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## ***Histology of Organogenesis in Sesamum indicum L.***

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### **Objectives**

*Sesamum indicum L.* is very recalcitrant for genetic transformation although the establishment of efficient regeneration system by organogenesis. In this study, we investigated the histological changes of sesame during organogenesis to determine the critical time for genetic transformation by *Agrobacterium*.

### **Materials and Methods**

#### **1. Material**

Plant – *Sesamum indicum L.*, cv. Dasak, various calli according to developmental stages.

#### **2. Methods:**

Calli collected at different time points were immediately fixed in 2.5% glutaraldehyde in 0.1M sodium phosphate buffer, pH 7.0. The tissues were dehydrated in ethanol series followed by xylene-wax infiltration and finally embedded in Paraplast Plus (Sigma). Microtome sections (10 $\mu$ m) were mounted in glass, stained with safranin, haematoxylin, and observed under brightfield microscope.

### **Results and Discussion**

We developed an efficient and reproducible protocol for in vitro regeneration system using organogenesis from cotyledonary explants in sesame. Multiple shoot induction could be achieved by supplementing MS basal medium with BAP(5 mg/l), IAA(1 mg/l), ABA(1 mg/l) and AgNO<sub>3</sub>(5 mg/l). A first term-culture(30 days) of explant in high sucrose(9%)-containing medium followed by transfer and maintenance in low sucrose(3%)-containing medium resulted in healthy, multiple plantlets. For the successful genetic transformation by *Agrobacterium* methods, the histology of different developmental stages involved during organogenesis was analyzed using microscopic observation and microtome sectioning at various stages. These observations can be useful for the choice of critical stage of organogenesis for genetic transformation.

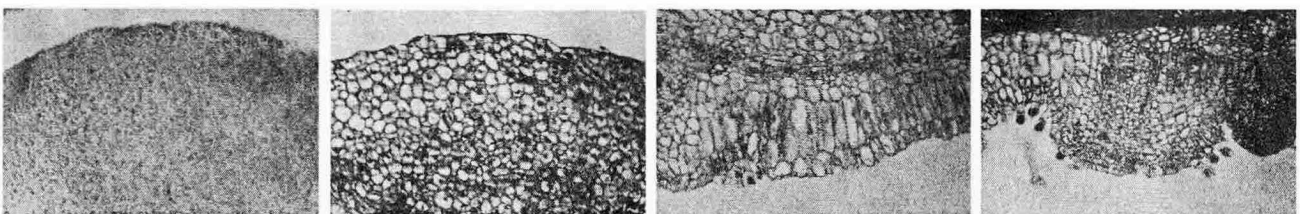


Figure 1. Characteristics of histological changes according to developmental stages.