Chapter 3

Objectives and plan of work

In view of the importance of pharmacological activity attributed to *B. juncea* and in availability of scientific evidence pertaining to the hepatoprotective effect of *B. juncea* seeds in particular, the present study has been initiated with following objectives:

- Establishment of APAP-induced hepatotoxicity model on HepG2 cell line
- Isolation of phytoconstituents from *B. juncea* seeds and phytochemical of the extract
- Cytotoxicity assessment and hepatoprotective efficacy testing *in vitro*

**PLAN OF WORK**

The work was divided into three parts in line with objectives and presented in the following chapters.

1) **Establishment of APAP-induced hepatotoxicity model on HepG2 cell line**

- Expansion and freezing of HepG2 cell line
- Cytotoxicity assessment of APAP for selecting the dose
- Establishment of hepatotoxicity model based on liver indices
- Microscopic examination of cytotoxicity
- Detection of generation of ROS and cell cycle analysis of the APAP-induced damage
2) Isolation of phytoconstituents from *B. juncea* seeds and phytochemical of the extract

- Preliminary phytochemical and physicochemical characterization of seeds
- Extraction and fractionation of bioactive phytoconstituents from seeds
- *In vitro* antioxidant assessment of the phytoextract
- Fingerprinting of phytoextract using multiple detection system

3) Cytotoxicity assessment and hepatoprotective efficacy testing *in vitro*

- Cytotoxicity assessment of the phytoextract
- Hepatoprotective evaluation of the phytoextract on APAP-induced hepatotoxicity model
- Assessment of hepatoprotective activity of phytoconstituents detected in mustard seed extract in the APAP-induced hepatotoxicity model