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Thesis submitted by

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section of the Master of Agricultural Science Degree.

AN INVESTIGATION OF METHODS FOR
THE ESTIMATION OF MEDULLATION IN
WOOL SAMPLES.

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AN INVESTIGATION OF METHODS FOR
THE ESTIMATION OF MEDULLATION IN
WOOL SAMPLES.

INTRODUCTION.

In studies of medullation in New Zealand wools the need for an accurate and rapid technique capable of giving a numerical index for the medullation of a sample of wool early became apparent. The matter was complicated by the fact that medullation can only be detected by hand and eye when it exceeds a certain coarseness, and to meet this difficulty the Benzol Test for the detection of hairiness was developed by Elphick. (1932)

The quantitative application of the test has proved difficult; in discussing the evaluation of the medullation revealed Elphick has pointed out that there are three factors which must be considered.

- (1) The average diameter of medulla.
- (2) The percentage of fibres medullated.
- (3) The average distance down the fibres which medullation extends.

In order to arrive at an empirical index he estimated by eye the average percentage of fibres medullated over the staple and weighted the result arbitrarily according to the type of medulla.

After using the method in work on fleece mapping, Elphick came to the conclusion that while it was the best means available at the time for classifying the very large number of samples under examination it could not be regarded as satisfactory owing to the personal element involved, especially since the classification of medulla diameter into three groups masked the comparatively accurate estimation which could be made of the other two factors.

Since the method used was purely empirical it was proposed to relate the figures obtained to the percentage volume of medulla determined by some absolute means. Preliminary investigations on the determination of the Specific Gravity of the wool samples had been commenced when unforeseen circumstances compelled Elphick to discontinue the work.

SOME OBSERVATIONS ON THE ACCURACY OF
ELPHICK'S METHOD.

To obtain some idea of the nature of the problems involved about 180 samples from stud Romney ram hoggets were examined using the above method. Repeats on the same samples even after quite short intervals shewed that considerably more experience than that gained by testing a few hundred staples would be necessary before any degree of reliability could be achieved.

In order to obtain an indication as to the effect of the personal factor, four persons experienced in wool research were invited to estimate by eye the percentage of hairy fibres in staples laid out in benzol. Table I shews the results.

TABLE I. ESTIMATION OF PERCENTAGE MEDULLATION BY DIFFERENT PERSONS

<u>Person</u>	<u>Sample Numbers</u>			
	<u>1</u>	<u>2</u>	<u>3</u>	<u>4</u>
A	65	30	25	15
B	75	40	20	15
C	50	45	25	30
D	50	30	20	10

The samples were not disturbed between the estimations and there is reason to believe that the variation would have been greater if similar fresh samples had been taken and teased out separately for each person.

A staple evaluated by Elphick as 22/300, Plate I, was

mounted in kerosene in a tray and covered with a glass plate ruled with parallel lines. With the aid of a travelling binocular, the number of medullated and pure fibres cutting each line was counted, taking no account of the degree of medullation, at every 1/4 inch from the tip. Table II compares the results obtained with Elphick's estimates. It would appear that even with an experienced observer the percentages estimated do not necessarily coincide with the actual figures. From the general appearance of the staple I would suggest that the visual estimate of percentage is affected by the coarseness of the medulla to a much greater extent than might be considered probable.

TABLE II. COMPARISON OF ESTIMATED WITH ACTUAL PERCENTAGES.

<u>DISTANCE</u> <u>FROM TIP</u> Inches	<u>MEDULLATED</u>	<u>NON-MEDULLATED</u>	<u>TOTAL</u>	<u>PERCENTAGE</u> <u>MEDULLATED</u>	<u>ELPHICK'S</u> <u>ESTIMATE</u>
0.25	15	50	75	20.0	
0.5	37	132	169	21.9	} 60% Moderate
0.75	82	212	294	27.9	
1.0	150	230	380	39.5	
1.25	221	227	448	49.5	
1.5	257	303	560	46.0	} 10% Slight
1.75	254	393	647	39.4	
2.0	261	456	717	36.4	
2.25	228	517	745	30.6	
2.5	171	601	772	22.0	
<u>Repeats</u>					
1.0	152	213	365	41.6	
1.25	221	231	452	49.0	

I cannot pretend to be able to improve upon Elphick's excellent treatment of the problems of visual estimation of percentage volume of medulla, but would suggest that of the factors affecting the figure obtained the diameter of medulla is the least easy to estimate accurately, while the relation of diameter to volume amplifies any errors. Since Elphick shews that the total range of

medulla volume per unit length varies as much as 256 times, while in practice it is not possible to divide this range into more than four classes, it seems that any attempt at visual estimation of percentage volume of medulla can never give accurate results even assuming accurate estimation of the other factors concerned.

