3.1 INTRODUCTION

MPG has been shown to afford radioprotection in mice *in vivo* and in several *in vitro* studies as well as in limited clinical trials (Nawalkha *et al*., 1984). Of several radioprotective drugs that has been used in radiotherapy of cancers to diminish the damaging effects of radiation to healthy tissues, MPG has come out to be a promising radioprotector and detoxifying agent (Sugahara and Srivastava, 1976; Nawalakha *et al*., 1984). Several clinical trials have been conducted and MPG has been accepted as a moderate radioprotector at an effective dose of 20 mg kg⁻¹ of body weight significantly below its reported toxic dose of 2100 mg kg⁻¹ body weight (Ayene and Srivastava, 1985; Uma Devi and Saharan, 1978; Saini and Uma Devi, 1979; 1980; Sugahara and Srivastava, 1976; Kumar *et al*., 1985; Sharan *et al*., 1992; 1995). Of the two related drugs, MPG and WR-2721, the latter has been reported to be more effective radioprotector than MPG for bone marrow (Gupta and Uma Devi, 1985) but its toxicity is too high to use it in clinical radiotherapy (Meistrich *et al*., 1984).

In conventional protocols, 20 mg kg⁻¹ dose of MPG is administered, either through oral or intraperitoneal route, 30 min prior to irradiation because its effectiveness depends not only on the dose of MPG but also on the interval between drug administration and irradiation (Bisth *et al*., 1990). As discussed in chapter 2, the main reason for the effectivity of MPG is its nontoxic nature at its optimum effective dose for radioprotection (20 mg kg⁻¹ of body weight) approximately 30 min before irradiation (Sugahara *et al*., 1970; Nagata *et al*., 1972; Tanaka, 1972; Nawalkha *et al*., 1984). Perhaps this time interval between administration of MPG and irradiation is required to permit metabolic accumulation of MPG in its optimum concentration in the tissues that are envisaged to be protected. However, it must be noted that once administered, the distribution and metabolic fate of MPG is totally physiology dependent. Therefore, accumulation of MPG in different tissues and metabolic alterations of MPG cannot be controlled. This fact suggests that depending on the physiological state of the subject, radioprotective effect of MPG may show variations. This is a serious matter which ought to be considered when MPG is to be administered to humans in chemo-radiotherapy protocols.

There have been reports where the radioprotective effect of MPG was reversed and it behaved as a radiosensitizer. MPG was found to behave like a radiosensitizer against spontaneous and chemically induced microsomal lipid peroxidation *in vitro* (Cheeseman *et al*., 1981; Ayene and Srivastava, 1985). It has been proposed that MPG may be at the level of secondary radical scavenger or chain breaking antioxidant to influence lipid peroxidation. It was, nonetheless, shown that MPG affordable radioprotection was restored in the presence of EDTA (Cheeseman *et al*., 1981). Similarly, inactivation of enzyme catalase *in vitro* in the presence of MPG showed both radioprotective and radiosensitizing effects of MPG under different situations. The MPG affordable radioprotection was reversed to radiosensitization when Fe²⁺/Fe³⁺ ions
were allowed to interact with MPG resulting in formation of an unstable catalase Fe\textsuperscript{2+}/Fe\textsuperscript{3+} complex (Wary and Sharan, 1988). MPG also induced radiosensitization at higher doses of gamma rays in human lymphocytes in vitro when assayed for radiation induced DNA strand break (Wary et al., 1989; Sharan, 1990). The possible reasons for reversal of MPG affordable radioprotection are:

(a) Alteration of the chemical structure or characteristics of MPG after its administration losing its radioprotective form.

(b) Sub-optimal concentration of MPG in the tissue envisaged to be protected.

(c) Circumstantial interaction of MPG with other macromolecules.

Further, due to physiologically dependent tissue distribution of MPG, it is possible that undesirable tissues (like tumors) show radioprotection thereby the desired effect of radiotherapy. All these have seriously limited use of MPG in clinical chemo-radiotherapy.

In order to maintain MPG in its radioprotective form it is, therefore, important that the chemical structure and characteristics are not metabolically altered in the 30 min prior to irradiation period to keep MPG in its radioprotective form. Secondly, the effective concentration of MPG in the tissues envisaged to be protected should be optimum for radioprotection and its concentration be minimum or totally nil in tissues (such as, tumors) that are not envisaged to be protected. Thirdly, circumstantial interactions of MPG with cofactors or ions, known to effect affordable radioprotection (Ayene and Srivastava, 1985; Wary and Sharan, 1988; Wary et al., 1989; Sharan, 1990) should be avoided as much as practicable. In principle, MPG after liposome encapsulation gets endowed with characteristics that may satisfy all the above three points. Due to liposome encapsulation, the MPG molecules are not immediately accessible to metabolic enzymes and co-factors that may alter the chemical structure and characteristics of MPG. Therefore, alterations of the chemical characteristics of MPG after administration is slowed down. For the same reasons, circumstantial interaction of MPG with other macromolecules is seriously hindered. Finally, as liposomes may be potentially targeted to tissues of interest, it will be possible to ensure optimum MPG concentration for radioprotection in tissues of choice and limited or no MPG in other tissues that are not envisaged to be protected.

