C_{19}H_{22}N_2O_2$ | 9.03 | 9.08 |
| 1-Heptanol | 53.2– 54.0 | 64 | $C_{20}H_{24}N_2O_2$ | 8.64 | 8.59 |
| 1-Octanol | 56.8– 57.6 | 68 | $C_{21}H_{26}N_2O_2$ | 8.28 | 8.28 |
| 1-Nonanol | 53.6– 54.2 | 65 | $C_{22}H_{28}N_2O_2$ | 7.95 | 7.97 |
| 1-Decanol | 59.0– 59.6 | 52 | $C_{23}H_{30}N_2O_2$ | 7.65 | 7.51 |
| 2-Methyl-1-propanol | 66.2– 67.6 | 31 | $C_{17}H_{18}N_2O_2$ | 9.92 | 9.60 |
| 3-Methyl-1-butanol | 52.4– 54.2 | 62 | $C_{18}H_{20}N_2O_2$ | 9.46 | 9.67 |
| 4-Methyl-1-pentanol | 61.2– 62.6 | 46 | $C_{19}H_{22}N_2O_2$ | 9.03 | 9.05 |
| 5-Methyl-1-hexanol | 44.4– 46.0 | 35 | $C_{20}H_{24}N_2O_2$ | 8.64 | 8.03 |
| 6-Methyl-1-heptanol | 32.2– 33.4 | | $C_{21}H_{26}N_2O_2$ | 8.28 | |
| 2-Propanol | 87.0– 87.4 | 53 | $C_{16}H_{16}N_2O_2$ | 10.44 | 10.36 |
| 2-Butanol | 48.8– 50.0 | 50 | $C_{17}H_{18}N_2O_2$ | 9.92 | 9.83 |
| 2-Pentanol | 50.0– 51.0 | | $C_{18}H_{20}N_2O_2$ | 9.46 | 9.71 |
| 2-Hexanol | oil[b] | | $C_{19}H_{22}N_2O_2$ | 9.03 | |
| 2-Heptanol | oil[b] | 47 | $C_{20}H_{24}N_2O_2$ | 8.64 | |
| 2-Octanol | oil[b] | 45 | $C_{21}H_{26}N_2O_2$ | 8.28 | |
| 2-Nonanol | 44.5– 46.5 | 57 | $C_{22}H_{28}N_2O_2$ | 7.95 | 8.00 |
| 2-Decanol | 36.0– 37.6 | 47 | $C_{23}H_{30}N_2O_2$ | 7.65 | 8.41 |
| Cyclohexanol | 88.0– 88.4 | 27 | $C_{19}H_{20}N_2O_2$ | 9.07 | 9.11 |
| Ethyl glycol monoethyl ether | 66.6– 67.6 | 67 | $C_{17}H_{18}N_2O_3$ | 9.39 | 9.05 |
| Benzyl | 91.2– 92.0 | 24 | $C_{20}H_{16}N_2O_2$ | 8.85 | 9.17 |

[a] Microanalyses by the Dumas Method were performed by the Du Good Chemical Laboratories, St. Louis, Missouri. [b] Oil at room temperature which did not solidify on cooling.

mixture melting points taken with these isomeric pairs gave an appreciable depression.

The use of these alkyl *p*-phenylazobenzoates as derivatives for the identification of low molecular weight alcohols appears to have limited application. Table I shows that a very large number of these esters melt within the narrow range of 40–60°.

Since the *p*-phenylazobenzoates were colored substances varying from orange to deep red, they seemed to afford a method of separation of closely related alcohols through the technique of adsorption chromatography. No particular study of the specific absorption bands, which they appear to furnish, was attempted by the authors.

There has been a very limited study of the separation of aliphatic alcohols in chromatographic procedures by use of their colored derivatives. Colored urethans (2) have been prepared in the reaction between a few aliphatic primary alcohols

