The hairy root culture of *Plumbago indica* L., induced by *Rhizobium rhizogenes* strain K599, was found to regenerate plantlets easily, but morphological characters of plantlets were different from those of the original species. These phenotypic differences could be divided into four different groups. *Group I*: plants had long and elliptical blade with curly leaf edge and wrinkled leaf surface and plants were dwarf. *Group II*: plants had general characters similar to *group I* plants except they were tall. *Group III*: plants had short and elliptical blade with curly leaf edge and wrinkled leaves, found only on the top of the plant, and they were dwarf. *Group IV*: plants had general characters similar to the *group I* but had a higher degree of abnormality such as the leaf apex was tightly rolled backward and plants were extremely dwarf. The detection of *rol* genes by PCR technique and the analysis by HPLC of plumbagin content in root of these plantlets were performed to determine the relationship between the abnormal characters and the presence of the *rol* genes that were transferred from bacterial to plant genome. The results revealed that *rolA* gene might be involved in the dwarfism. However, the plants that contained *rolA*, *rolB* and *rolC* genes were found to be much more abnormal than that contained only 1 or 2 genes. The *rolC* gene might be involved in the increase of plumbagin content as the roots of the *rolC* containing plants showed higher plumbagin content than those of the control roots. For *rolB*, the correlation to the phenotypes was not clearly relevant. Southern hybridization analysis of some plants that contained all of 3 *rol* genes but differed in morphological character showed that they contained 1 or 2 copies of *rol* genes. The analysis of genetic variation in all lines using AFLP technique with 35 pairs of primers generated 1,270 DNA bands with 174 polymorphic bands, which were equal to 13.7% of total bands. It was indicated that the genetic variation was induced in *P. indica* L. regenerated from hairy root.