Figure 2. Changes in leucine aminopeptidase activity during growth of sugarcane callus under shoot forming (o-----o) and non-shoot forming (o--o) conditions. Activity is expressed as μ moles of β-naphthylamine liberated per min per mg protein.

The increase in activity of LAP after day 8 (the day of visual manifestation of shoots) in SF tissue suggest that LAP has some major role in later stages of differentiation. Higher levels of LAP activity during initial stages have been reported in bush bean cotyledon suspension cultures grown in presence of 2,4-D and kinetin as compared to the corresponding cultures grown without growth regulators. They have also reported higher LAP activity, in absence of growth regulator, only during later stages of growth. A sharp peak in activity in NSF tissue on days 2 and 6 and in SF tissue on day 10 may be attributed to the appearance of some new isoenzyme forms growth, as suggested for LAP activity profile in bush bean cotyledon suspension cultures. Efforts in this direction are in progress.

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SYNTHESIS OF 4-HYDROXYCOUMARINS

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Some 4-hydroxycoumarins have been prepared by the method of Shah et al. The coumarins have been characterised by preparing the corresponding, bromo, nitro, sulphonic acid and phenylazo derivatives.

\[
\begin{align*}
I & : R_1 = R_3 = CH_3, \quad R_2 = R_4 = H \\
II & : R_1 = R_2 = H, \quad R_3 = R_4 = CH_3 \\
III & : R_1 = R_4 = H, \quad R_2 = Cl, \quad R_3 = CH_3 \\
IV & : R_1 = R_2 = R_3 = H, \quad R_4 = Ph \\
\end{align*}
\]

Various phenols were condensed with malonic acid in presence of phosphoryl chloride and fused zinc chloride at 70°C for 10–12 hr when the corresponding 4-hydroxycoumarins were obtained in 60–70% yield. The period of reaction had to be varied according to the nature of the phenol.

Thus, 3,5-xylenol gave 4-hydroxy-5,7-dimethylcoumarin [I, m.p. 210-11°, UV λ235,λ220 (log ε) 239

max
INFRA-RED IDENTIFICATION OF Fe–S BOND IN ASSAM COAL

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The bondings of organic sulphur in high sulphur Assam coal has been known as C–S linkage. Thiophenol, disulphide and thiophene have been reported as components of organic sulphur. The nature of sulphur bondings are very important from the viewpoints of desulphurization and liquefaction and also in understanding the constitution of high sulphur coal; this study has therefore been carried out. As x-ray photoelectron spectroscopy normally does not provide structural information of organic sulphur in Assam coal, IR technique was used to examine such structural components.

As regards minerals occurring in coal, Estep et al. have identified the IR absorption bands of pyrite and marcasite as:

pyrite – 284(w), 340(m), 391(vw), 406(s) cm\(^{-1}\)
marcasite – 285(w), 321(m), 350(m), 396(s), 407(s), 422(vw) cm\(^{-1}\).

Both these minerals exhibit two common bands (nearby 400 cm\(^{-1}\)) and the only difference between them is that pyrite shows an unique band at 340 cm\(^{-1}\).

The absorption band in the region 600–200 cm\(^{-1}\) in the spectrum of Assam coal has not been evaluated earlier. Therefore a high sulphur coal sample (c = 80.7 %) containing 0.6 % pyrite and 2.0 % organic sulphur was taken and treated with warm dilute (2N) nitric acid to remove sulphate and pyrite sulphur. The acid-washed coal sample was then subjected to x-ray investigation to determine whether any pyrite is present. The IR spectrum of this sample was recorded in KBr pellet using a Perkin-Elmer model 521 spectrophotometer in the range 4000–200 cm\(^{-1}\).

Careful investigation of IR spectrum reveals two broad absorption bands (at 340 and 540 cm\(^{-1}\)) in the region 600–200 cm\(^{-1}\). Absence of 400 cm\(^{-1}\) band in the spectrum indicates the absence of pyrite and marcasite in the sample.

The major part of the organic sulphur in Assam coal has been reported to contain SH groups. During sample preparation, no changes occurred in the SH group as nitric acid would not attack the organic sulphur. No absorption band is observed for S–H stretching vibration which otherwise occurs in the

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3. a. Sonn, A., Ber., 1917, 50, 1292.
4. c. Da Re, P. and Sandri, E., Ber., 1960, 93, 1085.