lated by the procedure of Fischer.[7] The ratioactivity of each hemin sample was measured in duplicate. The radioactive CO_2 was precipitated and measured as $BaCO_3$ after the addition of a known amount of carrier sodium carbonate to each sample. All samples were counted in a windowless flow counter and the reported counts per minute were corrected to infinite thinness.

The results in Table I show that (1) heme is synthesized in the red cells of vitamin B_6-deficient ducklings from glycine-2-C-14 at a rate which is half or less than half of that found with control ducklings; (2) addition of pyridoxal-5-phosphate *in vitro* restores the ability of the deficient cells to synthesize heme at a normal rate; (3) there is no stimulatory effect of pyridoxal-5-phosphate on heme synthesis by normal duckling cells; and (4) the addition of pyridoxal-5-phosphate accelerates the conversion of glycine-2-C-14 to $C^{14}O_2$ in both normal and deficient red cells. Since hemolysates of duck cells also show a stimulation of pyridoxal-5-phosphate on heme synthesis, the observed effect is not dependent on the presence of intact cells.

TABLE I

HEME SYNTHESIS AND CO_2 PRODUCED BY THE INCUBATION OF GLYCINE-2-C-14 WITH DUCK BLOOD

Each vessel contained 2 ml. of heparinized blood from either vitamin B_6-deficient or control ducklings and 0.1 ml. of glycine-2-C-14 (23.5 μM.; 230,000 c.p.m./μM.). In addition, 1 mg. of crystalline pyridoxal-5-phosphate monohydrate in 0.1 ml. of saline was added to appropriate flasks and 0.1 ml. of saline was added to the others. Values given are the averages ± standard errors obtained from 4 deficient and 4 control ducklings. The p-values for 1, 2 and 3 are <0.01, >0.2 and <0.01, respectively, when calculated without regard to the paired nature of the data. When calculated by matched pair formula,[8] the p-values for the stimulatory effects of pyridoxal-5-phosphate are between 0.01 and 0.05 for 1 and 2 and less than 0.01 for 3.

	Hemin, c.p.m./mg.		C.p.m./total collected CO_2	
	No additions	Plus pyridoxal-5-phosphate	No additions	Plus pyridoxal-5-phosphate
Vitamin B_6-deficient	535 ± 83	1185[1] ± 147	2656 ± 616	3790[2] ± 520
Control	1309 ± 112	1256 ± 102	4370 ± 344	7478[3] ± 391

Results with succinate were similar to those found with glycine. The incorporation of sodium succinate-2,3-C-14 into heme was depressed in vitamin B_6-deficient duck blood whole cells and hemolysates, and stimulated by added pyridoxal-5-phosphate. However, δ-aminolevulinic acid-2,3-C-14 was incorporated equally well into the hemes of the B_6-deficient and control bloods and was not stimulated by added pyridoxal-5-phosphate. Since δ-aminolevulinic acid is a porphyrin precursor formed from glycine and succinate,[9,10] it appears that pyridoxal-5-phosphate acts specifically in the formation of δ-aminolevulinic acid from glycine and succinate.

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DEGRADATION OF AMYLOPECTIN TO NIGEROSE

Sir:

Although the α-D (1→4) linkage is the principal glycosidic bond in amylopectin with branching occurring through α-D-glucopyranosidic (1→6) bonds, some linking at positions other than 4 and 6 is not excluded. Assuming complete reaction, the detection of a small amount of glucose by paper chromatography in the hydrolysate of periodate-oxidized (with subsequent reduction) amylopectin furnishes analytical evidence[1] for the presence of a small number of (1→3) or of both (1→2) and (1→4) linkages in amylopectin. We wish to present definitive evidence herein for the existence of an α-D-(1→3)-bond in amylopectin. This evidence consists of the isolation of nigerose (3-*O*-α-D-glucopyranosyl-D-glucose) as its crystalline β-D-octaacetate from an amylopectin hydrolysate produced under conditions which are known to minimize its formation by reversion to a negligible quantity.[2] A 0.4% solution of amylopectin (130 g., waxy maize starch) in 0.1 *N* hydrochloric acid, was hydrolyzed by heating at 97° to 67% completion. This hydrolysate, after removal of the acid by ion-exchange resin, was subjected to fractionation on a carbon (Nuchar C, unground) column by the general method of Whistler and Durso.[3] The fraction known to contain maltose and isomaltose was acetylated to give 40 g. of sirupy material from which most of the maltose was removed as β-maltose octaacetate by direct crystallization from ethanol, yield 18 g., m.p. 155–156°, $[\alpha]^{28}D$ +64° (*c* 4.5, chloroform). The material from the mother liquor was subjected to fractionation by silicate column extrusion chromatography and produced β-isomaltose octaacetate, 1.67 g., m.p. 144–146°, $[\alpha]^{30}D$ +98° (*c* 4.4, chloroform), and β-nigerose octaacetate, 350 mg., m.p. 140–145°, $[\alpha]^{25}D$ +80° (*c* 3.0, chloroform), X-ray powder diffraction pattern identical with that of known β-nigerose (sakébiose[4]) octaacetate[5] ("*y*-acetate"[6]). Upon further purification the melting point was 151–153°.

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SYNTHESIS OF PHTHALIMIDINES FROM SCHIFF BASES AND CARBON MONOXIDE

Sir:

When a solution of 5 g. of benzaldehyde anil in 50 ml. of benzene was heated with 1 g. of dicobalt octacarbonyl catalyst[1] under 100–200 atmospheres pressure of carbon monoxide at 220–230° for 5–6 hours, 2-phenylphthalimidine (I), m.p. 263°, was obtained in 80% yield.

Anal. Calcd. for $C_{14}H_{11}ON$: C, 80.38; H,

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