## **Characterization of Herberine Hydrochloride Nanoparticles**

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Abstract : A drug-loaded nanoparticles containing berberine hydrochloride (BH/FA-CTS-NPs) was prepared. The physicochemical characterizations of BH/FA-CTS-NPs and the inhibitory effect on the HeLa cells were investigated. Folic acidconjugated chitosan (FA-CTS) was prepared by amino reaction of folic acid active ester and chitosan molecules; BH/FA-CTS-NPs were prepared using ionic cross-linking technique with BH as a model drug. The morphology and particle size were determined by Transmission Electron Microscope (TEM). The average diameters and polydispersity index (PDI) were evaluated by Dynamic Light Scattering (DLS). The interaction between various components and the nanocomplex were characterized by Fourier Transform Infrared Spectroscopy (FT-IR). The entrapment efficiency (EE), drug-loading (DL) and in vitro release were studied by UV spectrophotometer. The effect of cell anti-migratory and anti-invasive actions of BH/FA-CTS-NPs were investigated using MTT assays, wound healing assays, Annexin-V-FITC single staining assays, and flow cytometry, respectively. HeLa nude mice subcutaneously transplanted tumor model was established and treated with different drugs to observe the effect of BH/FA-CTS-NPs in vivo on HeLa bearing tumor. The BH/FA-CTS-NPs prepared in this experiment have a regular shape, uniform particle size, and no aggregation phenomenon. The results of DLS showed that mean particle size, PDI and Zeta potential of BH/FA-CTS NPs were  $(249.2 \pm 3.6)$  nm,  $0.129 \pm 0.09$ ,  $33.6 \pm 2.09$ , respectively, and the average diameter and PDI were stable in 90 days. The results of FT-IR demonstrated that the characteristic peaks of FA-CTS and BH/FA-CTS-NPs confirmed that FA-CTS cross-linked successfully and BH was encapsulated in NPs. The EE and DL amount were  $(79.3 \pm 3.12)$ % and (7.24 ± 1.41) %, respectively. The results of in vitro release study indicated that the cumulative release of BH/FA-CTS NPs was (89.48±2.81) % in phosphate-buffered saline (PBS, pH 7.4) within 48h; these results by MTT assays and wund healing assays indicated that BH/FA-CTS NPs not only inhibited the proliferation of HeLa cells in a concentration and time-dependent manner but can induce apoptosis as well. The subcutaneous xenograft tumor formation rate of human cervical cancer cell line HeLa in nude mice was 98% after inoculation for 2 weeks. Compared with BH group and BH/CTS-NPs group, the xenograft tumor growth of BH/FA-CTS-NPs group was obviously slower; the result indicated that BH/FA-CTS-NPs could significantly inhibit the growth of HeLa xenograft tumor. BH/FA-CTS NPs with the sustained release effect could be prepared successfully by the ionic crosslinking method. Considering these properties, block proliferation and impairing the migration of the HeLa cell line, BH/FA-CTS NPs could be an important compound for consideration in the treatment of cervical cancer.

 ${\bf Keywords:} folic-acid, chitosan, berberine hydrochloride, nanoparticles, cervical cancer$ 

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