

## COMMUNICATION TO THE EDITOR

### A Quick Biuret Method for Protein in Wheat

DEAR SIR:

Since introduction of the rapid biuret method for protein in grains [Johnson and Craney, *CEREAL CHEMISTRY* 48(3): 276-282 (1971)] interest has been expressed in an even faster test for wheat. Although several samples can be tested at the same time by the above method, total *elapsed* time per sample is 35 to 40 min. For some purposes, such as testing wheat during loading, a prompt result on a single sample is needed.

The following procedure has been tested on wheat and appears to meet the need for a simple, accurate means of protein determination with an elapsed time of less than 10 min.

#### MATERIALS AND METHODS

##### Reagents

1. Alkaline-alcohol solution: In a 1,000-ml. volumetric flask place 5.6 g. potassium hydroxide pellets. Add 600 ml. isopropyl alcohol. Make to volume with distilled water.
2. Isopropyl alcohol (reagent grade).
3. Cupric carbonate (reagent grade).

##### Apparatus

1. Mill (Udy cyclone-hammer mill with 0.024-in. screen, or equivalent).
2. Balance.
3. Waring Blendor, laboratory model, with stainless-steel jar.
4. Vacuum-filter assembly (No. 3 Gooch crucible with two 2.1-cm. glass fiber filters).
5. Colorimeter (Leitz Photrometer or equivalent).
6. 50-ml. test tubes.
7. Electric interval timer.
8. Automatic pipet, 100 ml.
9. Pipettor, 2 ml.

##### Procedure

1. Grind a representative sample of about 25 g. of wheat and mix well.
2. Pipet 2 ml. isopropyl alcohol into blender jar.
3. Add 1.00 g. ground wheat. Shake to dampen sample.
4. Add 1.00 g. cupric carbonate.
5. Add 100 ml. alcohol-alkaline solution.
6. Blend at low speed (approximately 12,500 r.p.m.) for 4 min. Repeat steps 2 through 6 in second blender jar.
7. Filter about 10 ml. each sample through two glass fiber filters in Gooch crucible.
8. Using 10 × 10-mm. cell, read absorbance at 550 nm. Add duplicate values together and refer to standard curve for protein content.

One hundred twenty-five samples of wheat were tested by this method and compared with Kjeldahl protein determinations (Fig. 1). Two 1-g. portions of each ground-wheat sample were tested and the absorbance values added together for increased accuracy. Correlation was 0.995, standard error  $\pm 0.24\%$  protein. All classes of wheat were included in the study.

With two blenders so that the two portions can be blended simultaneously, time for testing one sample is less than 10 min.; with one blender, 13 min.

The metal blender jar should be rinsed in cold water and inverted to drain between samples. By using a metal container there is no heat buildup as there is with a glass jar. There is no need to dry the jar, as the 2 ml. of alcohol will prevent the grain from sticking to the wet jar or becoming lumpy.

Cost of reagents and filters is about 10 cents per test.

Because of the difficulty of obtaining cupric carbonate of uniform quality, several samples of wheat with known absorbance values should be checked when changing to a new lot of cupric carbonate, and a correction factor applied or a new standard curve calculated.

In establishing a standard curve for the analysis, select 35 or more samples having a good distribution over the normal protein range. A regression equation for the standard curve can be calculated using the absorbance readings and Kjeldahl protein contents.

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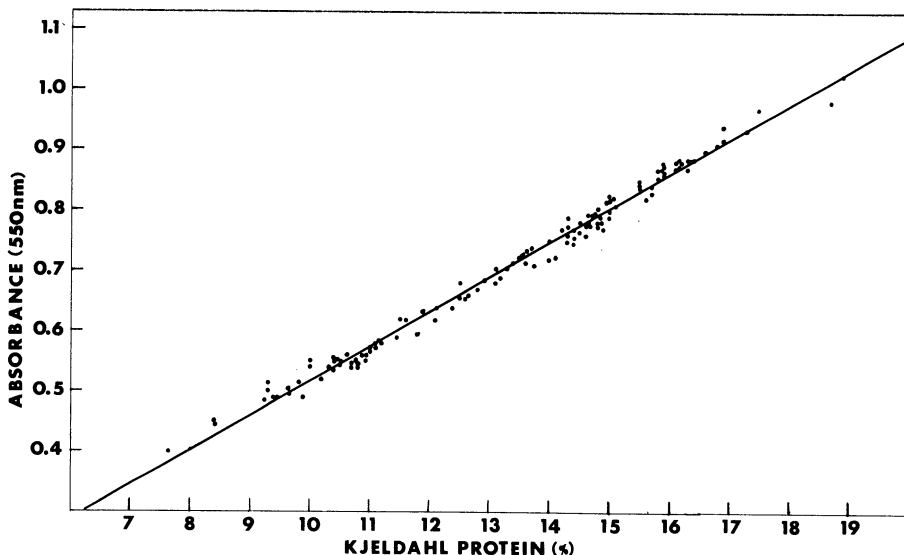


Fig. 1. Relationship of biuret absorbance values for wheat with Kjeldahl protein.