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# A Photoelectric Colorimeter: Its Application in the Measurement of the Concentration of Colored Substances in Solution

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## A PHOTOELECTRIC COLORIMETER; ITS APPLICA-TION IN THE MEASUREMENT OF THE CONCENTRATION OF COLORED SUBSTANCES IN SOLUTION

#### F. M. TURRELL AND LOUIS WALDBAUER

The concentration of plant substances such as the chlorophylls and carotenes can most readily be quantitatively measured by colorimetric methods. Colorimetry also offers a rapid method for the quantitative determination of organic substances such as sugars, oxidases, and inorganic ions as magnesium, iron and phosphates, in which colors can be developed when in combination with certain other ions in solution. The personal equation in such measurements has long been known to contribute a source of error where such methods have been used. An extended bibliography of which part 2 of this paper is composed, has several references which show that the use of photoelectric cells in this regard improves the accuracy



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of colorimetric investigations. A simple photoelectric colorimeter is here described and experimental data presented in support of certain specifications of construction and operation.

The colorimeter, (figure 1) of design similar to that reported by Oltman,<sup>1</sup> consisted of a 32 candle power automobile light bulb mounted in a large flash light reflector attached to the one end of a base board. At the other end of the board a Weston photronic cell was mounted in a U. X. radio socket. Midway between the lamp and the photocell a partition with a circular opening one and a half inches in diameter was so mounted as to permit the light from the auto lamp to shine on the photoelectric cell. Brass mountings were placed on either side of the partition to hold a two inch square Wratten filter and the absorption cell. A bakelite panel was attached to the base board and served as a mounting for the rheostat controlling the light source and the binding posts for the current source and the microammeter connections. The size of this unit was such that it could be easily slipped into a dissecting microscope box to eliminate external light. Electrical connections are shown in figure 2.



#### CURRENT SOURCES

Experimentation with various current sources was necessary in order to obtain a satisfactory current supply. University 110 volt alternating current with a 500 watt light bulb gave large fluctuations in light intensity. University 110 volt direct current showed the same characteristics. When transformers were employed to step down the voltage for use with the 6 volt automobile lamp the percentage variation was increased. Attempts were made to regulate the input line voltage fluctuation by the insertion of a Clarostat radio line voltage regulator. Curves 3 and 2, figure 3, show respectively the fluctuation of the light intensity of Uni-

1 Oltman, R. A. A new method and instrument for quantitative determination of chlorophyll. Plant Physiology 8:321-326, 1933.

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versity alternating current and commercial domestic alternating current as measured with a photoelectric cell and microammeter.



Two 6 volt storage batteries connected in parallel were found to constitute the most satisfactory current source. Curve 1, figure 3, shows a large initial fall in light intensity during the first two minutes of operation of the photocolorimeter with the battery source. The curve is, however, smooth and indicates that over a period of time in which a determination might be made that the light intensity may be approximately constant after the first two minutes. Over a longer period of time in which it might be desired to make many measurements, the light intensity could be regulated with a rheostat. For this purpose it was found that a drum wound rheostat in which the rotating contact touched every part of the resistance as in a slide wire resistance was far superior to the ordinary type of rheostat. Adjustment of the light intensity after the first two minutes of operation could be made so that initial settings before insertion of the absorption cell were made with an error of less than .5 of a microampere.

## MONOCHROMATIC LIGHT

The principle of the colorimeter is based primarily on Beer's law. That is

$$I_d = Ie^{-kcd}$$

where I = penetrating radiation of monochromatic light,  $I_d$  = radia-Published by UNI ScholarWorks, 1935

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tion transmitted by layer of depth d, k = absorption coefficient dependent on  $\gamma$  and the nature of the medium, c == the molecular concentration. The approximate absorption and transmission bands for Wratten filters have been published <sup>2</sup> and a stable filter could be selected in each case with a transmission band within the region of the strongest absorption band of the substance to be measured.

#### EXPERIMENTAL

Chemically pure crystals of KMnO<sub>4</sub> and NiSO<sub>4</sub>  $\cdot$  6H<sub>2</sub>O were used in making up standard solutions. American Chlorophyll Company 5X chlorophyll crystals, the purest grade obtainable, were used in making up the chlorophyll solution. Aliquots were diluted and then measured in the photocolorimeter. The table below gives the concentration and the respective microammeter readings for the various solutions.

Table I -- Concentration of the colored salts in solution with the corresponding photocolorimeter readings in microamperes

Titre KMnO	Reading KMnO	Titre NiSO + 6H O	Reading NiSO • 6H O	Titre	Reading
4	4	4 01120	4 01120	cinoropii).i	( ) )
	1			(a+b)	(a+b)
.0669	18	.2953	145	.002102	51
.03345	20	.1582	245	.000238	95
.02223	24	.0722	332	.000103	145
.01675	24	.0684	345	.0000606	190
.01341	24	.0591	365	.0000562	197
.01113	25	.0492	371	.0000392	230
.00957	30	.0385	384	.0000290	256
.00607	39	.0269	404	.0000186	280
.00134	95	.0143	423	.0000145	295
.00067	142	.0083	430	.0000105	306
.00033	190	.0049	444	.0000000	355
.00000	315	.0016	449		

#### SUMMARY

A simple photoelectric colorimeter is described and experimental data are given which indicate the suitability of certain types of electrical current for operating the photocolorimeter. Data are presented for  $KMnO_4$ ,  $NiSO_4 \cdot 6H_2O$  and chlorophyll (a+b) solutions.

A bibliography on photocolorimeters is given in part II of this paper.

2 Wratten light filters, Eastman Kodak Company, Rochester, New York, 1934.

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