Integration of Fricke gel dosimetry with Ag nanoparticles for experimental dose enhancement determination in theranostics

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Abstract

The use and implementation of nanoparticles in medicine has grown exponentially in the last twenty years. Their main applications include drug delivery, theranostics, tissue engineering and magneto function. Dosimetry techniques can take advantage of inorganic nanoparticles properties and their combination with gel dosimetry techniques could be used as a first step for their later inclusion in radio-diagnostics or radiotherapy treatments. This work presents preliminary results of properly synthesized and purify silver nanoparticles integration with Fricke gel dosimeters. Used nanoparticles presented mean sizes ranging from 2 to 20 nm, with a lognormal distribution. Xylenol orange concentration in Fricke gel dosimeter was adjust in order to allow sample's optical readout, accounting nanoparticles plasmon. Dose enhancement was assessed irradiating dosimeters setting X-ray beams energies below and above silver K-edge.

Keywords: Fricke gel dosimetry; Ag nanoparticles; Dose enhancement

1.- INTRODUCTION

Radiation dosimetry is devoted to measure the amount of energy deposited by ionizing radiation. Measurements are carried out by dedicated devices, the dosimeters, with the aim of estimating the effective dose received by exposed phantoms or patients. Nowadays, different approaches and techniques are available for achieving reliable radiation dosimetry, including gene expression, somatic mutations, gas ionization, thermoluminescence, optically stimulated luminescence, electron paramagnetic resonance, neutron activation, gels, films, among others; all of them providing specific advantages and disadvantages in terms of accuracy, biological interpretation, analytical dose reconstruction and other issues. From a physical point of view, international protocols for radiation dosimetry clearly establish absorbed dose to water as the reference starting point for any validated traceable dosimetry technique.

Nanoparticles are materials with overall dimensions in the nanoscale, typically considered as one-dimensional nanomaterials when one of their main is larger than the nanoscale and zero-dimensional when all of their dimensions are below 100 nm (Murthy, 2007). Nanoparticles have numerous advantages over their bulk analogues, most of them related to their high surface area to volume ratio, which enhances and favor different types of chemical reactions and provides a large number of surface modifications sites to engineer multifunctional materials. The physicochemical properties of nanomaterials are often better described by atomic or molecular interactions than by common particle or bulk materials models. Because of that, some properties can be tuned by changing the particle size or size distribution obtaining unique final characteristics. These properties and advantages have been exploited in almost any field of application like medicine, food industry, energy or environmental projects. In regard to medicine, there has been a strong interest on using nanoparticles in different areas such as, drug and gene delivery, biodetection of pathogens, detection of proteins, tissue engineering, tumor destruction via heating, separation and purification of biological molecules and cells, phagoketogenic studies, MRI contrast enhancement and fluorescent biological labels (Salata, 2004). Specifically, for oncological purposes and cancer therapy, nanomaterials have been already successfully applied as non-targeted delivery systems with many FDA approved examples and some targeted, stimuli and combined delivery systems which are in Phase II/III (Shi et al., 2016). Also, some nanomaterials have

been approved by the FDA and the EMA as imaging agents (Anselmo and Mitragotri, 2016), which proves the potential and future perspective of nanomaterials. At the same time, a growing number of new promising materials for oncological therapies arise every year that must fulfil certain issues and challenges for their clinical use (Chapman et al., 2013). Currently, there are a broad number of studies on materials that combine imaging and therapy in oncological treatments, which gave birth to theranostic nanoparticles, defined as those that combine the capacity for tumor imaging with therapeutic efficacy and low toxicity. Theranostics is still at the preclinical stage but it provides the opportunity for real-time imaging of tumors while patients are undergoing therapy (Kiessling et al., 2014). There are many types of materials that can be used in theranostics, such as polymers, lipids, dendrimers and inorganics nanocarriers (Lim et al., 2014), and in particular those based on inorganic materials can be used together with X-ray therapy to enhance local dose delivery and to monitor the evolution of the therapy by means of their X-ray fluorescence emission signal. Characteristic X-rays of metallic nanoparticles can be detected outside the patient, making them interesting for biomedical applications and fluorescent emission can produce local dose enhancement within the nanoparticles infused volume (Huang et al., 2011) . Therefore, methods able to account for absorbed dose and possible dose enhancement becomes necessary and extremely for any treatment design. Among the different alternatives, gel dosimetry is suitable tool thanks to its tissue-equivalence properties and ability to register 3D dose distributions (Baldock et al., 2010; Mattea et al., 2017).

