MASS SPECTROSCOPY INVESTIGATION OF THE EFFECT OF GAMMA IRRADIATION ON THE MEAN VALUE OF THE NUMBER OF ETHOXY GROUPS IN THE TRITON X-100

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Abstract

The effect of gamma radiation from a 60Co source on the structure of a nonionic surfactant, namely TRITON X-100, was investigated. Three main regions can be distinguished in the behavior of the mean value of ethoxy groups with an increase in the absorbed dose. However just a slightly decrease on this mean value was obtained when the dose range from 0 to 70 kGy.

Key words: gamma radiation, cobalt 60, radiation doses, mass spectroscopy, sterilization, radiation dose distributions, polymers

INTRODUCTION

Studies on radiation chemistry in micellar systems is a relatively new field due to the fact that these systems can resemble biological tissues, and be therefore used in different pharmaceutical applications. A classical example of the biological importance of these systems is the study of lung surfactant solutions, which are used as a model to determine the type and extent of possible radiation effects in this organ [1,2]. Micellar systems have also being used in the encapsulation of drugs and labelled compounds, such as radiopharmaceuticals for therapy and diagnosis in medical applications. For example, injectable delivery systems, such as polymeric microspheres, and carriers used in gene therapy, such as amphiphilic aggregates (liposomes and niosomes), are intended for intravitreal administration and have to meet the pharmacopeia requirements of sterility. In the pharmaceutical field, one of the applications of ionizing radiation is the final sterilization of these biodegradable materials intended for parenteral use. A minimum absorbed dose of 25 kGy is regarded as adequate for the purpose of sterilizing pharmaceutical products without providing any biological validation [3]. Gamma-radiation as a form of electromagnetic radiation, characterized by high penetration at a very low dose rate, can modify the performance of irradiated drug delivery systems, prolonging the peroxidative radiolitic mechanism due to the exposure time. However, the degradation products generated can significantly alter the aqueous microenvironmental conditions, e.g. H+ concentration within the system.

The effects of γ-irradiation on polymer microspheres have been reported by different authors, depending on the type of polymer and the active component [4-6]. However, little information is till now available in the literature on the effect due to the use of such excipients on the biopharmaceutical performance of γ-irradiated microparticulate systems. The study of the effects of γ-irradiation on new formulations such as microspheres intended for intravitreal administration becomes necessary also because local toxicity is related to particular properties that can be affected by sterilisation.

In this work, we investigated the effect of the gamma radiation from 60Co on the mean value of the number of ethoxy groups in micellar solutions of the nonionic surfactant TRITON X-100, a very common and widely used tensoactive both in the
cosmetics and pharmaceutical Industries as
solubilizer, emulsifier and detergent. In that way,
the effect of the gamma radiation in the head of the
surfactant is assessed.

MATERIALS AND METHODS

Reagents

The nonionic surfactant used was polyoxyethylene-t-
octylphenyl ether, with an average of 9.5 oxyethylene
(EO) units per molecule (Triton X-100, figure 1), from
Rohm & Haas Co. Surfactant concentration was 1%
(wt), ca. 1.6 x 10⁻² mol dm⁻³ which is well above the
CMC of Triton X-100 (2.4 x 10⁻⁴ mol dm⁻³ [7]; the
percentage of surfactant is based on the amount of
water present (in all the experiments distilled water
was used). The surfactant was used as received.

The mixtures were prepared 24 hours in advance to
ensure full hydration of micelles. The cloud point
under these conditions was 64.5°C.

RESULTS AND DISCUSSION

Mass Spectrometry and structural changes

Commercial polyethoxylated surfactants, such as
Triton X-100, are obtained as a polymeric
distribution having the same tail structure but
different head, with a mean value of ethoxylated
(EO) groups of 9.5. To investigate the effect of the
dose in this mean value a mass spectrometry
technique was used. A typical mass spectrum of
the non irradiated surfactant solution at 1% wt
shows a distribution of peaks between 449 and
861, which corresponds to a distribution range of
EO units around 9 (figure 2).

Figure 1. Chemical structure of Triton X-100 (n = 9.5).

