We wish to thank Mr. E. B. Damon and Dr. W. A. Ray for the kind loan of apparatus used in this investigation.

HARVARD CHEMICAL LABORATORIES CAMBRIDGE, MASSACHUSETTS GEORGE DAVIS SCIENCE HALL KNOX COLLEGE GALESBURG, ILLINOIS RECEIVED JULY 18, 1938

## Changes in the Physical Properties of Regenerated Cellulose by Liquid Ammonia

## BY RICHARD G. ROBERTS

During the dialysis of proteins and hormones contained in bags made from regenerated cellulose film and placed in liquid ammonia, it was observed that the bags changed in size and in flexibility. Therefore, a series of comparative tests on some of the physical properties of the film and ammoniatreated film was made. The film used in these tests was du Pont Cellophane number 600.

The film was cut into strips of convenient size and placed in a Dewar flask. Liquid ammonia, previously dried over metallic sodium, was added to immerse the film sample completely. The film was not previously dried by us. The Dewar flask was tightly stoppered, and attached to a mercury seal. The liquid ammonia boiled off in about twenty-four hours, and any excess ammonia gas was removed by a vacuum pump.

Physical properties showing an increase:

Tear strength (Elmendorf test)	200.0%
Tensile strength	70.6%
Thickness (flat micrometer)	152.4%
Weight (per unit area)	27.3%
Physical properties showing a decrease: Length (with grain) Width (cross grain) Area	8.3% 17.8% 24.7%

Ratios: Increase of tear strength to decrease in area, approximately 8 to 1. Increase of tensile strength to decrease in area, approximately 3 to 1.

It has been shown that bags made from regenerated cellulose film may be used conveniently for dialyzing experiments in liquid ammonia, although some space must be allowed for shrinkage.

The author wishes to thank Montgomery Ward and Company, Chicago, Illinois, for the use of apparatus in their testing laboratory.

## The Specificity of the Fermentation Test for Vitamin B<sub>1</sub>

By Alfred S. Schultz, Lawrence Atkin and Charles N. Frey

The fermentation method for the determination of vitamin  $B_1^{1-3}$  has been in successful operation for some time. The effect of 2-methyl-5-ethoxymethyl-6-aminopyrimidine has been described.<sup>2</sup> We have assayed a wide variety of substances such as non-autoclaved yeast, rice polish, vitamin pills and concentrates, solutions of crystalline vitamin, and milk, and no evidence of the interfering substance has been found. In an investigation on the metabolism of vitamin B1,4.5 we found reason to believe that a portion of the fermentation stimulating effect of urine is not due to vitamin B<sub>1</sub>. While this did not appear to alter the significance of the results, it was thought very desirable to find a method for differentiating between the intact vitamin molecule and any possible breakdown product. A way of doing this has been found in the differential oxidation of the vitamin  $B_1$  in the presence of the aminopyrimidine.

Alkaline ferricyanide in the cold will readily oxidize the vitamin to thiochrome. Preliminary experiments with a sample of thiochrome obtained from Merck and Company showed it to be inactive in the fermentation reaction. The aminopyrimidine is more resistant to oxidation and it is a simple matter to oxidize  $B_1$  preferentially when present in addition to aminopyrimidine. A solution containing 8 gamma of the aminopyrimidine and 8 gamma of thiamin hydrochloride in a volume of 35 ml. was treated with 2.5 ml. of 1%K<sub>3</sub>Fe(CN)<sub>6</sub> and 2.5 ml. of 50% NaOH. After standing at room temperature for five minutes the solution was neutralized with dilute sulfuric acid and made to 100 ml. A 25-ml. aliquot of this was tested in the usual manner by gas test. It gave a stimulation which corresponded exactly to 2 gamma of the aminopyrimidine (*i. e.*, the  $B_1$ was destroyed). Parallel experiments showed that the neutralized oxidizing solution was without influence on controls with either thiamin hydrochloride or aminopyrimidine.

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