

Preparation and biological evaluation of paclitaxel loaded biodegradable PCL/PEG nanoparticles for the treatment of human neuroendocrine pancreatic tumor in mice

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Abstract

The main aim of this study was to develop and evaluate nanoparticulate system of paclitaxel loaded polymeric biodegradable Poly (ϵ -caprolactone) nanoparticles for targeting to neuroendocrine pancreatic tumors in mice. Nanoparticles were prepared by simple emulsion technique having surface modification with pluronic F-68. All formulations were evaluated for particle shape and size, zeta potential, encapsulation efficiency and *in vitro* drug release. Radiolabelling of nanoparticles with ^{99m}Tc was done for scintigraphy and biodistribution studies. Cytotoxicity studies were performed on BON-1 cell line using MTT cell proliferation assay. The *in vivo* tumor inhibition study was performed after i.v. administration of paclitaxel nanoparticles. Our results showed that optimized nanoformulation was found to have size range from $100\pm 0.03\text{nm}$ to $250\pm 0.06\text{nm}$ with smooth spherical shape. Negative zeta potential value confirmed the surface modification and stability of nanoformulations. The amount of drug released after 24h from the formulation was found to be $73.3\pm 2.7\%$. More pronounced cytotoxicity was found with nanoparticulate formulation as compared with paclitaxel. The PCL-Ptx nanoparticles reduced tumor volume significantly in comparison with paclitaxel. Higher concentration of Ptx-NPs were found in tumor which was also revealed by high quality scintigraphic image of BON-1 tumor bearing mouse model. In conclusion, polymeric nanoparticulate formulation of paclitaxel was very much efficient for chemotherapy of human pancreatic neuroendocrine tumor in mice.

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Introduction

The majority of neuroendocrine tumors (NET) occur in the gastrointestinal tract (67.5% and the bronchopulmonary system (25.3%). Within the gastrointestinal tract, most NET occur in the small intestine (41.8%), rectum (27.4%) and stomach (8.7%). Less than 1% of NET occur in the pancreas. The incidence of gastroenteropancreatic NET is approximately 2.5 to 5 cases per 100 000 in the United States, which makes them much rarer than adenocarcinomas of the gastrointestinal tract.

The incidence and prevalence of gastroenteropancreatic NET, including gastrointestinal carcinoid tumors, have been substantially increasing in the United States (US) population over the last 30 years for unknown reasons. In particular, there has been a marked increase in diagnoses among African Americans. Typically, patients experience long delays before a diagnosis (5-7 years), and most lack access to the multidisciplinary care necessary for optimal management of these complex tumors. Disappointingly, in the last 30 years, there has been no change in mean overall survival for US patients with gastroenteropancreatic NET [1, 2]; the majorities are still diagnosed with metastatic disease, and no specific antineoplastic treatment exists. However, their incidence has increased substantially in the past 30 years, as indicated by an analysis of the National Cancer Institute's Surveillance, Epidemiology and End Results database.

Neuroendocrine tumors are a highly prevalent and serious condition with limited therapeutic options, particularly in advanced stages. A large number of drugs are used for its treatment. Octreotide a somatostatin analog is also useful for this purpose. The drug suffers with various adverse effects. However these problems can be overcome by encapsulating the drug in

nanocarriers which would impart good therapeutic impact and reduce toxicity. In last two decades, paclitaxel (Ptx) has been one of the most important potent anticancer drug used effectively for the treatment of different types of tumors, including ovarian, breast, lung and pancreatic cancer. Paclitaxel has also shown widespread activity against cancer [3-7]. Induction of apoptosis has been produced by Ptx because of the various pharmacological effects in human cancer cells including polymerization and stabilization of microtubules, resulting in a loss of the natural microtubules' dynamics, blockade of cells at the G/M phase of the cell cycle and inhibition of DNA synthesis [8, 9].

