The present study entitled “In vitro production of Swertiamarin from Swertia chirata using liquid culture” was carried out in plant tissue culture laboratory, Department of Biotechnology, Shoolini University, Bajhol, Solan. The summary of the findings is as under:

- For the micropropagation of Swertia chirata, leaf part from field grown plant was used as an explant. For the sterilization of explant 0.1% Mercuric chloride with 2-4 drops of 0.2% bavistin for 5-10 minutes was found to be the best combinations.

- Aseptic conditions were maintained for the initiation of the callus culture. Different callus was optimized namely hard compact, compact and friable, compact green, soft friable light yellowish and friable yellowish and embryogenic callus was resulted. Callus was obtained when MS media supplemented with:

  1. NAA (1.5mg/l) + 6-BA (3mg/l).
  2. 2,4-D (2mg/l) alone.
  3. 2,4-D (2.5mg/l) + 6-BA (0.7mg/l).
  4. 2,4-D (1mg/l) + 6-BA (0.5mg/l) + TDZ (0.5mg/l).

Among all treatments, the best concentration for callus culture (compact green) was obtained with 94.4% of percentage response when MS medium was supplemented with 2, 4-D (1mg/l) + 6-BA (0.5mg/l) + TDZ (0.5mg/l).

- For the multiplication of callus culture, the most suitable medium was MS + 2, 4-D (1mg/l) + IAA (0.5mg/l) with percentage response of 88.8%.

- In the present study Swertia chirata plants were regenerated from the callus. Once the callus was multiplied and maintained, it was transferred to the regeneration medium. Among all the treatments tried, maximum number of shoots were obtained in MS medium supplemented with Kn (1.5mg/l) + IAA (2mg/l) + GA3 (0.2mg/l) with an average rate of 7.6 shoots per explants. The shoots regenerated in about four weeks of incubation period.

- The effect of Kn, IAA and GA3 at concentrations varying between 0.2 to 2.0 mg/l was studied on multiplication and growth response of shoot initiation. Regenerated Swertia chirata plants were then multiplied on MS basal and on
MS medium supplemented with Kn, IAA and GA₃, either alone or in combinations. There was significant variation in terms of number of shoot buds induced per explant.

- Cell suspension cultures were established from compact green callus and transferred in to 250ml conical flasks containing 60ml of liquid medium. Flasks were tightly closed and placed in an orbital shaker (110 rpm) at 25 ± 2°C for 15 days. Cell viability was checked by using erythrosine dye under light microscope and 90 – 95% cells found viable.

- In this study, the plan was to develop a cell suspension protocol for *Swertia chirata*. Different combinations of growth regulators were used. Out of all treatments, MS + Kn (0.3mg/l) + BAP (0.4mg/l) + NAA (0.3mg/l) + TDZ (0.1mg/l) was found to be the best treatment for cell proliferation with 83.3% of survival rate.

- In liquid aerated culture vessels, different concentrations and combinations of growth regulators were used. The hormonal concentration, MS+ Kn (1mg/l) + BAP (2mg/l) + GA₃ (1.5mg/l) was found to be the best for multiplication of shoots with an average number of 17.5 shoots per explants after 17 days of incubation period.

- The quantification of Swertiamarin, mangiferin and amarogentin was carried out by using the reverse phase High Performance Liquid Chromatography (HPLC Waters 515) and elutions were carried out under isocratic conditions using acetonitrile: water (70: 30) at a flow rate of 1.0 ml/min and equilibrated for 5 min at 240 nm UV wavelength.

- Swertiamarin, mangiferin and amarogentin content in leaves derived callus cultures was assayed in present study. But there was negligible content of these metabolites were found after HPLC analysis So, to enhance the production of Swertiamarin, mangiferin and amarogentin, the effect of elicitor (methyl jasmonate) at different concentrations for different time incubation was tested by using liquid cultue and semi-solid medium.
From the present study it can be concluded that the higher swertiamarin content (4.38%) by using liquid culture with 50µ g methyl jasmonate for 16 days incubation, while in semi-solid medium highest Swertiamarin content (2.52%) was found with 100µ g methyl jasmonate after 16 days incubation.

Antimicrobial activity of *Swertia chirata* was checked against E. coli and S. aureus using agar well diffusion methods and compared with standard antibiotics (Ciprofloxacin). The extract of *Swertia chirata* gives good results against S. aureus (12.3 ± 0.5) on Muller Hinton agar.