The present work focuses on a preliminary analysis on the feasibility and reliability of integrating high Z nanoparticles, like AgNPs, with Fricke gel dosimeters aimed at profiting from the potential individual advantages of the dosimetry system and the AgNPs. *A priori*, AgNP-infused Fricke gel dosimetry may be capable of achieving local dose enhancement, mainly due to secondary electrons produced by silver re-emissions; as well as exhibiting potentiality for external monitoring by detecting and recording Ag K-lines emissions. Spatial distribution of AgNPs concentration may be assessed by detecting Ag K-lines during irradiation, thus providing a promising framework for tumor targeting and, simultaneously, a local dose enhancement for a better protection of the surrounding organs at risk. To this aim, suitable modifications may be necessary for the preparation protocol of standard Fricke gel in

order to achieve a chemically stable and physically useful product. Moreover, synthesis of AgNPs may also require specific modifications or adaptations to obtain adequate AgNP suspensions for this specific application. Besides, dedicated X-ray detection system needs to be incorporated into the proposed configuration for further investigation about the potentiality of correlating the X-ray fluorescent signal with AgNPs spatial distribution and local absorbed dose, even at real-time level.

2.- MATERIALS AND METHODS

2.1.- Nanoparticle synthesis and characterization

In order to obtain silver nanoparticle suitable for gel dosimetry a synthesis process where particles are being formed and stabilized at the same time with a gel dosimetry compatible substance was used as previously described elsewhere (Mattea et al., 2017). Briefly, a 300-mM aqueous solution of silver nitrate (99.9 % acquired from Prodesa S.C.A., Buenos Aires, Argentina) and a 96-mM aqueous solution of gelatin (250 Bloom purchased from Sigma Aldrich, Saint Louis, MO, USA) were mixed and stirred for 60 minutes. The obtained solution was kept in a sealed reactor at 90 ºC for at least 15 hours. The synthesis product was then purified by a dialysis and freeze-drying process ensuring that no unreacted material was present in the final product. The obtained material was then characterized by transmission electron microscope (TEM) with a JEM-JOEL 1200-EX II TEM microscope (USA).

2.2.- Gel dosimeters elaboration, irradiation and characterization

Two different Fricke gel dosimeters were prepared; on one hand, a sensitive material was manufactured based on the method described elsewhere (Valente et al., 2007), where a 250-bloom gelatin solution (3.00 % w/w) was stirred for 20 min at 50 °C. Then, the resulting gel was mixed at 28 ºC with sulfuric acid (1.38 % w/w), xylenol orange (0.04 % w/w) and ferrous sulfate (0.06 % w/w). On the other hand, a AgNPs infused Fricke gel dosimeter was

manufactured with a similar protocol by reducing the xylenol orange concentration to 70 parts per million. Both materials were contained in PMMA cuvettes of 1×1×4.5 cm^3 and stored at 4 °C for at least 12 hours before irradiation. Sulfuric acid and xylenol orange were provided by Sigma Aldrich (USA), and ferrous sulfate was purchased from Research AG, (Buenos Aires, ARG).

For the X-ray beam irradiations, two different irradiation setups were used based on a previous study (Mattea et al., 2017) For the silver fluorescence detection study, the X-ray source generator was set to 50 kVp and the scattered spectrum was collected with an AMPTEK XR-100T Cd-Te γ/X-ray detector. In order to study the dose enhancement, two different incident spectrums were used, the first beam was configured by setting the X-ray generator at 50 kVp and 14.4 mA and the second beam was configured with 25 kVp and 34.5 mA. In both cases, the resulting dose rate was (25.9±0.8) cGy/min, measured by a calibrated ionization chamber (TN 30013, PTW, Freiburg, Germany).