Low aqueous solubility is the major problem associated with Ptx formulations. Solubility in cremophor EL-ethanol (1:1 v/v, Taxol®) and diluted with sterile 0.9% (w/v) sodium chloride just prior to administration is associated with severe life-threatening hypersensitivity reactions as well as leaching of i. v. infusion sets [10, 11]. There is always the problem in developing formulations for Ptx due to the low aqueous solubility of the drug. Alternative Ptx formulations have included cosolvents, emulsions, cyclodextrin complexes, liposomes, and microspheres [12-17]. Because of the limitations of the various other alternative Ptx formulations, biodegradable polymeric microspheres has gained the wide acceptance, which are physically stable and are not known to produce toxicity. As a result, advanced cases of cancer have even been successfully treated with controlled release polymeric microsphere formulations [18-20]. The microspheres can improve treatment efficacy while reducing toxicity [21-23]. The intravenously injected poly lactic glycolic acid-based, Ptx-loaded microparticles for the treatment of lung cancer have been proposed by the group of Horikoshi (1996) [24].

Poly (ϵ -caprolactone) (PCL) possesses unique properties such as higher hydrophobicity and neutral biodegradation end products, which do not disturb the pH balance of the degradation medium among US Food and Drug Administration approved polyesters [25, 26]. Over the years, using PCL as polymeric material, drug delivery systems have been developed [27-29]. In achieving efficient surface modification strategy, higher hydrophobicity of PCL plays a key role that is purely dependent on hydrophobic interactions between the center block (poly propylene oxide) of the stabilizing surfactant and polymeric core (PCL nanoparticle) [30]. The purpose of the present study was to develop and biologically evaluate Ptx-loaded PCL/PEG based nanoparticles against BON-1 human neuroendocrine pancreatic tumor induced in mice.

Materials

Paclitaxel was the generous gift from Dr. Reddy's Laboratories Ltd. (India) and was stored at 4°C. Analysis by High Performance Liquid Chromatography (HPLC) showed that Ptx was >99% pure. Poly (ϵ -caprolactone) PCL 85000Da, polyethylene glycol (PEG) 5000Da, stannous chloride dehydrated ($\text{SnCl}_2 \cdot 2\text{H}_2\text{O}$), tween 80 and cremophor EL were purchased from Sigma-Aldrich. Dichloromethane (DCM) and dimethylsulfoxide (DMSO) were purchased from Merk. Pluronic® F-68, a non-ionic surfactant composed of poly (ethylene oxide)/poly (propylene oxide)/poly (ethylene oxide) triblock copolymer, was obtained from Sun pharmaceuticals Ltd. (India). Solvents of HPLC were purchased from Fisher Scientific Company (Fair Lawn, NJ, USA).

Cell culture

Monolayer cultures of human pancreatic carcinoma BON-1 cells (obtained from Institute for Biophysics, University of Frankfurt, Germany) was maintained at 37°C in a humidified CO_2 incubator (5% CO_2 , 95% air) in Dulbecco's Modification of Eagle's Medium (DMEM) (Sigma, USA) supplemented with 10% foetal calf serum (GIBCO, USA), 50µg/mL penicillin, 50µg/mL streptomycin sulfate and 2µg/mL nystatin.

Methods

Preparation of paclitaxel loaded nanoparticles

Nanoparticles (NP) were prepared by simple emulsion technique [31]. Three milligrams of Ptx was mixed with the polymer solution having 50mg of PCL and 5% PEG in 3.0mL of dichloromethane and were emulsified in to the 2.0mL of 0.25% pluronic F-68 before a sonication of 15sec (70W) LABSONIC U sonicator. The emulsion formed was added drop-wise on 500mL of 0.5% pluronic F-68 under magnetic stirring at 1000rpm at room temperature and ambient pressure used to stabilize the emulsion and prevent nanoparticle aggregation during the process. To evaporate dichloromethane, the solution was stirred at room temperature under magnetic stirring or under reduced pressure at 35°C for approximately 1h. To remove the non-encapsulated drug, the suspensions were filtered (0.22µm) and ultra centrifuged at 22,000g for 10min at 4°C. The pellets were suspended in 10mL of ultra-purified water and were centrifuged (Eppendorf). This procedure was repeated three times to remove the residual surfactant and lyophilized for 48h LABCONCO, triad™ lyophilizer. Then lyophilized Ptx loaded nanoparticles (Ptx-NP) were stored in refrigerator at 4°C.