It was suggested that a method of grading based on the estimated percentages of medullated fibres at various levels in the staple might be possible. One hundred and eight samples from six Romney hoggets were graded using a tentative method, but the results were not satisfactory owing to the complex mode of distribution of the medulla over the samples. Until more is known of the genetics of medullation, and the effects of environment upon the expression of the genetic make up of the follicles, the only sound plan is to evaluate medullation by the use of a simple quantitative index.

OTHER INDICES OF THE MEDULLATION INVESTIGATED.

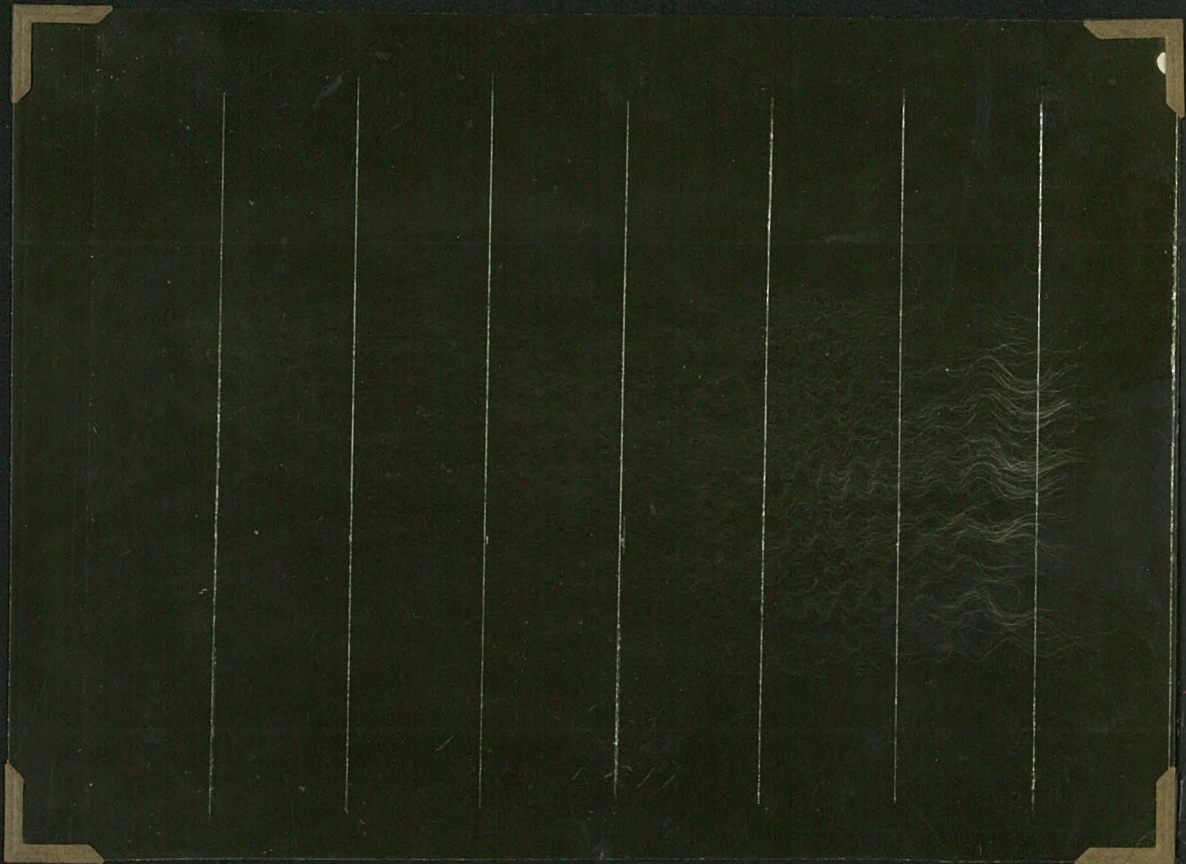
The indices of medullation discussed in the present paper are as follows :-

- Part I. The percentage of the total fibre length in the sample affected by medullation.
- Part II. The amount of light reflected or absorbed by the wool when immersed in benzol.
- Part III. The percentage volume of air space in the sample estimated from accurate determinations of specific gravity.

