It can be seen from Table 2 that the condensing enzyme in all the fractions is inhibited by magnesium ion whereas acetyl coenzyme *A* deacylase is activated. It will also be observed that the purified condensing enzyme is still found to be inhibited by magnesium ion. This suggests that the condensing enzyme of *A. niger* may be different from that obtained from other sources in respect of its magnesium requirement.

My thanks are due to Miss Mary Clements, Division of Applied Biology, National Research Council of Canada, for the gift of the strain of *A. niger*, *N.R.C.* 233, and lyophilized *E. coli*.

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A Rapid Paper Chromatographic Procedure for the Quantitative Determination of Hydroxyproline

In a recent communication[1], we described a descending paper chromatographic procedure for the quantitative determination of hydroxyproline which required a solvent development of 40 hr. Using horizontal paper chromatography at an elevated temperature, comparable results have been obtained after development for only 4 hr.

Four standard hydroxyproline solutions (*p*H 6·5) containing, respectively, 0·050, 0·075, 0·100 and 0·125 μgm. of amino nitrogen are applied to the paper in 0·5 μl. volumes. The unknown solution is spotted (in 0·5 μl. volumes) to fall within the 0·50–0·125 μgm. range of the standard solutions.

Whatman No. 1 filter paper buffered[2] at *p*H 8·4 and measuring 12 cm. × 17 cm. is used. The origin is 2·5 cm. from one end. A 1·5-cm. tab, folded at right angles to the paper, is inserted into the solvent trough.

The chromatographic chamber and the manner in which the filter paper strip is supported in a horizontal position have been described[3,4].

The developing solvent is prepared at 22° C. in a temperature-controlled room by equilibrating equal volumes of redistilled *m*-cresol (Matheson Co., Inc., practical grade) and *p*H 8·4 buffer (0·067 *M*)[2]. The buffer solution must be within ± 0·1 *p*H to obtain the separation reported here. The solvent-rich phase is placed in the trough and the bottom of the chamber is covered with the buffer-rich phase. The covered chamber is housed in a mechanical convection oven at a temperature of 60° C.

After spotting, the filter paper strip is placed in the chamber and a solvent development of 4 hr. at 60° C. separates hydroxyproline from the other common amino-acids. Following solvent development, the chromatogram is dried for 1 hr. at 60° C. using a mechanical convection oven, dipped in a ninhydrin–isatin solution, and the colour of the hydroxyproline spots is developed by placing the chromatogram in an oven at 60° C. for 15 min. The colour reagent is composed of 270 mgm. of ninhydrin, 130 mgm. of isatin and 2 ml. of triethylamine made up to 100 ml. with water-saturated *n*-butanol. The merits of this hydroxyproline reagent have been discussed previously[5].

The maximum densities of the hydroxyproline spots are measured with a 'Densichron' transmission densitometer (Model 3835 *B*, W. M. Welch Scientific Co., Chicago) using a 1-mm. diameter aperture and no filter. A standard curve is prepared by plotting the logarithm of the known concentrations against the densities. The concentration of hydroxyproline in the unknown solution is then calculated by referring to the standard curve. The standard curves are not reproducible from one paper strip to another, and the standards must be run each time on the same chromatogram with the unknown.

Table 1. Accuracy and Precision of the Rapid Horizontal Paper Chromatographic Determination of Hydroxyproline

Chromatogram	Calculated concentration (mgm./100 ml.)
1	48·7
2	48·7
3	49·6
4	48·7
5	45·9
6	47·7
7	49·6
8	44·0
9	46·8
Average	47·7
Standard deviation	2·21
Deviation from theoretical value, 46·8 mgm./100 ml. (per cent)	1·9

The accuracy and precision of this rapid paper chromatographic procedure were established by determining the hydroxyproline content of a solution, *p*H 6·5, containing eighteen common amino-acids. The average value of nine chromatograms, 47·7 mgm./100 ml., differed from the theoretical value by 1·9 per cent (Table 1). The standard deviation was 2·21 mgm./100 ml. These values are in good agreement with an error of 0·9 per cent and a standard deviation of 1·41 mgm./100 ml. obtained when the solution was assayed using a descending paper chromatographic procedure[1]. The main advantage of the horizontal procedure is the shortening of the solvent development time from 40 hr. to 4 hr.

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Anæmia and Liver Damage in X-Irradiated Animals

A number of authors have found disturbances in various liver functions, damage to the liver in the form of fatty degeneration, and increase in the total liver fats in various mammals after total body irradiation with lethal doses of X-rays. According to Ellinger[1] these changes occur after irradiation with doses exceeding *LD*80, and the mechanism of their origin is not yet quite clear. Severe anæmia after irradiation, and resulting hypoxia of the liver and of other organs, might also occur. We have investigated this possibility, and have found that after total body irradiation with