Physicochemical characterization of nanoparticles

Surface morphology. Zeta potential measurements. Encapsulation efficiency

The shape and morphological examination of nanoparticles was performed using a transmission electron microscope (TEM) following negative staining with sodium phosphotungstate solution (0.2%, w/v), at various magnifications.

Suitably diluted aqueous suspension of nanoparticles at concentration of $1.0 \pm 0.3 \text{ mg/mL}$ was taken for zeta potential measurements using Malvern Zetasizer (DTS Ver, 4.10, Malvern Instruments).

The drug loading efficiency was determined in triplicate by HPLC. The mobile phase consisted in acetonitrile/water (70:30v/v). The column temperature was maintained at 30°C. The flow rate was set at 1.0mL/min and the detection wavelength was 227nm. Sample solution was injected at a volume of 50µL. The HPLC (Agilent 1200 series) was calibrated with standard solutions of 5 to 100µg/mL of Ptx dissolved in acetonitrile (correlation coefficient of $R^2=0.9965$). Nanoparticles were dissolved in acetonitrile and vigorously vortexed to get a clear solution. The encapsulation efficiency was defined by the ratio of measured and initial amount of Ptx encapsulated in nanoparticles.

In vitro release study

Based on the method given by Burt et al (1995) [32] with some modifications, Ptx nanoparticles (3mg) were suspended in 3mL of 10mM phosphate buffered saline (PBS) with 0.4% albumin (pH=7.4) containing 0.1%w/v tween 80. The tubes were placed into an orbital shaker maintained at 37°C and

shaken at 300rpm. At specific time interval, the tubes were centrifuged at 4000g for 5min and 2.0mL of the supernatant was carefully removed and 1mL of DCM was added to the PBS samples. The contents were then allowed to phase separate for 30min and the aqueous phase (drug-free) was aspirated from the 1mL DCM phase (drug-rich). The DCM was then evaporated to dryness at 40°C under nitrogen gas and the contents were reconstituted in 1mL of acetonitrile: water (60:40v/v) and analyzed by HPLC methods (C18 reverse phase column, a mobile phase of 58:37:5 acetonitrile: water: methanol flowing at 1mL/min) at ultraviolet spectroscopy detection at 227 nm.

Radiochemical synthesis of technetium-99m (^{99m}Tc) Ptx nanoparticles

Paclitaxel was labeled with ^{99m}Tc by direct labeling method as described by others [33]. For radiolabelling, 0.1mL of ^{99m}Tc (185MBq/mL; obtained by solvent extraction method from molybdenum) was mixed with 0.1mL of stannous chloride solution in 10% acetic acid solution (1mg/mL), and the pH was adjusted to 7.0 using sodium bicarbonate solution (if required). To this solution, 1mL of Ptx solution (1mg/mL) was added and incubated for 15min at room temperature. Paclitaxel nanoparticles were labeled in a similar way. The labeled formulations were stored in sterile evacuated sealed vials for subsequent studies.

Radiochemical purity of ^{99m}Tc Ptx nanoparticles

Radiolabelling efficiency was determined by ascending thin layer chromatography (ITLC)-Silica gel coated fibre sheet (Paul Gelman science Inc., USA) using 100% acetone as a mobile phase. A measured amount of 2-3μL of radiolabeled complex was applied at a point 1cm from one end of an ITLC-SG strip. The strip was developed in acetone and the solvent front was allowed to reach 8cm from the origin. Each TLC was cut in 0.5cm segment and counts of each segment were taken. The radioactivity was counted by Biodex scintillation well counter; USA. The free ^{99m}Tc, which moved with the solvent ($R_f=0.9$), was calculated. By subtracting the activity moved with solvent front by using acetone from the net amount of ^{99m}Tc with paclitaxel and ^{99m}Tc with Ptx nanoparticles not activity was calculated.