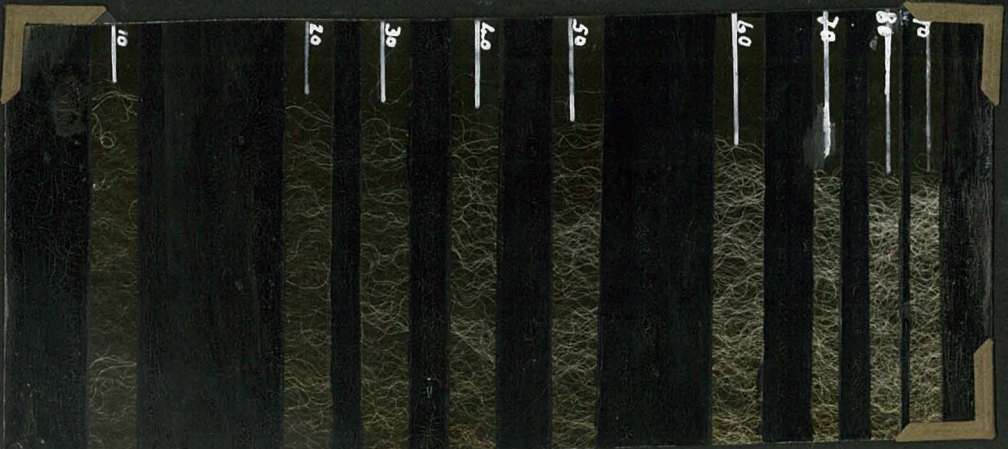
PLATE I

- (a) Sample evaluated by Elphick.
- (b) "Thickness of spreading" standard
 (300 fibres /1").
- (c) "Percentage" standard.

(Plate 1a was taken by Mr. M. T. Gabriel, the remainder of the photographs reproduced in this thesis are the work of the author).



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PART I. THE PERCENTAGE OF THE
TOTAL FIBRE LENGTH IN
THE SAMPLE AFFECTED
BY MEDULLATION.

While not of such a fundamental nature as the percentage volume of medulla, this index offers a sound basis for estimation and selection.

It has already been shewn that unaided visual estimates of percentage medullated fibres for a given region in a staple are apt to be inaccurate and variable from person to person; attempts were made to devise a technique whereby accuracy and standardization could be obtained.

Since Elphick's unpublished experiments on infilling have shewn that there is no certain way of preserving an actual staple shewing a given degree of hairiness indefinitely, one has to fall back on photographs of such standard locks. It had been suggested that it might be possible to produce a series of standard photographs of locks shewing zones over which the percentage of medullated fibres was fairly constant. Several locks were selected and after photographing were counted in kerosene at every 1/4 inch from the tip. (See Appendix A). Very marked differences in the appearance of similar percentages were found to be associated with differences in the thickness of spreading the sample, making a standardization of the latter essential, and the question arose as to the accuracy with which a given degree of spreading could be reproduced. Attempts were made to match the thickness of spreading of a counted staple mounted on black velvet under a glass plate. Scoured samples were teased out roughly with fingers, placed on black velvet and pressed flat with a suitable sheet of glass, the appearance being compared with that of the standard. Small changes in thickness could be made by working the staple with one edge, or a corner, of the glass. In making comparisons it was found that the average size of the spaces be-

between the fibres was the best feature on which to base one's ^{power} judgment of similar thicknesses of spreading. A medium hand lens was found helpful.

To test the accuracy of the above method and also to get an indication as to the magnitude of the personal factor, two members of the College Wool Research staff kindly consented to tease out samples to be checked up by microscopic counts. Table III shews the results of these tests. It would appear that while a given thickness of spreading is reproducible with fair accuracy, there is a tendency to concentrate attention on the centre of the staple.

TABLE III. ACCURACY OF TEASING TECHNIQUE.

Person	Lock Number	Distance from Tips.	Fibres per inch.	
			Teased sample	Standard
Writer	1	1 $\frac{3}{4}$ "	232	296
		2 $\frac{3}{4}$	240	282
		3	258	265
		4	278	288
		4 $\frac{1}{4}$	267	288
		4 $\frac{1}{2}$	261	279
	2	2"	290	300
		3	288	300
		4	290	300
"A"	3	1 $\frac{3}{4}$ "	224	296
		2 $\frac{3}{4}$	246	282
		3	250	265
		4	281	288
		4 $\frac{1}{4}$	293	288
		4 $\frac{1}{2}$	295	279
"B"	4	3"	218	265
		4	264	288
		4 $\frac{1}{4}$	274	288
		4 $\frac{1}{2}$	265	279

A thickness of 300 fibres to the inch was considered suitable as a basis for tentative standardization and was adopted in all subsequent work.