As it has been reported in chapter 2, encapsulation of MPG by reverse-phase evaporation method was very satisfactory, this chapter deals with testing the biological effectiveness of MPG as a radioprotector after liposome encapsulation. To quantify the MPG affordable radioprotection in biological system two parameters are envisaged to be used:

(a) Protection of γ-irradiation induced cell killing by assaying viability of cells.

(b) Protection of γ-irradiation induced release of membrane bound acetylcholine esterase
of liver by assaying the amount of the enzyme in the soluble fraction.

Over all viability of cells after radiation assault is of utmost importance for assessment of impact of radiation. This strategy has been employed in several studies as the most important parameter of assessment of radiation induced damage as well as for monitoring the impact of factors which can rescue cells from radiation induced damage. Since membrane constitutes the major component of a cell, dye exclusion technique to monitor viability of cell has been very widely used (Wary and Sharan, 1988; Sharan et al., 1995).

Acetylcholine esterase (acetylcholine acetylhydrolase, AChE), an oligomeric enzyme, is predominantly membrane anchored (Massoulié and Toutant, 1988) mediating cholinergic neurotransmission by rapid hydrolysis of transmitter acetylcholine. Radiations are known to disrupt membrane structure, thereby, releasing the membrane bound or anchored enzyme (Bacq and Alexander, 1966). Thus, accumulation of AChE in the soluble fraction of liver has been used as a parameter to monitor radiation induced damage (Sharan et al., 1995). The cellular substrate of AChE is acetylcholine that is hydrolyzed by the enzyme to choline (Jürss and Maclicke, 1981; Bazelyansky et al., 1986).

3.1.1 Aim And Objectives
In order to achieve the objective, the investigation aimed to do the following:

1. To assess the effect of increasing dose of γ irradiation on the survival of bone marrow and spleen cells of mice.

2. To monitor the effect of increasing dose of γ irradiation on the release of membrane bound acetylcholine esterase (AChE) enzyme of liver of mice.

3. To monitor the two parameters mentioned above when the mice had been administered bioequivalent dose of either free MPG or LEM prior to irradiation in order to establish the biological effectiveness of MPG after liposome encapsulation.

3.2 METHODS AND MATERIALS

3.2.1 Chemicals
2-mercaptopropionylglycine (MPG) was obtained from Santen Pharmaceuticals Co., Japan. Dipalmytoyl phosphatidyl choline (DPPC); Dicetylphosphate (DCP); Cholesterol (Chol), Triton X-100, Trypan blue and Acetylcholine were purchased from Sigma Chemical Co. USA;
Dithionitrobenzoic acid (DTNB) from SRL, India and Sepharose CL-4B from Pharmacia Fine Chemicals, Sweden. Minimum essential medium (MEM) was product of from HiMedia, India. Other chemicals of highest purity grade were purchase from local suppliers. Glass double distilled water was used for all preparations.

3.2.2 Animal
Female Swiss albino mice (6-8 weeks), inbred colony maintained in animal room (22±2 °C) on standard dry pellet feed and water ad libitum, were used in this study. The average weight of the mice was 20±3 g.

3.2.3 Experimental Groups
The animals were divided into the following groups for this investigation:

1. Control group - This consisted of normal mice without the injection of MPG or LEM. The animals were not irradiated. 15 mice was used for each group.

2. Positive control group - A group of animals were irradiated to different dose γ rays (1 Gy, 2 Gy, 4 Gy, 6 Gy and 8 Gy).

3. MPG group - The mice belonging to this group received intraperitoneal injection of aqueous solution of MPG 30 min prior to irradiation at different doses (1 Gy, 2 Gy, 4 Gy, 6 Gy and 8 Gy). Three different doses of MPG used were: 10 mg kg⁻¹ body weight, 20 mg kg⁻¹ body weight and 40 mg kg⁻¹ body weight.

4. LEM group - The mice belonging to this group received intraperitoneal injection of liposome encapsulated MPG (LEM) 30 min prior to irradiation at different doses (1 Gy, 2 Gy, 4 Gy, 6 Gy and 8 Gy). Three different doses of MPG used were: 10, 20 and 40 mg kg⁻¹ body weight.