In vitro biological evaluation

Cytotoxicity of Ptx nanoparticles

Cellular cytotoxicity was assessed by using tetrazolium dye based (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT), a yellow tetrazole) assay. Exponentially growing cells were plated in 96-well microtitre plate at a uniform cell density of 10,000 cells/well 24h before treatment. Cells were treated with varying concentrations of Ptx in only controls and Ptx nanoparticles (0.025, 0.25, 2.5, 12.5 and 25μg/mL range) for 72h and MTT assays were performed. At the end of treatment, negative control and treated cells were incubated with MTT at a final concentration 0.05mg/mL for 2h at 37°C and the medium was removed. The cells were lysed and the formazan crystals were dissolved using 150μL of DMSO. The absorbance of individual wells was noted on 570nm via an ELISA plate reader at 25.8°C. Average values in triplicate were subtracted from average value of control and the survival fraction of cells was calculated by the formula:

$$\text{Survival fraction} = \frac{\text{Absorbance of experimental group}}{\text{Absorbance of control group}} \times 100$$

In vivo biological evaluation

Tumor transplantation

Athymic mice (10-12 weeks old) used in these studies were obtained from the institute's central animal facility and weighed 18-20gr at the time of tumor implantation. The institutional animal ethics committee of Institute of Nuclear Medicine and Allied Sciences (INMAS), New Delhi had duly approved the protocol for in vivo studies. BON-1 tumor cells obtained from Institute for Biophysics, University of Frankfurt, Germany were maintained by serial passage of tumor cell suspension in the peritoneal cavity of strain A mice. Subcutaneous tumors were implanted by intramuscular injection of 15×10^6 cells (in 0.1-0.15mL volume) into right hind leg. Tumor diameter was calculated with the help of a electronic caliper. Experiments were performed when the tumor had attained a diameter of 9 ± 0.3 mm (5-6 days after implantation).

Tumor growth inhibition studies

Mice bearing subcutaneous tumor were treated with the nanoparticles intravenously through the tail vein. The injection volume of the treatment was prepared in normal saline and was 0.1-0.15mL. When tumors reached 9.0 ± 0.3 mm in diameter, the mice were divided into three groups (six animals each group). The first group was taken as control and the second group was injected with Ptx nanoparticles (Ptx conc.1mg/kg); mice in the third group were treated with Ptx only controls at a dose of Ptx concentration of 1mg/kg in every second day for 14 days. The animals were kept under observation. The results were statistically analyzed by using Student's t-test with a 95% confidence level ($P \leq 0.05$), and were reported as mean standard deviation (SD).

Tumor scintigraphy

Tumor imaging was performed in BON-1 cell line implanted in tumor bearing nude mice by administering 100μL of the ^{99m}Tc labeled Ptx nanoparticles preparation containing 2.96MBq of radioactivity. Images were taken using planar gamma camera (SPET, LC 75-005, Diacam, Siemens, Hoffman Estates, IL, USA). Images were obtained at different time intervals starting from 15min to 24h after post injection.

Biodistribution studies

For analyzing the distribution of nanoparticulate formulation of Ptx in tumor bearing mice, we divided the mice into three groups having six mice in each group. After that, administration of drug was done through the tail vein. The mice of first group were injected with Ptx (10mg/kg), second with Ptx loaded nanoparticles (^{99m}Tc labeled Ptx), third group of mice was kept as control. For biodistribution experiment, the animals were sacrificed by cervical dislocation after 1, 2, 4h time periods. Surface blood was washed with cold water and immediately after radioactivity was counted.