The characterization of the dosimeters was carried out by means of optical methods, in particular, the absorbance change between the non-irradiated and the irradiated materials was measured with a Shimadzu UV-1800 spectrophotometer (Japan) at room temperature.

2.3.- Nanoparticle detection and dose enhancement in AgNP-FXGD

Ionization of Ag electrons is responsible of an appreciable part of the collective dose enhancement effect. Therefore, in order to study dose enhancement, X-rays beams capable or not of exciting silver K-edge (25.5 keV) were used, mentioned before as 25 kVp and 50 kVp. Irradiations were carried out with a 3×3 cm^2 field size, a source-to-phantom distance of 93 cm and a collimator to sample distance of 13.5 cm.

2.3.- Dose enhancement and nanoparticle detection by Monte Carlo simulations

There are numerous Monte Carlo codes properly benchmarked and currently used for different applications of radiation transport, like EGS4 , GEANT4, MCNP, FLUKA and PENELOPE, among others. Specifically, PENELOPE (PENetration and Energy Loss Of

Positrons and Electrons; photon simulation was introduced later) Monte Carlo code 2001 has demonstrated to provide excellent agreements with experimental data for the so called "low energy range", *i.e.* < 0.1MeV, approximately, as it is the case of medical radiology. In this context, PENELOPE appears as a suitable option to simulate AgNPs-infused Fricke gel dosimeters with the aim of characterizing some relevant physical properties as well as evaluating the absorbed dose distributions for different configurations.

PENELOPE main code provides a comprehensive database containing pure elements and some compounds, commonly used during radiation-matter interaction processes, like biological tissues, some plastics and alloys. However, user-specific materials, such as Ag nanoparticles suspended in Fricke gel, require a proper definition by the user, which can be reasonably achieved by using the additivity rules incorporated in the *material.exe* module of PENELOPE. Clearly, the validity of the obtained results is strongly dependent on this approach, which may be adequately applied for relatively low AgNPs concentrations, assumed to be uniformly distributed within the Fricke gel and no relevance minor differences of AgNPs dimensions may be also required. Once all such conditions are satisfied, simulation may provide acceptable description of absorbed dose distribution as well as emerging photon spectrum. Finally, a simplified setup was designed according to the *pengeom* module to sketch 10x10x40 mm^3 spectrophotometry vials containing AgNP-infused Fricke gel, which can be filled at different Ag concentrations, irradiated by X-ray beams, as produced by conventional X-ray tubes, and emerging photon fluence was recorded, as first approximation, integrating all the 4π solid angle.

3.- RESULTS

Obtained results are presented with the following structure: Synthesized AgNPs characterization, AgNP-infused Fricke gel dose-response, AgNPs detection by X-ray fluorescence and estimation of local dose enhancement due to AgNPs.

Silver nanoparticles within a porcine skin gelatin matrix were successfully obtained with mean sizes ranging from 2 to 20 nm. A TEM image of the synthesized NPs is shown in

Figure 1, along with the nanoparticle area equivalent diameter (AED) histogram and its fitted Lognormal distribution. The materials used in the AgNP-infused Fricke gel dosimetry study had a mean AED of 7.8 nm with a standard deviation of 3.1 nm and didn't caused any modification to the stability of the FGD or severe changes to their optical properties, as reported in previous studies when a silver salt was used as the modifier (Mattea et al., 2017)

