



Timber Talks



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CONIFER TISSUE ANALYSES

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Physiological studies concerned with plant reproduction require an understanding of seasonal changes in carbohydrate food reserves in buds, foliage, twig bark and wood associated with the development of reproductive primordia. The carbohydrates include such soluble sugars as fructose, glucose, sucrose, raffinose, stachyose and xylose. Variation in chemical properties of these sugars makes analysis difficult. Large numbers of samples are required, some of the plant tissues are small, and a rapid semi-micro method of analysis is necessary; additional complexity occurs when tissues from conifers are being analyzed due to their particular physical characteristics, and to erroneously high apparent sugar levels obtained by using sugar reducing methods. Improvement in sample preparation and in the analysis of coniferous tissues for variations in soluble sugars was investigated.

To minimize stimulation of enzymes during oven-drying of tissues, and possible heat loss of carbohydrates, samples were dissected in the field and transported to the laboratory in a dry-ice refrigerated chest. Tissues were freeze-dried, ground in a Wiley mill, bottled and re-dried before analysis. Small samples were weighed into glass fibre filter paper thimbles and soluble sugars separated from insoluble materials by 4 hours of continuous extraction with 80 per cent ethanol. Residues were dried and maintained until required for analysis.

To analyze for total soluble sugars, the ethanol was evaporated and the extract maintained in an aqueous solution. Proteins and other substances precipitated from the extract by saturated neutral lead acetate solution caused negligible interference with colorimetric procedures. Removal of pigments and phenolic interfering substances, by treatment with powdered bone charcoal, was essential for valid and reproducible results.

A stabilized diluted anthrone solution was prepared by dissolving 1.5g anthrone in one litre of a mixture of 20% ethanol and sulphuric acid. This reagent produced greater color intensity than other commonly used anthrone reagents and retained its stability for 2-3 weeks. Concentrations of total sugar in sample extracts was determined by color produced on reaction with anthrone for exactly 7.5 minutes in a boiling water bath, in comparison with that of a series of standard glucose solutions. Greater similarity of values were obtained for the principal free sugars than when other reagents or methods were used.