Gamma irradiation of the samples

Samples were irradiated in non de-aerated glass
ampoules using gamma rays from a 60Co gamma
source, at 25°C. The activity of the radiation
chamber was 2.12 kCi and the dose rate was
1.373 kGy/h, measured by Fricke and ceric sulfate
dosimeters. Small aliquots of Triton X-100
aqueous solutions (1% wt) were irradiated at
doses between 0.1 and 70 kGy.

Mass spectrometry analysis

The mass spectrometry analysis was performed in
the reflective mode of a BRUKER / BIFLEX III mass
spectrometer, equipped with a 337 nm UV nitrogen
laser (3 ns FWHM, 200 μJ mean energy per pulse)
from Laser Science Inc. Samples were analyzed
using Laser Desorption Ionization (LDI) and Matrix
Assisted Laser Desorption Ionization (MALDI). The
standard dried droplet method was used for the
sample preparation and a TX-100 typical
concentration of 10⁻⁴ w/w was used. The TX-100
molecule ionization was ensured by applying small
quantities of Na and K ions in the sample solution.
For the LDI and MALDI analyses a laser intensity of
0.75 and 0.15 GWcm⁻² was used, respectively. The
4-Hydroxy-α-cyanocinnamic acid (α-CHCA) was
used as a matrix for the MALDI analysis, with a
concentration of analyte (TX-100) to matrix (α-
CHCA) molecules of 1:10. All the mass spectra
correspond to an average of 15 and 10 laser shots
for LDI and MALDI analyses, respectively.

Figure 2. A typical mass spectrum of the non irradiated
surfactant solution at 1% wt of Triton X-100.

Calculations using a more detailed graph of this
distribution, expressed as peak area vs. number of
EO groups per monomer, gave an average
number of 9.3 (figure 3), which was considered as
the mean value and used for comparison with the
irradiated samples.

Figure 3. Mass spectrometry results for Triton X-100 micellar
solution (1%), without irradiation. ne, number of EO groups
per monomer.

Small aliquots of Triton X-100 aqueous solutions
(1% wt) were irradiated at doses between 0.1 and
100 kGy, and analysed using the mass
spectrometry technique. The mean distribution
values were calculated using the same method
and compared to the non irradiated (figure 4).
The $n_e$ values for very low doses (less than 1kGy) show an average value of 9.25 ± 0.31, and a slight decrease is observed towards a dose value of 50 kGy. For larger doses, the change in the mean values relative to the non-irradiated sample is equivalent to losing one group in the EO polymeric chain. These changes as a function of the irradiation dose are probably a consequence of the indirect action of gamma radiation on surfactant molecules, i.e. the interaction with free radical products of the water radiolysis. The direct interaction is less efficient, considering that the irradiation was performed in a dilute aqueous solution.

For the indirect interaction, there are three possible domains in the chemical structure of Triton X-100 (figure 1) that can be sensitive to these radical attacks: the hydrocarbon chain, the aromatic ring and the EO polymeric unit. However, considering that these solutions were well above CMC of surfactant, it is logical to assume that most of the primary degradation of the surfactant molecule would occur on the polyoxyethylene chain (EO), due to the shielding effect that the ethoxylated groups have over the tail [8]. A similar decrease was reported by Pellizeti et al. for the mean value of the EO groups, due to the action of hydroxyl (OH) radicals in an aqueous system of a nonionic ethoxylated surfactant (Igepal CO-720, 6.0 x 10^-4 mol L^-1, [9]). In an early report on effects of gamma irradiation upon aqueous solutions of different kinds of surfactants, it was found that bond cleavage of oxyethylene in polyoxyethylene surfactant (POE) was the main chemical reaction occurring after radiolysis of water [10].

CONCLUSIONS

Gamma-irradiation of aqueous solutions containing Triton X-100 at concentrations above CMC affects some of the surfactant structure, in particular at the dose range that is characteristic for radiation sterilisation (15-30 kGy). However, despite the slight decrease observed in the mean value of the ethoxy groups, this variation could lead to great changes in the physical chemical properties of the above mention surfactant. This fact will be study.

REFERENCES