formation of an acid containing two primary amino groups from the acidic biotin, the inactivation of biotin with nitrous acid without evolution of nitrogen are all in keeping with the interpretation which we placed on the action of $Ba(OH)_2$ on biotin. We should now like to report the resynthesis of biotin from this diaminocarboxylic acid.

If our interpretation of the degradation reaction were correct, it should be possible to convert the diaminocarboxylic acid back to biotin through reactions employed for the synthesis of urea derivatives. Accordingly 10 mg of the diaminocarboxylic acid were treated with phosgene under conditions ordinarily employed. Crystalline biotin was obtained from the reaction mixture in 98 per cent. yield. The compound melted at 228–230° (uncorrected), which agrees with that recorded by us for natural biotin.³ The melting point of a mixture of the synthetic compound with biotin isolated from natural sources showed no depression. The specific rotation of the resynthesized biotin was $[\alpha]_{22}^{22} = +92^{\circ}$ (0.2 per cent. solution in 0.1 N NaOH). By treatment of the synthetic compound with diazomethane, a methyl ester (m.p. 166-167°) was formed which showed no depression in melting point when mixed with a sample of biotin methyl ester. As tested by the yeastgrowth method the synthetic biotin exhibited the same degree of activity as natural biotin.⁴ Since the resynthesized biotin is identical in melting point, optical activity and biological potency with the natural product, it is obvious that little or no racemization could have taken place during the Ba(OH)₂ treatment of biotin. The synthesis of biotin from the diamino compound affords additional and conclusive proof for the cyclic urea structure in biotin. The possible relation of the urea structure of biotin to the affinity of biotin for avidin is being subjected to experimental test.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

AN INEXPENSIVE SQUARE-WAVE GENERATOR

BECAUSE the square wave contains an infinite series of harmonics of its fundamental frequency, and because its precise wave-form is readily recognizable on an oscilloscope it is an easily applied severe test for an amplifier. It shows at a glance the high and low frequency cut-offs, other frequency and phase discrimination, resonance, overshoot, etc.

The generator here described is light, compact,

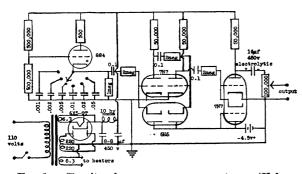


FIG. 1. Circuit of square-wave generator. (Values are not critical.) The 100,000 ohm output potentiometer should be tapered as for an audio gain control.

simple and portable. Its principle is to feed the output of an oscillator first into a limiter which clips off the peaks and gives waves with flat tops and bottoms, thence into an amplifier and second limiter, etc. The

⁸ V. du Vigneaud, K. Hofmann, D. B. Melville and J. Rachele, Jour. Biol. Chem., 140: 763, 1941.

sides of the wave become steeper with each amplifier stage.

Frequency stability is provided by the thyratron oscillator. The fundamental is variable in six steps from 35 to 1,200 cycles. The grids of the amplifier tubes together with the diodes constitute effective limiting circuits. The output is variable from 40 volts to less than 100 microvolts by a single control. The rates of rise in the two sides of the square wave are not exactly equal, but the slow one is faster than 10 microseconds. Thus the generator will test an amplifier to over 50,000 cycles.

The power transformer should be a well-shielded one, and should be mounted a few inches away from the thyratron tube to prevent magnetic action on the latter.

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AN ADJUSTABLE RESISTOR FOR FLOWMETERS

ONE of the most common types of flowmeters for air is that consisting essentially of a resistor and a gage for measuring the pressure drop across it. The pressure gage may be a manometer with its two arms connected to the air line, one ahead of and the other following the resistor; or if one end of the resistor is open to the atmosphere, so may be the correspond-

⁴ We wish to express our appreciation to Miss Eleanor Hague of this laboratory for carrying out the assays.