The extremely variable nature of the percentage gradient in various samples examined made it obvious that it would be difficult to produce standards with zones showing definite percentages of medullated fibres and attempts were made to devise a means whereby this percentage could be estimated, by comparison with standard photographs, at a number of regions, the results to be averaged to give the mean percentage over the staple.

A comparator whereby a slightly enlarged image of a portion of the sample teased to standard thickness and immersed in benzol could be viewed in the same microscopic field as the photograph did not give satisfactory results either when the images were made to fill apposing halves of the eyepiece field or when one image was colored with a red filter. The apparatus, however, was not particularly suited to the particular type of comparison.

It was suggested that greater accuracy and ease of comparison would result if the attention of the observer were concentrated on a narrow band running across the staple rather than upon the whole sample. A glass cover plate was ground with coarse carborundum leaving $1/4$ inch intervals of clear glass at every inch. Water soluble pigment was rubbed into the ground areas, the intention being to shew the wool clearly at the $1/4$ inch window intervals, the remainder of the plate being sufficiently opaque to obscure direct vision, while at the same time allowing observation of the general distribution of medullation, e.g., the occurrence of zoning.

Similar $1/4$ inch bands were marked out on the standard and a number of samples were examined by various persons using the apparatus but it became apparent that judgment was made on general appearance, involving coarseness of medulla, rather than on percentage of fibres medullated.

The most satisfactory comparison was found to be obtain-

ed by using a grid consisting of lines etched on the cover glass and filled with water soluble pigment. This was placed upon the teased sample and the average distance between hairy fibres crossing the various lines on the plate compared with the average distance between hairy fibres on the standard.

Using this method of estimation repeats on the same samples after re-teasing gave concordant results which were found to compare favourably with the actual percentages of medullated fibres obtained by counting under the microscope. (Table IV).

TABLE IV. COMPARISON OF ESTIMATED PERCENTAGE OF
MEDULLATED FIBRES WITH ACTUAL PERCENTAGE.

<u>Level</u>	<u>LOCK A.</u>		<u>LOCK B.</u>	
	<u>Estimated %</u>	<u>Actual %</u>	<u>Estimated %</u>	<u>Actual %</u>
1	40	50	40	33
2	40	35	60	57
3	40	40	80	76
4	35	34	90	84
5	40	36	90	82
6	40	39	60	69
7	40	40	30	41
8	30	35	20	21
9	30	35	10	12
10	25	26		
11	20	18		
12	10	11		

In preliminary work estimations were made at 1/4 inch intervals but in later work it was considered to be sufficient to divide the staple length into seven equal portions, using the 1/4 inch grid, and to take the average of the seven estimations as being representative.

It would appear that a method similar to the above would be capable of expressing, with reasonable accuracy, the percentage of

the total fibre length in the staple affected by medullation, although it cannot be regarded as established without further checking against microscopic counts for samples involving different types and degrees of medullation, different counts, and in particular a study of the number of estimations required on each staple in order to get sufficient accuracy.

Although it has been used for routine work the technique is unsatisfactory for such purposes owing to the length of time required to complete an estimation. It seems reasonable to expect that experience in its use would suggest "short cuts" such as the device of matching the roughly teased sample with standards round about the 300 fibre mark, and using an appropriate percentage standard, rather than working to one set figure.

It seemed desirable to exhaust the possibilities of other methods of measuring medullation before proceeding with the elaboration of the technique above described, since in all scientific work it is general experience that while visual evaluations by comparison with standards is often both simple and accurate, these advantages are offset by the length of time involved.

PART II. THE AMOUNT OF LIGHT
REFLECTED OR ABSORBED
BY WOOL WHEN IMMERSSED
IN BENZOL.

It was suggested that examination of wool samples in monochromatic light might indicate further lines of investigation. Raw and scoured wool, as well as a sample in benzol, was examined using various lighting. The only unusual appearance was found in the case of illumination from a quartz mercury vapor source, under which wool heavy in condition gave an orange fluorescence. It is possible that if the samples were dyed before viewing under the quartz lamp differences might be observed owing to the varying effects of dyes on medullated and non-medullated wool, and the fact that some dyes fluoresce in ultraviolet light.