Statistical analysis

Pearson correlation coefficients were used to evaluate the extent of a relationship between two data sets. Coefficients of determination were calculated. Statistical differences among groups were analyzed using ANOVA. A P-value of 0.05 was considered statistically significant.

Results

Characterization of Ptx loaded nanoparticles

The method was optimized to produce drug nanoparticles of the small size having high entrapment efficiency. The formulation characteristics of the optimized formulations prepared by PCL/PEG in presence of pluronic F-68 are given in Table 1. The nanoparticles were generally spherical in shape and do not show any detectable free drug crystals. The average range of nanoparticles was 100 to 250 nm as visualized by the TEM (Fig. 1). No appreciable drug degradation (< 0.1%) was detected under storage at 4°C for three months.

Table 1. Formulation characteristics for the optimized nanoparticle formulations

Diameter (nm) (mean±SD)	236.3±1.06
Polydispersity index (PI) (mean±SD)	0.10±0.03
Zeta potential (ZP ⁻) (mV) (mean±SD)	-21.60±0.18
Encapsulation efficiency (EE* (%)) (mean±SD)	84.15±1.01

+ Polydispersity index; - Zeta potential; * - Encapsulation efficiency; Each value is mean of three independent determinations.

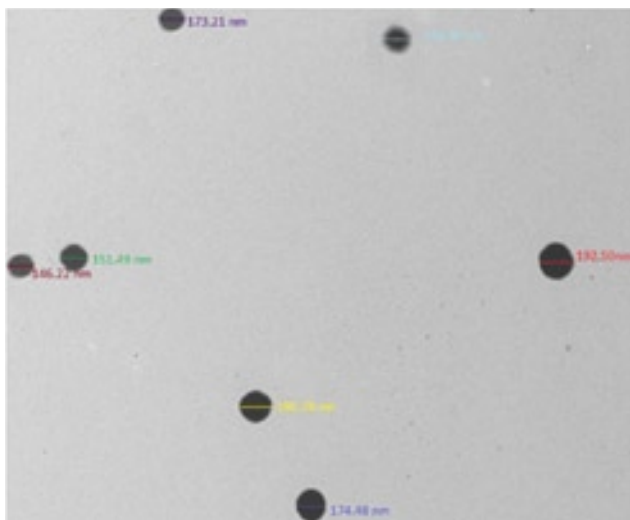


Figure 1. TEM photograph of Ptx loaded nanoparticles.

In vitro release profile

In vitro percentage drug release-time profile of Ptx-NP in release media was investigated at 37°C shown in Figure 2. After the initial burst release for about 4h, Ptx release followed zero order kinetics. Release of Ptx in the first 4h was equivalent to 39.5±0.29% of the initial drug load of nanoparticles. After that, amount of Ptx released was 66.1±0.69% at the first 30h.

Cytotoxicity study

Figure 3 shows the cytotoxicity of free Ptx and Ptx loaded nanoparticles by the MTT assay using the BON-1 cell line. The range of concentrations of Ptx (0.025 to 25µg/mL) corresponds to plasma levels of the drug achievable in humans [34]. It was observed that the cytotoxicity is positively correlated to its concentration for Ptx only controls and Ptx nanoparticles. The cytotoxicity of free Ptx and Ptx nanopar-

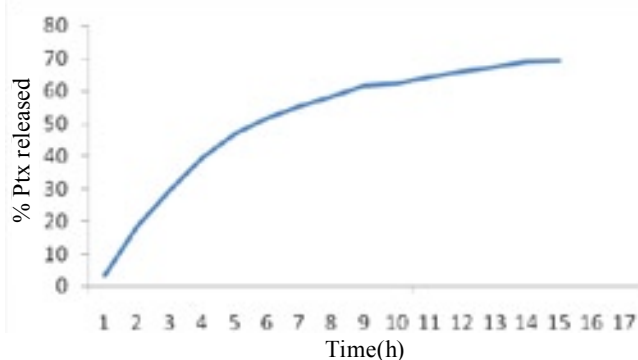


Figure 2. In vitro release pattern of paclitaxel from PCL/PEG nanoparticles. Mean±SD (n=3); (P≤0.05).