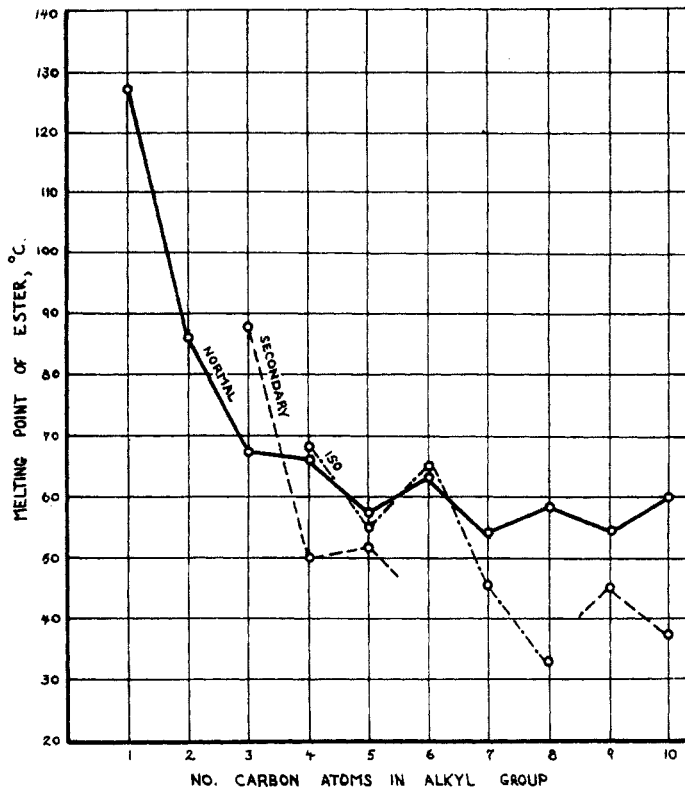


FIG. 1. MELTING POINTS OF ALKYL ESTERS OF *p*-PHENYLAZOBENZOIC ACID

and *p*-phenylazophenyl isocyanate. Mixtures of these urethans have been separated into the respective components by the chromatographic technique. Strain (3) suggested that colorless alcohols may be identified and separated by chromatographic adsorption through the colored esters of keto acid dinitrophenylhydrazones. However, chromatographic studies of colored derivatives of higher molecular weight compounds containing alcoholic hydroxyl groups have been carried on rather extensively. Chromatographic adsorption separations have been reported for colored esters formed in the reaction of *p*-phenylazobenzoyl chloride with terpene alcohols (4), triterpenes (5), sterols (6), and sugars (7).

The *p*-phenylazobenzoates of low molecular weight aliphatic alcohols have been found to be suitable derivatives for chromatographic adsorption separations on alumina. The individual esters moved down a column of alumina with increasing rate when Skellysolve-B, benzene, and ethyl acetate were used as solvents respectively in the order listed. By means of these derivatives it was possible to separate and identify many closely related aliphatic alcohols.

Table II shows the results obtained by the adsorption of sixteen pairs of *p*-phenylazobenzoates involving ten *n*-alkanols, three *n*-2-alkanols, and three iso-alkanols. Two pairs were separated sufficiently to make two zones visible

TABLE II

BINARY MIXTURES OF ESTERS SEPARATED INTO ZONES

methyl—isobutyl
methyl—isoamyl

BINARY MIXTURES OF ESTERS FORMING CONTINUOUS BAND

| | |
|---|---|
| methyl—*n*-butyl | *n*-butyl—*n*-heptyl |
| *n*-hexyl—*n*-decyl | methyl—2-propyl |
| ethyl—2-butyl | isoamyl—*n*-octyl |
| isoheptyl—*n*-nonyl | isobutyl—*n*-octyl |

BINARY MIXTURES OF ESTERS GIVING NO SEPARATION

| | |
|---|---|
| *n*-propyl—2-propyl | 2-pentyl—*n*-octyl |
| isobutyl—*n*-butyl | methyl—ethyl |
| methyl—*n*-propyl | *n*-butyl—*n*-amyl |

with a colorless band between. Eight pairs formed a continuous band. Sectioning of this with subsequent elution yielded an almost homogeneous top and bottom section with intervening sections of varying composition. Six pairs did not give satisfactory separation under the conditions employed. The first member of each pair listed in Table II, wherein separation was obtained, was the most strongly adsorbed derivative. As expected the higher molecular weight derivative formed the lower or least readily adsorbed band. The separations as indicated were confirmed by melting points of the materials obtained from the chromatogram. The individual esters were purified chromatographically before mixing. It is quite possible that further investigation of the chromatographic properties of these esters on other adsorbents might provide a larger number of resolutions.