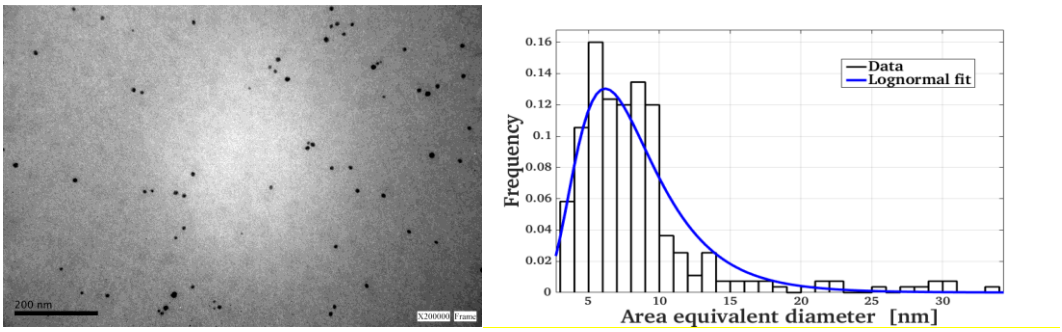

Figure 1.- TEM image of the silver nanoparticle synthesis product (left) and particle size distribution (right).

In order to quantify the dose enhancement effects due to the presence of silver nanoparticles, AgNPs-infused Fricke gel dosimeters were irradiated with X-ray beams with kVp energies below and above silver K-edge and a total absorbed dose of 12 Gy. The obtained absorption spectra are shown in Figure 2. An increase in the absorbance change was observed when the dosimeters were irradiated above silver K-edge, thus proving a dose enhancement because of their presence. For comparison purposes, absorption spectra of Fricke gel dosimeters without nanoparticles and irradiated under the same conditions are also included in Figure 2.

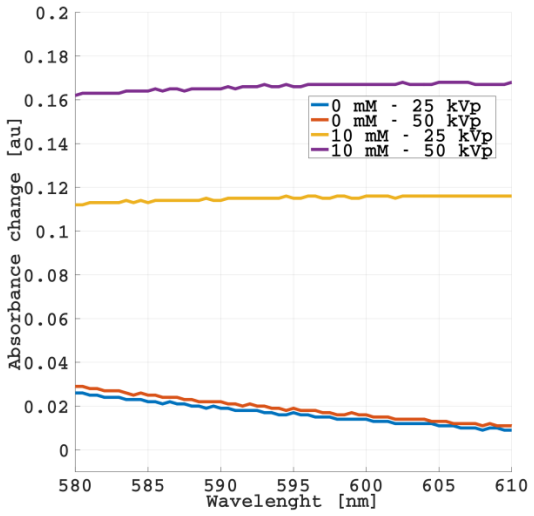

Figure 2.- Absorption spectrum for silver nanoparticles infused Fricke gel samples and standard BFGD.

From these results, it becomes clear that nanoparticles are increasing the base absorbance value in the material, but if a proper concentration of xylenol orange and nanoparticles are used a clear dose enhancement can be measured with the proposed method. Silver characteristic X-ray fluorescence was detected in a water equivalent phantom placing a nanoparticle doped volume at different depths from the surface of the phantom.

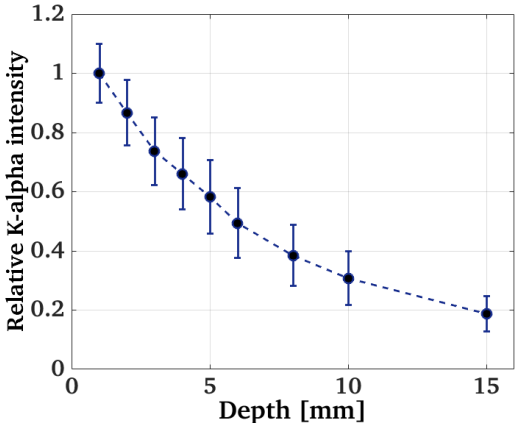

Figure 3.- Silver K-alpha intensities at different depths.

The intensity of the resulting K-alpha lines is depicted in Figure 3, normalized to the minimum experimentally achieved depth (1 mm).

Figure 4 reports results obtained from Monte Carlo simulations for the K alpha line intensity as a function of the water-equivalent depth at which AgNPs were located.