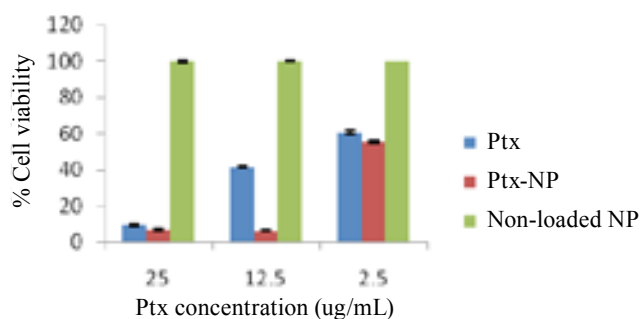


Figure 3. Colorimetric estimation of the mitochondrial activity for cytotoxicity of Ptx nanoparticle and Ptx (MTT assay) in BON-1 cell line after an incubation time of 72h at different concentrations.

ticles was not significantly different at the concentration of 0.25µg/mL. As the concentration increased, the difference of the cytotoxicity between the Ptx and Ptx-NP became obvious. At the concentration of 12.5µg/mL and 2.5µg/mL, all nanoparticles exhibited more prominent cytotoxicity than free Ptx.

Radiochemical synthesis and purity of nanoparticles

On the basis of chromatographic analysis, the radiolabeling efficiency was found to be more than 98% consistently. Technetium-99m has been used to directly label preformed nanoparticles based on stannous chloride as reducing agent. In the present study the radiolabelling efficiency on nanoparticles was always more than 90%. Technetium-99m (in the form of pertechnetate) is easily available, cost effective, and a low radiation dose. The half-life of 6h is long enough to synthesize the ^{99m}Tc-labeled radiopharmaceuticals and perform imaging studies. Reduced ^{99m}Tc is added to the nanoparticles, which can easily bind to the nanoparticles surface. This surface labeling approach is technically simple and good for the production of nanoparticles in a pharmaceutically acceptable form, and therefore used for the present study.

Tumor regression studies

The *in vivo* antitumor efficacy of paclitaxel loaded nanoparticles and free Ptx was evaluated in BON-1 tumor bearing mice. The Ptx-loaded nanoparticles inhibited tumor growth most efficiently, followed by free Ptx (P<0.05) (Fig. 4). The tumor volume was excessively enlarged (about 16.6mm) of the control (PBS) group on the 6th day and its tumor inhibition

study has been stopped while other groups lasted until the 9th day for free Ptx and the 13th day for Ptx-loaded nanoparticles ($P < 0.001$) (Fig. 4). Body weight measurements showed no significant differences between the groups throughout the study. There was a slight increase in body mass as a result of natural animal growth.

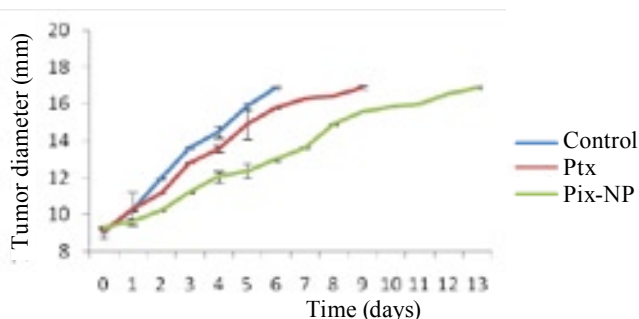


Figure 4. In vivo tumor growth inhibition study of Ptx loaded nanoparticles on BON-1 implanted tumor bearing mice.

Tumor scintigraphy

Scintigraphic images of ^{99m}Tc labeled Ptx nanoparticles in tumor grafted nude mice showed rapid accumulation of radioactivity in tumor. Imaging of animals was carried out at different time intervals after administering labeled compound via tail vein. The mice depicted the beginning of accumulation of activity in tumor at 30min, which reached to maximum at 1h (Fig. 5) and remained almost stable for 4h.