Since hair reflects considerably more light than pure wool when under the benzol test, it would seem that the amount of light reflected might be expected to serve as an index of the degree of medullation. At first sight it would appear probable that the amount of light reflected would be a function of the surface area of the medulla in the sample, in which case a linear relation between light reflected and percentage volume of medulla would not obtain. A given amount of fine medulla would have a greater surface area, and hence reflect more light, than the same amount of coarse medulla. There are such a large number of factors which might affect the amount of light reflected, however, that one cannot establish a relation on a priori grounds. The effect of the internal structure of the medulla upon its apparent whiteness, the distorting effect on apparent diameter due to the cortex of the fibre acting as a lens, reflection from the ends of small sections when the medulla is discontinuous, and probably many other factors, would have to be considered.

Attempts were made to measure the light reflected from a moderately hairy sample in benzol, when placed in the concentrated

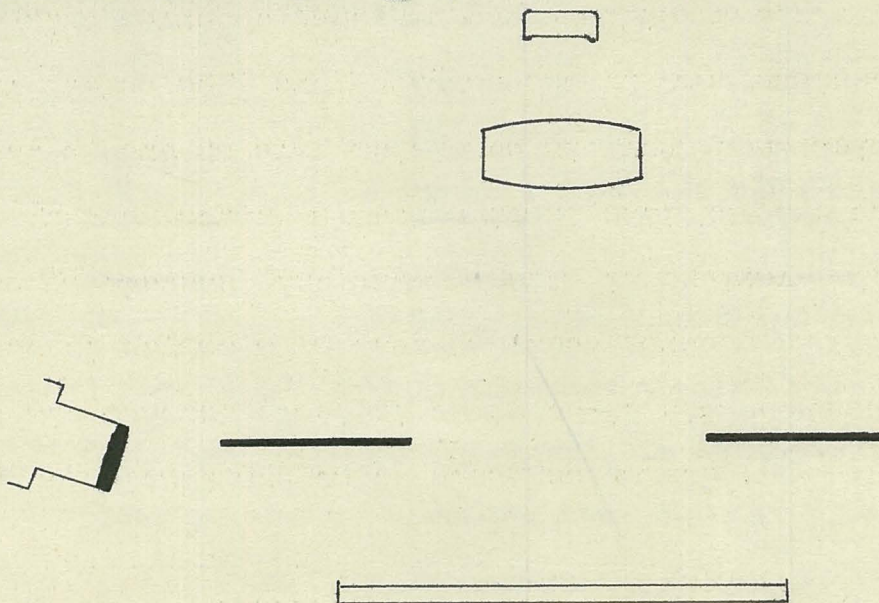
beam from a projection lantern, with a Lumner Brodhun cube but the illumination intensity was too low for the instrument to be successfully used.

PRELIMINARY EXPERIMENTS UTILIZING A PHOTOELECTRIC CELL.

The possibility of utilizing a photo cell for the purpose of measuring the amount of light reflected from samples of wool in the benzol test under standard conditions had been projected for some time. A survey was made of the literature on the subject, (Appendix B), and indicated that a photo cell of the rectifier type would be the most suitable for present purposes.

By courtesy of Standard Telephones and Cables (A'sia) Ltd., Wellington, a Weston Photronic cell was made available for preliminary experiments.

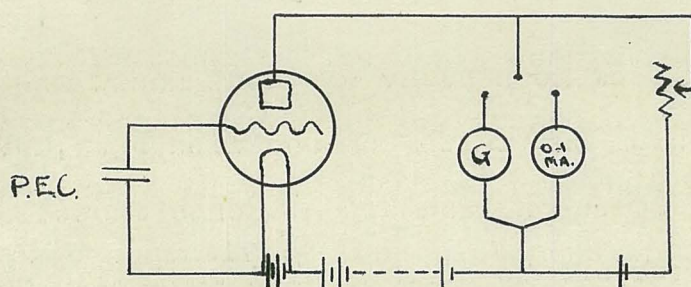
The optical apparatus was the same as that used with the Lumner Brodhun cube, an image of the brightly illuminated benzol test being focussed on the sensitive surface of the cell, which replaced the diffusing screen of the photometer, by means of a large aperture condenser. (See Figure I.).



The E. M. F. produced by the cell was measured using a

Tinsley potentiometer sensitive to 0.02 milli volt (m.v.) and was found to be of the order of 3 m.v. for a moderately hairy staple. A number of readings were taken using this apparatus but it was found that standardization of the potentiometer was necessary every few minutes - presumably owing to changing E.M.F., of the accumulator used.

