

03/04/2015

Seat No 14

## Biochemistry Practicals (Paper - 08)

### C<sub>2</sub> - Test.

Seat No: 14

#### \* Extraction of RNA from Spinach leaves.

##### → Extraction.

The spinach leaves are homogenised in lysis buffer. 1ml of 2M sodium acetate is added and shaken well. To this, 2ml of mixture of chloroform and isoamyl alcohol in the ratio of 49:1 is added and shaken vigorously for 5 mins. Keep the mixture for centrifugation at 10,000 rpm at 4°C

The aqueous layer is transferred in to a fresh tube and equal volume of ice cold isopropanol is added. The mixture is centrifuged kept in -20°C for 1 hour to precipitate RNA. It is then centrifuged at 10000 rpm at -20°C for 20 mins. The supernatant is discarded.

Equal volume of ice cold proponal is added to the pellet and kept at -20°C for 1 hr. It is then centrifuged at 10000 rpm at 4°C for 10 mins to get RNA out. The supernatent is discarded and pellet is washed with 75% alcohol. Centrifuged and vacuum dried until the alcohol is completely evaporated.

The pellet is stored at  $-20^{\circ}\text{C}$ . The RNA extracted can be purified, weighed & detected by orcinol method.

Result : The presence of RNA is detected by orcinol method.