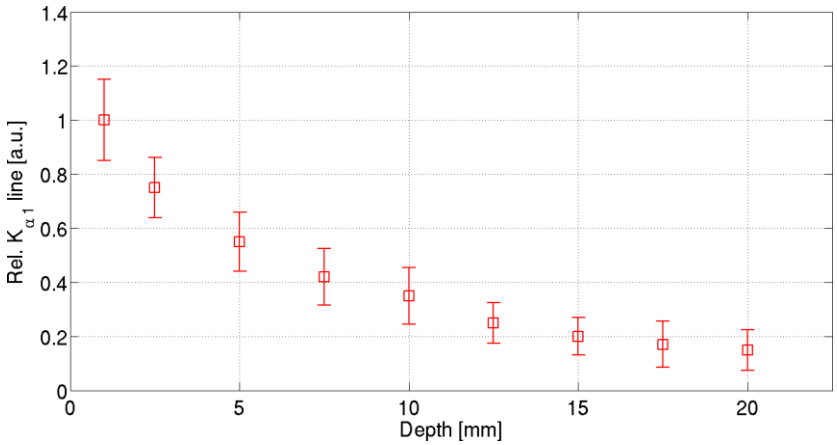

Figure 4.- Relative intensity (normalized at 1 mm depth) of detected K-alpha line positioning the 2% w/w AgNP-infused Fricke gel dosimeter at different water-equivalent depths.

Similarly, total absorbed dose was calculated for AgNP-infused samples for different AgNP concentrations, and the obtained results were normalized to 100% for the case of standard, i.e. no AgNPs, Fricke gel dosimeter. The obtained results are summarized in Table 1.

Table 1.- Dosimetry effect due to AgNP presence in Fricke gel dosimeters.

AgNP concentration [%w/w]	Norm. total dose [a.u.]
0	1.000±0.001
0.5	1.008±0.005
1.0	1.019±0.009

1.5	1.05±0.01
2.0	1.09±0.01
10.0	1.02±0.01

4.- DISCUSSION

During the first phases of this work, AgNP synthesis represented one of its main limitations and required an exhaustive study and the implementation several techniques to overcome the implications of using silver as a modifier for Fricke gel dosimetry. Also, because of the high demanding requirements in the final product, AgNPs with minimal ionic Ag concentration was mandatory and essential for the appropriate further implementation. Successive dialysis processes demonstrated to be a suitable solution without affecting significantly the properties of AgNPs, like their average size descriptors and their distribution. X-ray fluorescence was proposed for real-time external monitoring, because it is a well-known robust technique capable of being easily integrated or adapted to typical irradiation setups involving nanoparticle-infused materials. Moreover, the dose enhancement due to the presence of AgNPs was proved by measuring the difference in the absorbance of dosimeters irradiated above and below de silver K-edge.

Finally, very good agreement was found between Monte Carlo simulations and experimental data, thus supporting Monte Carlo technique as a valuable tool to model these types of physical processes involving nanoparticle suspensions. As expected, Monte Carlo was also capable of evaluating the dosimetric effects of AgNPs within aqueous systems.

5.- CONCLUSIONS

Preliminary, AgNPs and gel dosimetry were integrated by means of the AgNP-infused Fricke gel dosimeter. The dosimetry system maintained reasonable useful dose-response, comparable to that of standard gel dosimeters. Modifications proposed for adapting existing elaboration protocols for AgNP-infused Fricke gels proved to achieve good results in terms of feasibility and AgNP characterization. Experimental X-ray fluorescence demonstrated to be a robust technique for the characterization of emerging photons making possible further correlations with AgNP presence, concentration and location. Careful characterization about AgNP size distribution was achieved by means of dedicated image processing of electron microscopy. Experimental results and Monte Carlo predictions exhibit very good agreements, as reported in Figures 3 and 4, for the monitoring of emerging X-rays and its correlation with AgNP location. Additionally, dose enhancement was satisfactorily described by means of Monte Carlo simulations. These results might constitute a valuable contribution for current and further investigations and developments in the field.

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