Int. J. Chem. Sci.: 7(2), 2009, 961-968

RADIATION EFFECTS IN GELATINS

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ABSTRACT

Radiation effects in gelatins have been investigated by electron spin resonance spectroscopy. gelatins of different types have been taken and radiation effects have been compared. The ESR spectra of irradiated gelatins were quartet having complicated hyperfine structure. The spectra are thought to be superposition of component spectra arising due to •NHCO (I) and •CONH (II) radicals. Radical (I) Gives component quartet while radical (II) gives component singlet. Superposition of these two components will result in the experimentally observed spectrum. Area under ESR spectra were measured using double integration methods and the values are compared. Among the four gelatins investigated, it was observed that general grade gelatin degrades heavily than the others.

Key words : Gelatin, Irradiation effect, Electron spin resonance, Free radicals

INTRODUCTION

Gelatins are important class of biopolymers having innumerable applications like edible products pharmaceutical agents, photographic applications etc. Three types of materials essentially contribute to manufacture of gelatins[1]. They are (I) pig skin (II) beef skin and (III) bone. The gelatins essentially consist of collagen and is characterized by the bloom strength. gelatin obtained from pig skin is called as gelatin A and gelatin derived from beef skin is known as gelatin B.

Enzymatic degradation of gelatin was reported by Kamth et al.[2] These authors have also studied gamma irradiation effects in crosslinked gelatins and reported that cross linking density increases on irradiation. Thermal degradiation of gelatin is reported to

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cause a reduction in molecular weight and cross linking[3]. Under these conditions, the gelatin is reported to loose the property of water solubility[3]. Heating effects on gelatins has been investigated by Macsuga[4] using differential scanning calorimetry. These studies indicate formation of chemical bonds between gelatin molecules and formation of gels. Microbiological degradation of gelatins has been reported by various authors[5-7] who have investigated the effect of various microbiological agents on gelatins in different media.

In the present studies, the authors made an attempt to investigate radiation effects in gelatin using electron spin resonance spectroscopy. As irradiation causes cleavage of various chains and bonds in molecules, formation of free radiacals takes place. The free radicals can be identified by the ESR technique. Different types of gelatins are irradiated with gamma rays and ESR spectra are recorded. The total area under each spetrum represents various free radicals produced on irradiation. Therefore, areas under the ESR spectra have been calculated by double integration methods and the values are compared.

EXPERIMENTAL

The gelatins employed in the present studies are in powder form. They are gifted by Dr. S. Veerabhadraiah, Netherlands. These samples are exposed to gamma irradiations emitted by a cobalt 60 γ – source having a dose rate of 15 K Gy/ hour in air at room temperature. The ESR spectra have been recorded on Varian E line spectrometer operating as X-band frequencies and 100 K Hz modulation. Different grades of gelatin used in the present studies are as listed in the following Table 1.

Table 1 : Designation of gelatins

Name of the sample	Specification	Designation
Gelatin type A	162 bloom	G1
Gelatin type A	Derived from porcine skin	G2
Gelatin type B	225 bloom Bovine skin	G3
Gelatin	General	G4

RESULTS AND DISCUSSION

ESR spectra of unirradiated gelatins have not shown any signals indicating the absence of free radicals. However, irradiated gelatins have shown ESR signals, which are essentially quartets, with some minor deviations. The spectral parameters are listed in Table 2. The spectra are as shown in Fig. 1, 2, 3 and 4, respectively.

The spectral areas are calculated by double integration methods and the values are listed in Table 3. The spectral areas calculated by different methods are not same and hence, the average values are formulated in the table.

Table 2 : Spectral parameters of irradiated gelatins

Sample	No. of hyper-fine lines	Spread (G)	Line position (G)				Line spacing (G)			Descri -ption
			1	2	3	4				
G2	4	95	3219	3244	3264	3279	15	20	29	Fig. 1
G1	4	95	3223	3245	3264	3280	17	19	26	Fig. 2
G4	4	90	3216	3233	3265	3276	15	20	25	Fig. 3
G3	4	90	3215	3242	3260	3276	15	18	26	Fig. 4

Computer simulations

Irradiation of gelatins result in formation of free radicals. The observed ESR spectrum indicate the presence of free radicals. The total number of free radicals present in an irradiated polymer is proportional to the total intensity of ESR spectrum. Therefore, evaluation of total intensity of ESR spectrum is an important aspect. The authors have used double integration methods to evaluate the values of ESR intensity. The total areas can be computed by Simpson method, trapezoidal method and graphical methods. Computer program are written for these methods. The values of intensities obtained by these methods are compared.

ESR spectra of gamma irradiated gelatins are as shown in Fig. 1, 2, 3 and 4. The spectral parameters are as listed in Table 2. The values of spectral intensities obtained by different methods are not same and the average values are as listed in Table 3. Since the value of spectral intensity is proportional to the number of free radicals, the concentration of free radicals can be ascertained from spectral areas. It can be observed from Table 3, the number of free radicals produced in gelatins is in the following order.

