

red, yellow, green and blue; (b) that they must be regarded as operating in two bipolar units, red with green, and yellow with blue; (c) that brightness is to some extent dependent on the modulator units.

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¹ Granit, R., "Sensory Mechanisms of the Retina", Section IV (1947).

² Pickford, R. W., *Nature*, 157, 700 (1946).

A Simple Technique for the Differential Staining of Lignified Cell Walls

THE common methods for differential staining of lignified plant tissue are specific when used correctly, but minor errors in procedure may lead to unsatisfactory results. The following simple technique has been found a reliable, cheap and quick supplement to the other methods available, and to give good results in unskilled hands.

It is based on the observation that, in all cases which we have examined, a colourless solution of benzidine imparts an orange or yellow colour to lignified cell walls, but causes no colour change in cellulose or in cutinized or suberized walls. The most intense orange coloration is produced by a saturated solution of benzidine in glacial acetic acid. With lowered acidity the solubility of benzidine decreases, the resulting solutions producing a less brilliant but equally specific staining. This develops in sections after thirty seconds to sixty seconds treatment; but as the solution is colourless and the reaction occurs only in lignified tissue, sections can be left in it for many days without 'overstaining', and there is no need for subsequent washing or 'destaining'. This is an advantage when handling sections in bulk, it being possible to transfer them to the solution directly after cutting and to hold them in it until it is convenient to mount them with or without a light counter stain. A less acid solution may also be used for the vital staining of tissue, as vascular systems of leaves, etc., are clearly visible after impregnation. The colour is unaffected by treatment with alcohol or xylol, and has shown no signs of fading in sections exposed to daylight for three months.

In view of the constitution of benzidine, the chemistry of the staining is possibly similar to that of the colour reaction given by lignified cell walls with a number of primary and secondary amines.

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Influence of Time and Temperature of Storage on Dye-Reduction Tests in Milk

In applying overseas data to the determination of suitable dye-reduction standards for the grading of milk in New South Wales, it soon became apparent that particular attention would have to be paid to the influence of time and temperature of storage on the reduction time¹. Apart from Smythe's work in

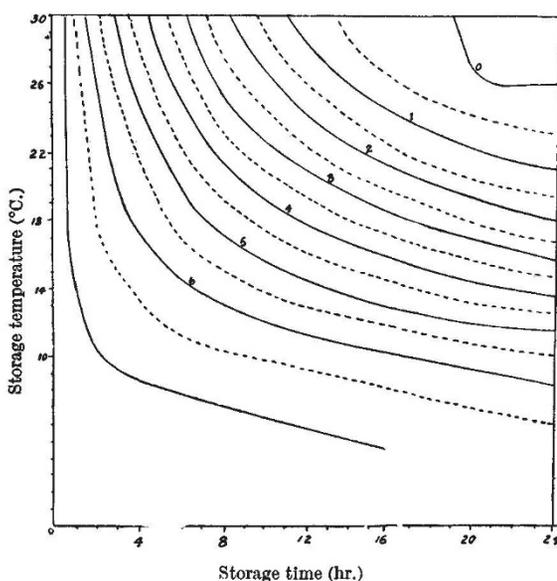
Queensland², for a relatively limited storage time, there is surprisingly little information concerning the significance of these factors, and it is thought that our own approach and results will prove of interest beyond Australia.

The primary aim of our work has been to study the trends in decrease of dye-reduction times with storage time and temperature, including within the range of our experiments conditions likely to be encountered both in Australia and Britain. By this means it has been possible to bring to bear on our own problem the large body of British experimental work and practical experience, as well as to arrive at a temperature compensation scale for local variations that are likely to be more extreme than those encountered in the United Kingdom. Samples from six producers, representing extremes of production conditions, were taken at the completion of milking and stored at combinations of time and temperature, namely, 4, 8, 12, 16, 20 and 24 hr. and 0, 10, 14, 18, 22, 26 and 30°C. Methylene blue and resazurin tests (both at 37.5°C.) were then performed on each sample.

With the advice of Dr. D. B. Duncan, of this Department, it has been possible to use the results so obtained to study the trends with storage time and temperature as a regression surface, having twelve parameters and allowing for quadratic variation with storage time and cubic variation with temperature as well as interaction between the two. The results for the modified methylene blue test are shown in the form of a contour graph (reproduced herewith) in which the lines join the points of equal reduction times. From this graph it is possible to arrive at an approximation of reduction time associated with any combination of storage time and temperature; but more fully the surface is represented by the equation:

$$Y = 7.2 - 0.005t + 0.00027t^2 - 0.047T + 0.019tT - 0.00044t^2T + 0.0047T^2 - 0.00293tT^2 + 0.000039t^2T^2 - 0.00010T^3 + 0.000048tT^3 - 0.00000005t^2T^3;$$

Y is reduction time in hours, t is storage time in hours, T is storage temperature in °C.



Influence of time and temperature of storage on methylene blue reduction time. Lines join points of equal reduction time in hour - half-hour intervals