

**Reaction of  $\beta$ -Hydroxypropionic Acid and Sodium Cyanide.**—The procedure in this experiment was identical with that described above for  $\beta$ -hydroxypropionitrile.  $\beta$ -Hydroxypropionic acid was first prepared by hydrolyzing the corresponding nitrile,<sup>2</sup> and the acid obtained was used without further purification. The amounts of  $\beta$ -hydroxypropionic acid, sodium cyanide and water used were 2.65 g., 8.62 g. and 15 ml., respectively. The succinic acid obtained in this case weighed 0.51 g. (14.9%) and gave m.p. of 181–182°.

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## NEW COMPOUNDS

### *p*-(2-Ethylhexoxy)-benzoic Acid

A mixture of 42 g. (0.2 mole) of 2-ethylhexyl bromide,<sup>1</sup> 28 g. (0.2 mole) of *p*-hydroxybenzoic acid, 22 g. (0.4 mole) of potassium hydroxide, 150 cc. of alcohol and 50 cc. of water was refluxed for 24 hours (two layers). In order to hydrolyze any ester which may still have been present, some 10% aqueous potassium hydroxide solution was added and the mass refluxed for another 30 minutes. Acidification gave an oily product which was isolated and distilled first under 35 mm. pressure (245°), then under 0.5 mm. (180°). The acid solidified easily and crystallized from a large amount of 80% formic acid in soft, white leaflets, which are

(1) Ch. Weizmann, E. Bergmann and L. Haskelberg, *Chem. and Ind.*, **56**, 1587 (1937).

converted at 60–61° into "liquid crystals," in analogy to the behavior of *p*-butoxybenzoic acid.<sup>2</sup>

*Anal.* Calcd. for  $C_{15}H_{22}O_3$ : C, 72.0; H, 8.8; mol. wt., 250. Found: C, 72.4; H, 9.1; mol. wt., 255 (titration).

(2) Bradfield and Jones, *J. Chem. Soc.*, 2660 (1929).

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### 2-Chlorophenyl-1-naphthylcarbinol<sup>1</sup>

**2-Chlorophenyl-1-naphthylcarbinol.**—A Grignard reagent was prepared from 11.6 g. of magnesium, 100 ml. of  $\alpha$ -bromonaphthalene and 300 ml. of dry ether. When the reaction was complete, a solution of 55.3 g. of *o*-chlorobenzaldehyde in 300 ml. of dry ether was added dropwise with stirring. The mixture was stirred overnight at room temperature and then heated under reflux for one hour, cooled, and finally decomposed with 75 ml. of cold, 20% ammonium chloride solution. The ethereal solution was decanted from the hard residue and the residue was washed twice with ether. The combined ether solutions were washed with water, dried over "Drierite," and finally concentrated. The residue was fractionated under reduced pressure and the fraction distilling at 215–216° (4 mm.) was collected; yield 89.5 g. (85%).

A small portion of the above viscous oil was crystallized three times from dilute ethanol to give soft white needles; m.p. 96–97°.

*Anal.* Calcd. for  $C_{17}H_{13}OCl$ : C, 75.98; H, 4.88. Found: C, 76.16; H, 4.91.

(1) This compound is a previously unisolated intermediate in the synthesis of 1-(*o*-chlorobenzyl)-naphthalene by the method of Bradsher, *This Journal*, **62**, 1077 (1940).

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## COMMUNICATIONS TO THE EDITOR

### THE MOLECULAR WEIGHT DETERMINATION OF POLYPEPTIDES

Sir:

In the course of our attempts to purify and characterize certain of the higher polypeptides by counter-current distribution it has become apparent that any change affecting a carboxyl or amino group produces a striking effect on the partition ratio of the peptide. Accordingly, the derivatives representing different stages of substitution which result from incomplete reaction can be separated readily. Since with unchanged peptide remaining, the band occurring in the distribution pattern nearest the unchanged substance would be the monosubstituted derivative, a general approach to the problem of molecular weights is suggested. Molecular weight determination by analysis for substituting groups has long been a standard procedure but a decision as to the numbers of groups involved has often been equivocal.

For instance, if a polypeptide with one free  $-NH_2$  is treated with sufficient 2,4-dinitrofluorobenzene<sup>1</sup>

(1) F. Sanger, *Biochem. J.*, **39**, 507 (1945).

so that only a fraction of it is converted, distribution of the resulting mixture in a suitable system gives two bands, one of the unchanged polypeptide and a yellow band of the DNP derivative. Determination of the distribution pattern by weight and by absorption at 350  $m\mu$  now permits calculation of the molecular weight, provided Beer's law holds. The value  $\epsilon$  of  $\delta$ -DNP-ornithine hydrochloride (16,250) can be used as the basis of calculation.

If the peptide contains two amino groups, four bands in the pattern are possible. One band containing the disubstituted product will be furthest removed from that containing the unchanged material. Two possible intermediate mono-substituted products would be expected to have very similar partition ratios and could form overlapping bands unless a very high number of transfers had been applied. Across any one colored band the ratio of weight to absorption should be, and in our experience thus far has been, constant.

With three amino groups in the molecule, two families of overlapping bands and one of the completely substituted peptide could occur. However,