Abstract

In the present study, about 140 samples of different indigenous plants were collected from different localities for the isolation of taxol producing endophytic fungi. About 310 endophytic fungal strains were isolated from these samples and out of which maximum strains (95) were isolated from the samples collected from the *Taxus* sp. whereas minimum samples (15) were isolated from the *Citrus limonum*. These strains were screened for the production of the taxol through surface culture in 250 ml Erlenmeyer flasks by growing in PD broth at 30°C for 20 days. It was found that out of 310 isolated fungal strains only 20 strains had the ability to produce the taxol (9 from *Taxus*). It was found that among these 20 strains, only three (UH-10, UH-230 and UH-275) showed reasonable amounts of the taxol production i.e. 140µg/l, 80µg/l and 1540µg/l, respectively. The identification of these strains was done on the basis of the morphological characteristics, structure and size of the spore and the genetic analysis. These strains i.e. UH-10, UH-230 and UH-275were identified as *Cladosporium cladosporioids, Asperigllus niger* and *Eurotium rubrum*, respectively. The surface culture fermentation was found to be the most suitable fermentation type for the production of taxol as compared to SmF and SSF. Ten different fermentation media were screened to enhance the production of taxol by the isolated fungal strains. *Eurotium rubrum* showed maximum taxol productivity among these three selected strains in medium M-3 containing peptone, glucose, sodium acetate and sodium benzoate. The optimization of the physical parameters was done for *Eurotium rubrum* (UH-275). The maximum amount of taxol was found to be produced when fermentation was carried out at 30°C for 21 days with initial pH (6.5). Among the nutritional parameters, glucose (1.5%) as carbon source was found to be the most suitable while among the organic and inorganic nitrogen sources, peptone (2%) and 0.3% sodium nitrate resulted in maximum productivity of taxol. Sodium benzoate at a concentration of 50mg/l, as a constituent of the medium showed better production of taxol. Spore inoculum at a concentration of 5% resulted in the highest amount of taxol production. Physical (UV) and chemical (Nitrous acid and EMS) mutagens were used in order to create random mutagenesis for enhancing the yield of taxol in the selected strain. Any of the variants produced after random mutagenesis did

not show enhanced production of taxol. The fermenter studies showed that the time of incubation for maximum production of taxol was reduced from 21 days to 14 days. Optimum rate of aeration and agitation in the fermenter were found to be 0.5 vvm and 100 rpm, respectively. In the bioreactor, highest amount of taxol production (4.8mg/l) was observed when the pH was maintained at 6.5. The down streaming was done with silica gel column chromatography to get the high purity taxol. The nuclear magnetic resonance (H-NMR) and infra-red (IR) spectrometry was done for the characterization of purified taxol. The spectra obtained confirmed the presence of high purity taxol in the samples. Mesoporous silica nanoparticles, both unfunctionalized and functionalized (aluminum and iron) with pore size greater than 1.8nm were synthesized and used for the drug delivery studies. Three different types of aluminum and two iron functionalized particles were used. Characterization of these mesoporous silica nanoparticles was done with scanning electron microscopy and BET analysis. Non porous silica particles were also used for comparison and it was found that the loading of drug was increased with increasing the pore diameter and functionalization of the MSNs. However, the release was not favored by the functionalization. Unfunctionalized MSNs showed best loading to release ratios.