Plant Regeneration in Chickpea

V.K. Sheila, J.P. Moss, C.L.L. Gowda, and H.A. van Rheenen[1]

Micropropagation is one of the most widely used tissue culture techniques for rapid asexual in vitro propagation. Such techniques for grain legume crops have been rare but are of recent interest. Chickpea *(Cicer arietinum* L.) is a major food legume in many countries and is one of the most important pulse crops in the world. A technique was standardized to propagate chickpea and its wild relatives. A medium was developed to induce multiple shoots from excised shoot tips of 15-day-old aseptically raised seedlings and 30-day-old greenhouse-grown plants. Shoot-tip explants (2-8 cm long) of six chickpea genotypes (L 550, K 850, ICCC 32 (ICCV 6), ICC 12237, ICCC 42, and C 235) and four wild *Cicer* spp (C. *bijugum* No. 201, *C. cuneatum* SL 157, *C. judaicum* No. 185, and *C. pinnatifidum.* No. 188) were cultured under two light regimes. Multiple shoots were produced from shoot tip explants of all the genotypes tested on medium containing 2.0 mg l^{-1} 6-Benzyl Aminopurine (BA) and 0.5 mg l^{-1} Indole Acetic Acid (IAA). All the chickpea genotypes except ICCC 32 and ICC 12237 produced a greater number of shoots under 16 h than 24 h daylength. Explants of *C. cuneatum* SL 157 and *C. judaicum* No. 185 also responded better under 16 h than 24 h daylength. *Cicer bijugum* and *C. pinnatifidum* were not tested under 16 h daylength but did produce shoots under 24 h daylength.

Shoots produced were excised and subcultured individually on fresh medium at 6-week intervals to ensure a high rate of multiplication. One shoot-tip explant of the genotype K 850 produced a total of 134 shoots after three transfers. The shoots were excised and transferred individually to a rooting medium supplemented with Indole-3 Butyric Acid (IBA) or Napthylene Acetic Acid (NAA). The IBA-supplemented medium induced normal roots, while NAA-supplemented medium induced short, thick roots. The plantlets were transferred to polythene bags or small plastic pots containing sterile sand and then drenched with an antimicrobial solution (Benlate® + Thiram® + Agrimycin® mixture) to reduce plant mortality. Finally, the plantlets were established in a sand + Vertisol (3:1) medium in pots and maintained in the greenhouse. Normal pods were produced from such micropropagated plants.

Attempts were also made to regenerate plants from callus. Callus was induced from a range of explants (leaflets, mature cotyledons, immature cotyledons, immature embryos, hypocotyls, epicotyls, shoot tips, and stems) of C. *arietinum* genotypes and wild *Cicer* spp. Callusing response varied with explant, media, light, and temperature. Medium containing 2,4-D, alone, or in combination with other hormones, induced callus with embryogenic structures that resembled embryogenic initials. Further differentiation into embryo-like structures did not occur. However, some of the calli produced roots.

Medium supplemented with BA and IAA induced callus from the cut end of stem explants in contact with the medium. Very few calli produced shoot buds when maintained on the same medium. The callus was transferred to medium without hormones for shoot elongation. The regenerated shoots produced roots on medium supplemented with IBA. This technique will be standardized to improve the frequency of regeneration.

1. International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru P.O., A.P. 502 324, India.

Sheila, **V.K.,** Moss, **J.P.,** Gowda, C.L.L., and van **Rheenen, H**.A. 1992. Plant regeneration in chickpea. Page 153 ***in*** Biotechnology and crop improvement in Asia (Moss, J.P., ed.). Patancheru, A.P. 502 324, India: International Crops Research Institute for the Semi-Arid Tropics.