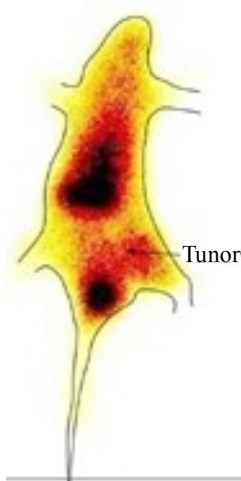


Figure 5. Whole body SPET image of female nude mice with subcutaneous somatostatin receptor positive tumor in right thigh after 1h following intravenous administration of ^{99m}Tc labeled Ptx nanoparticles (2.96MBq activities).

Biodistribution studies

To evaluate the potential significance of the nanoparticles uptake by various tissues with regard to a cytotoxic drug, the biodistribution study was performed using Ptx with ^{99m}Tc either as free complex or encapsulated in nanoparticles. These experiments were performed in BON-1 bearing mice models. The percentage of ID/g distribution to tissues in different organ at different time intervals for ^{99m}Tc Ptx only controls and ^{99m}Tc labeled Ptx nanoparticles are shown in Figure 6A and B. As expected drug loaded nanoparticles showed greater activity in tumor in comparison with Ptx only controls. It is clear from the tumor concentration of Ptx that PCL nanopar-

ticulate formulations could deliver a significantly higher concentration of Ptx in to the tumor than Ptx only controls. Depending on the dosage form (pure drug in solution or nanoparticles), the distribution pattern appears to be different. It was evident that free paclitaxel was eliminated from the body at a much faster rate than was any of its nanoparticulate formulation. The overall uptake of nanoparticle formulations by spleen was significantly lower than free Ptx.

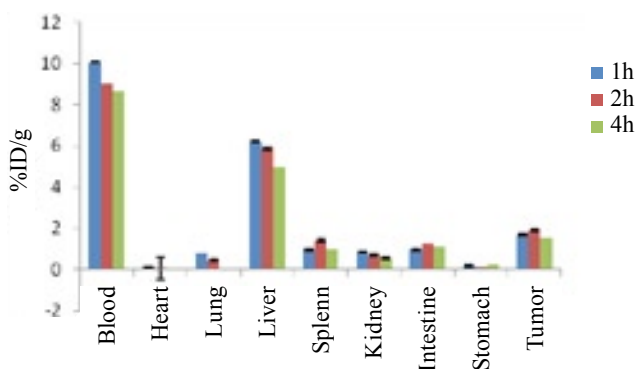
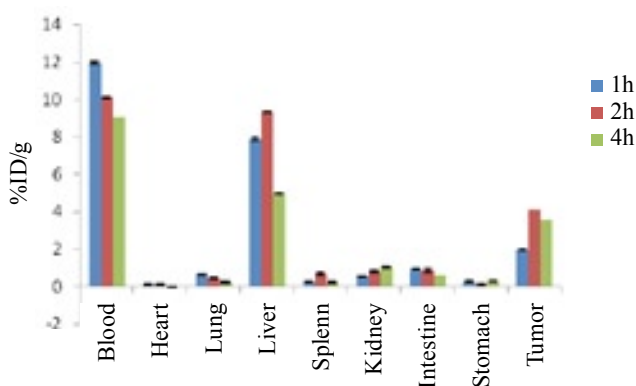


Figure 6A. Biodistribution of ^{99m}Tc Ptx in BON-1 cell tumor grafted in athymic mice injected with 3.7MBq activities.



Mean \pm SD; (n=3); $P < 0.05$

Figure 6B. Biodistribution of ^{99m}Tc labeled Ptx nanoparticles in BON-1 cell tumor grafted in athymic mice injected with 3.7MBq activities.