## EXPERIMENTAL

*Alcohols.* The *n*-alkanols, 2-propanol, 2-butanol, 2-hexanol, 2-octanol, isobutyl alcohol, isoamyl alcohol, cyclohexanol, ethyl glycol monoethyl ether, and benzyl alcohol were good commercial grades which were used without further purification. The 2-heptanol (b.p. 155–157°, $n_D^{16}$ 1.4170) was synthesized by reducing methyl *n*-amyl ketone according to the method of Whitmore (8). The remaining alcohols were synthesized by the method of the Grignard reaction using the following reagents: *n*-propyl bromide and acetaldehyde gave 2-pentanol b.p. 116–119°, $n_D^{16}$ 1.4061; *n*-octyl bromide and acetaldehyde gave 2-decanol b.p. 108–109° at 13 mm., $n_D^{16}$ 1.4351; *n*-heptyl bromide and acetaldehyde gave 2-nonanol b.p. 192–198°, $n_D^{27}$ 1.4272; 1-bromo-2-methylpropane and ethylene oxide gave 4-methyl-1-pentanol b.p. 147–153°, $n_D^{27}$ 1.4200; 1-bromo-3-methylbutane and ethylene oxide gave 5-methyl-1-hexanol b.p. 168–171°, $n_D^{26}$ 1.4230; 1-bromo-5-methylhexane (b.p. 169–171°, $n_D^{23}$ 1.4470 prepared on refluxing 5-methyl-1-hexanol with 48% hydrobromic acid) and formaldehyde gave 6-methyl-1-heptanol b.p. 185–190°, $n_D^{24}$ 1.4453.

*Preparation of esters.* A mixture of acid chloride[4] (approximately 0.1 g.), alcohol (excess), and a few milliliters of pyridine was refluxed gently for 1 hour. The red solution was poured with vigorous stirring into ice and 10% sodium bicarbonate solution. The mixture was left standing for a few minutes. The methyl, ethyl, isopropyl, and ethoxyethyl esters crystallized instantly. They were filtered, washed with water, and air-dried. Those esters which separated as oils were extracted with ether. The ether extracts were washed with water and

[4] Commercially available from Eastman Kodak Company.

saturated sodium chloride solution, dried over sodium sulfate and the ether was removed. The crude ester was dissolved in the minimum volume of Skellysolve-B or a mixture of Skellysolve-B and benzene and chromatographed on activated alumina on which the free acid was strongly adsorbed. The ester was eluted with absolute ethanol and the solvent was removed. The yield of orange-to-red colored esters ranged from 24 to 93%. The alkanol derivatives crystallized from a mixture of ethanol and water as thin fluffy plates, exhibiting varying degrees of static charge. The cyclohexyl, ethoxyethyl, and benzyl esters crystallized from a mixture of ethanol and water to give soft, fine needles. The esters of the $C_6$–$C_8$ *n*-2-alkanols could not be induced to crystallize.

*Methyl p-phenylazobenzoate.* A mixture of 0.1834 g. of acid chloride, 10 ml. of absolute methanol, and 10 ml. of distilled water was refluxed for 1 hour. The reaction mixture was poured into ice and 10% sodium bicarbonate solution. The red solid which deposited was collected. On chromatographing and recrystallizing from 95% ethanol, this material gave red plates (46% yield), m.p. 121.2–124.8°.

*Chromatographic separations.* A typical chromatographic separation on a mixture of two components was conducted as described below. A column 20 mm. by 40 cm. was used. Activated alumina (grade F-20) was packed in the column by filling it several times with the adsorbent while tapping the sides of the column with cork rings and tamping the surface of the adsorbent with a plunger. The adsorbent then was wetted with Skellysolve-B.

The mixture of esters (10 to 20 mg. of each component) was dissolved in the minimum volume of warm Skellysolve-B or a mixture of Skellysolve-B and benzene from which they were adsorbed on the column. The chromatogram was developed by passing Skellysolve-B, 5% benzene in Skellysolve-B and 2% ethyl acetate in Skellysolve-B through the adsorbent in the order listed. The adsorbent was pushed out and cut into bands or the bands were dug out of the column by a long narrow spatula, and the bands were eluted with 95% ethanol. When a continuous band was obtained, the band was arbitrarily cut into several sections. The pure components were obtained from the top and bottom sections and the intervening sections were mixtures. The eluents were concentrated, filtered into a tared flask, and the last traces of solvent were removed *in vacuo* under a stream of nitrogen. Melting points of the residues were determined. In several cases it was necessary to triturate the oily residues with traces of a mixture of alcohol and water to induce crystallization.

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SUMMARY

The *p*-phenylazobenzoates of the *n*-alkanols (from methyl to decyl), the isoalkanols (from isobutyl to isooctyl), the *n*-2-alkanols (from propyl to decyl), cyclohexanol, ethyl glycol monoethyl ether, and benzyl alcohol have been prepared and are of value for identification purposes.

These esters are colored and certain mixtures of them can be separated by chromatographic adsorption.

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