G3 > G1 > G4 > G2

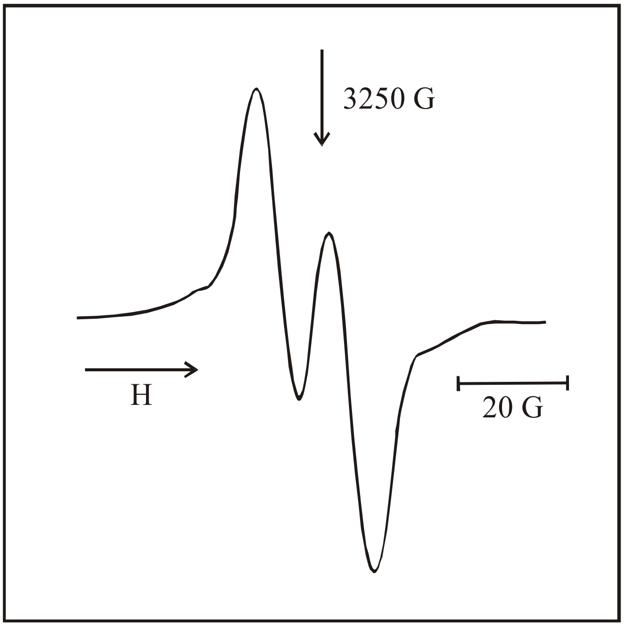


Fig. 1 : ESR Spectrum of gelatin A

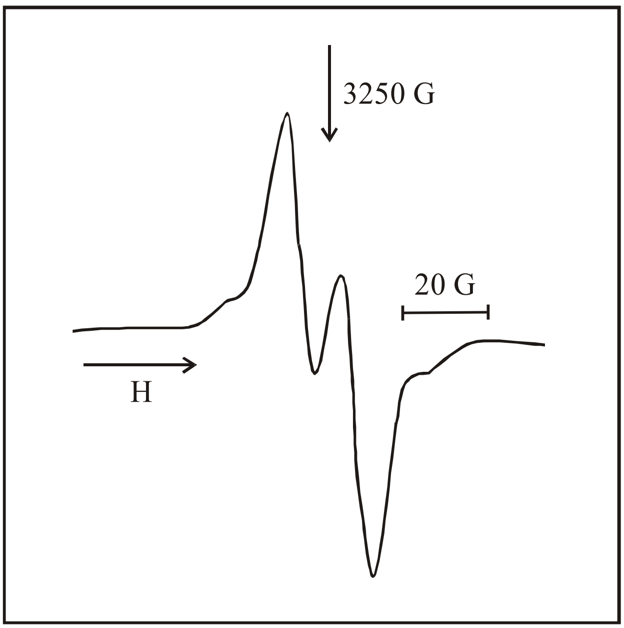


Fig. 2 : ESR Spectrum of irradiated gelatin A 162 blooms

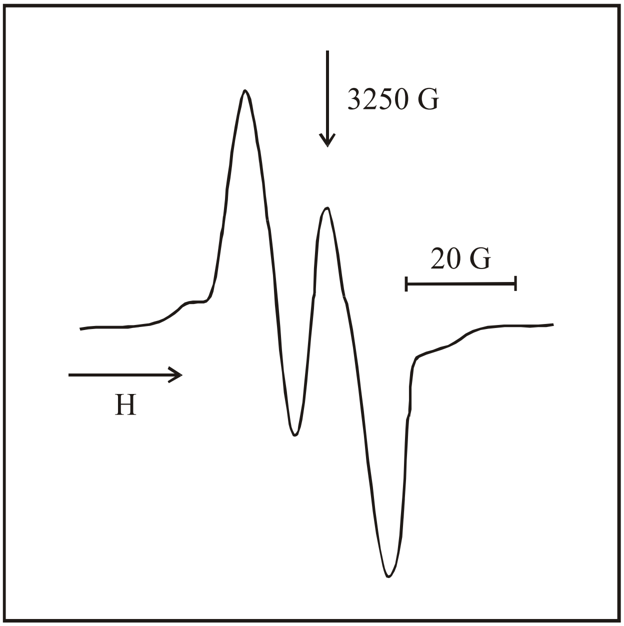


Fig. 3 : ESR Spectrum of irradiated gelatin B

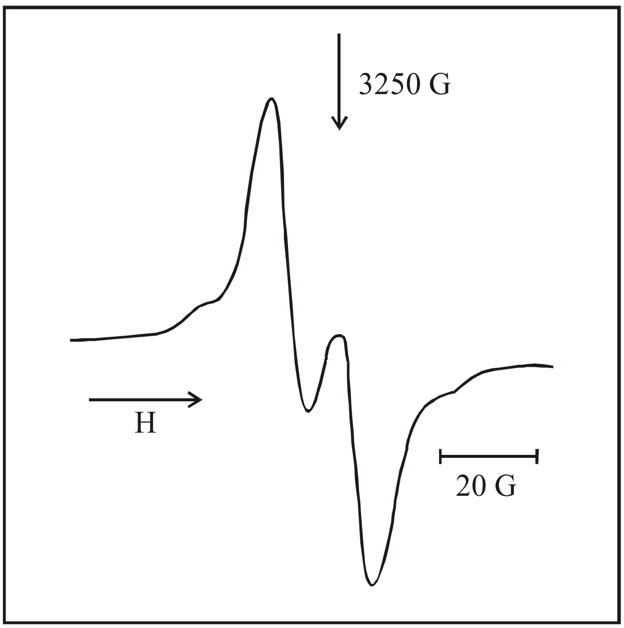


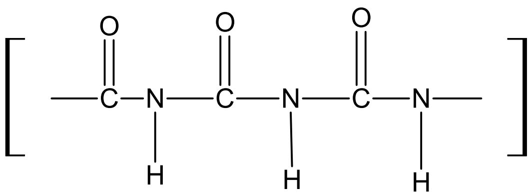
Fig. 4 : ESR Spectrum of irradiated gelatin

Table 3 : Spectral areas of irradiated Gelatins

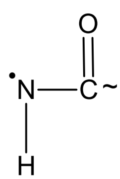
Sample	Average value of spectral intensity
G1	1812
G2	1552
G3	2310
G4	1810

Identification of radiation producers

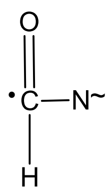
The chemical formula of gelatins is as shown below –



The radiation may attack at any one or many bonds. In view of the stability considerations, it is most probable that bond cleavage at position ‘a’ may takes place, leading to the formation of two types of radicals. They are –



Radical (I)



Radical (II)

Abraham and Whiffen[8] have reported that radicals of type (I) will give component quartet with a splitting of ~ 40 Guass. The intensity distribution quartet observed in the present studies is not equal to the quartet observed by Abraham and Whiffen. Therefore, contribution from radical (I) is also anticipated. In principle, radical (II) is not having either α or β protons in its nearest vicinity. Therefore, component singlet is more appropriate to expect from radical.