Discussion

Successful optimization of Ptx-loaded PCL nanoparticles yielded spherical and nanometric size range (100-250nm) nanoparticles. The encapsulation efficiency was high, averaging about 84%. This may be a result of the retention of Ptx in the organic phase, due to its high oil/water partition coefficient of >99 , and thereby became encapsulated as the nanoparticles solidify [35]. Consequently, the larger nanoparticles, which have smaller surface area/volume ratio, showed higher encapsulation efficiencies. The zeta potential was found to be negative, implying that the nanoparticle surface was negatively charged, confirming the presence of PEG chains as itself uncharged in nature. The burst release of Ptx may be due to the dissolution and diffusion of the drug that was poorly entrapped in the polymer matrix, while the slower and continuous release may be attributed to the diffusion of the drug localized in the PCL core of the nanoparticles. This sustained release could mainly result from the erosion and degradation of the polymer because Ptx has very poor dissolution in water [36].

As to *in vitro* evaluation, cytotoxicity of both free Ptx and Ptx-loaded nanoparticles increased as the drug concentration grew from 0.025 to 25µg/mL. The antitumor effect of the Ptx-loaded nanoparticles was similar to free paclitaxel at lower paclitaxel concentration while the cytotoxicity of the nanoparticles was superior to free paclitaxel at higher concentrations. This superiority may mainly due to the uptake of the nanoparticles and the sustained release of Ptx inside the cancer cells. It should be emphasized that in the case of Ptx a significant effect was attributed to the excipient cremophor EL (absent in nanoparticles), whereas in the case of Ptx-loaded nanoparticles the cytotoxicity observed was only attributed to Ptx (drug-free nanoparticles were non-cytotoxic). This decrease in cell viability, measured by the MTT test can result from an inhibition of cell growth or from cytotoxicity. Tumor growth inhibition is more pronounced as compared to the control group. When encapsulated into nanoparticles, Ptx could attain tumor site through enhanced permeation and retention (EPR) effect and maintain the effective therapeutic concentration for a long period of time. By non-specific endocytosis, the PCL nanoparticles will be taken up by the tumor cells which rapidly increase the intracellular concentration of the drug resulting in maximal pharmacological effect [37].

Paclitaxel-NPs were distributed mainly in tumor and liver and small quantities being found in kidney and spleen and are revealed by the scintigraphic images of tumor bearing mouse. Ptx-NPs accumulated only slightly in the heart and lung, and lower the toxic effect of Ptx in the heart and lungs. In the present study, we have used pluronic F-68 having 30 residues of propylene oxide (PO) and 76 residues of ethylene oxide (EO) residues. This mode of adsorption leaves the hydrophilic PEO side arms in a mobile state as they extend outwards from the particle surface and provide stability to the particle suspension by repulsion effect through a steric mechanism of stabilization involving both enthalpic and entropic contributions [38]. The end result of such an assembly is a stable, slow-eroding colloidal system that is less phagocyte-prone and hence long circulating. Once accumulated within the tumor interstitium by exploiting vascular abnormalities that allows freer access to the tumor mass, the PCL nanoparticle system would increase the drug concentration inside the tumor cells as a result of non-specific endocytic process, followed by gradual release of the drug [39].

Social utility of work lies in its therapeutic potential causing healing of tumors at high rate as compared to non-encapsulating drug. Paclitaxel is known to cause various side effects which affect lifestyle of the cancer victims. Use of Ptx loaded nanocarrier showed beneficial effects in terms of reduced toxicity, adequate bioavailability and highly acceptable therapeutic index. Nanocarriers are promising drug delivery systems that provide agreeable degree of therapeutic effect.

In conclusion, all our experimental results show that nanoparticulate formulation of Ptx effectively reduces the volume of human neuroendocrine pancreatic tumour injected in mice and thereby has the promising potential to improve the therapeutic efficacy of Ptx and reduce drug associated toxicity.

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The authors declare that they have no conflicts of interest.

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