Since the low internal resistance of the photo-cell (7,000 ohms) obviates the use of electrostatic methods of measurements a vacuum tube voltmeter used with a reflecting galvanometer, as indicating instrument, was decided upon as the most convenient means of measuring the cell E.M.F. The usual circuit (Figure 2) was fitted up and various types of tube tried.



A Phillips A.415 valve was found to give the most stable reading on the galvanometer.

Deflections were obtained by placing a black screen between the lens and the photo-cell and recording the "zero" deflection, the screen then being removed and the deflection again recorded. The apparatus was so arranged that the difference between these two readings was almost entirely due to light from the wool under test.

Table V shews some of the results obtained.

TABLE V.

DEFLECTIONS OBTAINED WITH BADLY HAIRY

TIPPED SAMPLE IN THE TRAY.

<u>Galvanometer Readings</u>		<u>Deflection</u>
<u>Zero</u>	<u>Screen Removed</u>	
Wool + Tray		
16.7	20.7	4.0
17.2	21.2	4.0
17.2	21.2	4.0
17.6	21.5	3.9
Wool in tray condensed into a much smaller area		
2.1	6.5	4.4
2.6	6.6	4.0
2.8	6.8	4.0
2.9	6.9	4.0
Wool Removed from Tray - Tray Alone		
4.2	5.3	1.1
4.5	5.6	1.1
4.5	5.6	1.1
4.8	5.8	1.0

To obtain an indication of the range of values, a series of graded samples was examined, the results being shewn in Table VI, deflections per unit weight of wool being calculated as a basis of comparison.

TABLE VI. PRELIMINARY DETERMINATIONS OF PHOTO ELECTRIC

INDEX OF HAIRINESS.

Sample	Weight	Deflection	Deflection Due to Wool	Deflection per gram
Tray only		1.1		
A Almost Pure	0.29 gr.	1.7	0.6	2.1
B Hairy Tip	0.33	3.0	1.9	5.7
C do do	0.25	3.0	1.9	7.6
Same Sample Condensed	0.25	3.0	1.9	7.6
D General Medullation	0.29	5.5	4.4	15.1
E do do	0.44	8.0	6.9	15.7
F Bad Hairy Tip	0.31	9.3	8.2	27.0
G Hairy Sample	0.35	11.4	10.3	29.5

The photo electric indices are not accurate owing to the fact that the small central bakelite area on the cell used was insensitive to light, hence any portion of the hairiness image falling thereon would not be recorded¹; moreover the illumination was not even. The results indicate, however, that stable readings may be obtained over a wide range of values.

Table VII shews the results of repeat determinations on some of the above staples after 24 hours, the samples being placed as nearly as possible in their original positions in the tray, - reasonable agreement is indicated.

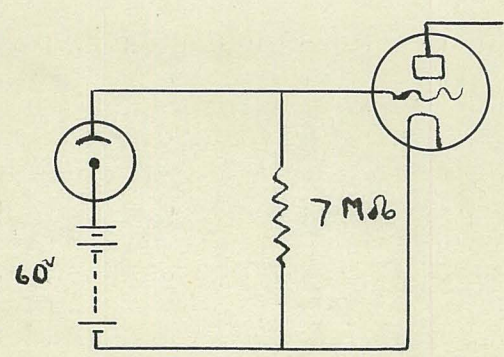
¹ Standard Telephones and Cables advised that cells without the central bakelite area were being manufactured and were available from the U.S.A., to special order.

TABLE VII. COMPARISON OF ORIGINAL AND REPEAT DEFLECTIONS
FROM STAPLES AFTER 24 HOURS.

<u>SAMPLE</u>	<u>ORIGINAL DEFLECTIONS</u>	<u>REPEAT</u>
A	1.7	1.7
E	8.0	8.2
F	9.3	8.8
G	11.4	11.3

Experiments with a Vacuum Photo-Cell.

A photo-cell - probably of the vacuum type - kindly loaned by the Dominion Observatory was fitted up in the circuit shewn in Figure 3, and used with the same optical arrangement as before. Stable readings could not be obtained with the temporary electrical apparatus used owing to variable leakage effects, but the sensitivity did not appear to be so great as with the Weston cell.



CONCLUSIONS.

It appeared that a Weston Photronic cell of the type without the central bakelite area, used in conjunction with a well designed vacuum tube voltmeter, would be capable of measuring the light reflected from wool under the benzol test with considerable accuracy - 2 to 3%, provided that a constant source of illumination capable of producing an even brightness over the tray could be obtained, so that a given amount of medulla would

give a constant deflection without reference to its position in the tray.