RADIOPHARMACEUTICALS IN NANOSCALE: DEVELOPMENT OF LABELING PROCESS FOR NANOMEDICINE.

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INTRODUCTION

Drug delivery systems as polymeric microparticles represents a significant development area in the pharmacy field. Both the investment and research has been increasing each day. The polymeric microparticles have great stability, industrial capacity and allow adjustments to achieve the suitable release profile and /or direction for a particular site of action. The use of poly(lactic-co-glycolic) acid nanoparticles (PLGA NPs) has emerged as a powerful potential methodology for carrying small and large molecules of therapeutic importance as well as scaffolds for tissue engineering applications. Polymeric micelles are used as pharmaceutical carriers to increase solubility and bioavailability of poorly water-soluble drugs. Different ligands has been used to prepare targeted polymeric micelles. Liposomes have a decade-long clinical presence as nanoscale delivery systems. However, their use as delivery systems of nanoparticles is still in the preclinical development stages. Liposome-nanoparticle hybrid constructs present great opportunities in terms of nanoscale delivery system engineering for combinatory therapeutic-imaging modalities. Moreover, many novel materials are being developed in nanotechnology laboratories that often require methodologies to enhance their compatibility with the biological milieu *in vitro* and *in vivo*.

The recent developments of nuclear medicine in oncology have involved numerous investigations of novel specific tumor-targeting radiopharmaceuticals as a major area of interest for both cancer imaging and therapy, moreover combining the more recent nuclear imaging multimodalities which provide high sensitivity and anatomical resolution such as PET/CT (positron emission tomography/computed tomography) and SPECT/CT (combined single photon emission computed tomography/computed tomography system) with the use of these specific tumor-targeting carriers constitutes a promising rally which will, hopefully in the near future, allow for earlier tumor detection, better treatment planning and more powerful therapy.

OBJECTIVE

The aim of this study is highlight the use, limitations, advantages and possible improvements of different nano and microcarriers as potential vehicles for radionuclides delivery in cancer nuclear imaging and radiotherapy.

METHODS

Nanoparticules and Microparticules

Five samples of nanoparticules and Microparticules were analyzed, as follow: samples I and II are micelles, constituted by DSPE-PEG, TGPS and tamoxifen; sample III is a nanocapsule of PLA and tamoxifen and sample IV is also a nanocapsule made of PLA-PEG and tamoxifen, all obtained from the Laboratório de Tecnologia Farmacêutica USP-Ribeirão Preto.

Chromatography

The labeling process consisted on a 150 µL withdrawn of the nanoparticules and microparticules solution, each of them were incubated with stannous chloride ($SnCl_2$) solutions (80 µL/mL) obtained from Sigma-Aldrich for 20 minutes (room temperature). Then, another incubation followed with 100 µCi (300 µL) of freshly eluted technetium-99m (IPEN/CNEN) for other 10 minutes.

In order to characterize the labeled nano and microparticules whatman nº 1 chromatography was performed by using 2 µl of labeled sample in acetone (Proquimios) as mobile phase. The radioactivity of the strips was verified in a gamma counter (Packard, Cobra II).

Biodistribution

Biodistribution studies were done with ten mices, two for each nanoparticle labeled sample (I, II, III and IV) and two for the microparticules sample. The

labeled samples (3,7 MBq/0,2 mL) were administred after catheterization of the jugular. Planar images were obtained 30 minutes post-injection at a Milennium Gamma Camera (GE Healthcare, Cleveland, USA). Counts were acquired for 5 min in a 15% window centered at 140 KeV. Then, animals were sacrificed and their organs removed, weighted and the radioactivity uptake counted in a gamma counter (Packard-Cobra II). Results were expressed as percentage of injected dose per gram of tissue.

RESULTS

Whatman n°1 chromatography

Results are shown in tables I, II and III.

TABLE I– Ascending chromatographies of the ^{99m}Tc-sample I and ^{99m}Tc-sample II comparing to free pertechnetate ($Na^{99m}TcO_4^-$).

Samples	Solvent	Bottom (%)	Top (%)
^{99m}Tc-sample I	Acetone	80.1	19.9
^{99m}Tc-sample II	Acetone	86	14
$Na^{99m}TcO_4^-$	Acetone	0.3	99.7

TABLE II – Ascending chromatographies of the ^{99m}Tc-sample III and ^{99m}Tc-sample IV comparing to $Na^{99m}TcO_4^-$.

Samples	Solvent	Bottom (%)	Top (%)
^{99m}Tc-sample III	Acetone	92.2	7.8
^{99m}Tc-sample IV	Acetone	87.1	12.9
$Na^{99m}TcO_4^-$	Acetone	0.3	99.7

Biodistribution Studies

The results for each labeled sample are in the tables below:

TABLE III– Biodistribution %gram per tissue versus organ of the labeled samples in mice:

Organs	Sample I	Sample II	Sample III	Sample IV
Heart	1,88±0,69	1,17±0,95	0,61±0,43	4,12±0,38
Right lung	2,31±0,43	0,86±0,83	1,04±0,82	4,48±1,52
Left lung	2,29±0,40	0,94±0,91	1,20±0,80	3,06±2,10
Liver	8,06±1,75	3,19±4,33	9,08±2,49	8,41±0,01
Spleen	1,42±0,33	1,43±1,51	2,34±0,25	2,49±0,39
Stomach	0,81±0,40	1,04±0,07	0,21±0,09	1,34±0,77
Intestine	0,46±0,08	1,88±0,96	0,17±0,12	0,92±0,60
Right kidney	8,93±0,86	4,49±6,17	2,72±0,86	8,52±2,37
Left kidney	8,88±1,01	4,50±6,17	2,70±0,96	8,05±2,51

CONCLUSION

All the structures analyzed in this study showed a great behavior in labeling process. Moreover, the labeling process seems to be a important technique in the evaluation of nanoescales products. Otherwise the production of nanoradiopharmaceuticals is absolutely possible since the labeling process of nanoescales is viable.

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