Regarding radical (I), it has nitrogen atom coupled to the unpaired electron in α–position. As the nuclear spin of nitrogen is ‘1’, it should give three lines. There is a proton coupled to the α – nitrogen. The proton contributes to two hyperfine lines. The total number of hyperfine lines expected from radical (I) may be six (3 x 2 = 6). However, only quartet spectrum is observed experimentally.

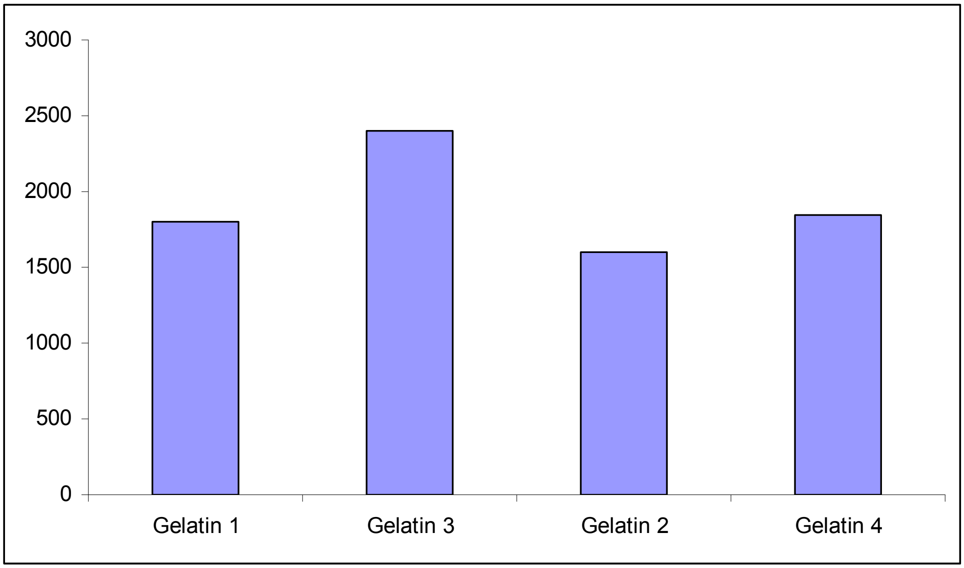


Fig. 5 : Comparison of spectral areas of gelatins

The reason for the decrease in number of hyperfine lines is attributed to the interplay of hyperfine interaction. Such type of hyperfine interaction has also been reported by Sanjeeva Rao et al.[9, 10] for irradiated poly (methacrylic acid) and irradiated poly (phenyl methacrylate). The areas under ESR spectra of irradiated gelatins have been calculated by different double integration methods. The values obtained by these methods compared by plotting histograms are depicted in Fig. 5. It indicates that the number of free radicals in G3 is greater than other types of gelatins.

CONCLUSION

In conclusion, irradiation of gelatins leads to the cleavage of C-N bond, forming radicals (I) and (II). Radical (I) gives component quartet while radical (II) gives component singlet. Superposition of these two components results in the observed quartet spectrum. Among the four gelatins studied, G3 degrades heavily than the other types gelatins.

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Accepted : 22.03.2009