3.2.4 Preparation Of Liposomes
Liposomes were prepared by reverse phase evaporation method. The details of the methodology has been described in chapter 2 (section 2.2.3). Briefly, using 5 mg of DPPC, 1 mg of DCP and 2.5 mg of cholesterol which were dissolved in chloroform:ethanol (v/v, 1:1). To the solution, an aqueous solution of MPG was slowly added while vortexing. Chloroform and ethanol were removed by rotary evaporator at 40 °C for about 45 min. LEM was separated from the free MPG by centrifugation or by gel filtration on Sepharose CL-4B. The concentration of MPG in LEM form was calculated by the assay of -SH group as described in chapter 2 (section 2.2.5).
3.2.5 Dose And Mode Of Administration Of MPG
Separate solutions of 2.5 mM, 5 mM and 10 mM MPG were prepared as described in chapter 2 (section 2.2.3.1). The same MPG solutions were used for entrapment into liposome. Half ml of the MPG solutions (free MPG) or 1 ml of LEM were intraperitoneally injected onto mice to deliver 0.204 mg, 0.408 mg and 0.816 mg, respectively, of MPG per mouse. The dose of MPG in these three cases was respectively equivalent to 10, 20 and 40 mg MPG kg\(^{-1}\) body weight of mouse. MPG was administered by a single intraperitoneal injection about 30 min prior to irradiation.

3.2.6 Dose And Mode Of γ-Irradiation
Mice were whole body irradiated to doses of 1 Gy, 2 Gy, 4 Gy, 6 Gy and 8 Gy of γ rays in a Gamma chamber 900 (Bhabha Atomic Research Center, Bombay). The source of radiation was a \(^{60}\)Co delivering radiation at a dose rate of 23.65 Gy min\(^{-1}\).

3.2.7 Assay Of Cell Viability
The method was based on monitoring the number of cells which are able to exclude trypan blue dye indicating that they are metabolically active and live (Wary and Sharan, 1988). Therefore, non-blue cells were taken as live while cells which accumulated dye and became blue were taken as dead.

3.2.7.1 Preparation of required buffers/solutions: The following reagents were prepared:

(A) The reagent for dye exclusion assay was made by boiling the following in 90 ml of water.

- Trypan blue: 400 mg
- Sodium chloride: 800 mg
- Dipotassium hydrogen phosphate: 60 mg
- Methyl p-hydroxy benzoate: 50 mg

The pH of the solution was adjusted to 7.2 with 0.1 N sodium hydroxide, the volume made up to 100 ml and the solution was stored at room temperature (Phillips, 1973).

(B) Minimum essential medium (MEM): MEM (11.7 g) was dissolved in 1000 ml of double distilled water. After autoclaving, it was stored refrigerated.

3.2.7.2 Methodology:
Immediately after irradiation, the mice were killed by cervical dislocation. The cells were flushed out of the spleen and the bone marrow and care was taken to minimize cell damage. The
spleen and the bone marrow cell suspensions were prepared in MEM. To 0.9 ml of the cell suspension 0.1 ml of the assay reagent were mixed and incubated at 37 °C for 5 min. Viable (non-blue) and dead (blue) cells were counted on a Burker chamber under a Zena phase contrast microscope. The cell number was calculated using the following formula:

\[
\text{# of cells ml}^{-1} = \text{# of cells square}^{-1} \times 25000 \times \text{dilution factor}
\]

### 3.2.8 Calculation Of Viability Modification Factor (VMF)

To quantify the radioprotective effect of MPG or LEM the VMF was calculated. It is a factor derived by dividing the % viability of the cells after irradiation in the presence of either free MPG or LEM by that of radiation alone.

\[
\frac{\text{% viability of cells in the presence of either free MPG or LEM after } X \text{ Gy } \gamma\text{-irradiation}}{\text{% viability of cells after } X \text{ Gy } \gamma\text{-irradiation}}
\]

### 3.2.9 Assay Of Acetylcholinesterase (AChE)

The enzyme assay methodology was based on Ott et al. (1975) with minor modifications.

#### 3.2.9.1 Preparation of required buffers/solutions:

The following reagents were prepared and stored refrigerated for use:

(A) PBS (pH 7.9) - The constituents of this buffer was:

<table>
<thead>
<tr>
<th>Substance</th>
<th>Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Na₂HPO₄</td>
<td>0.1 M</td>
</tr>
<tr>
<td>NaH₂PO₄</td>
<td>0.1 M</td>
</tr>
<tr>
<td>NaCl</td>
<td>0.9 M</td>
</tr>
</tbody>
</table>

appropriate amounts of Na₂HPO₄ and NaH₂PO₄ were separately dissolved in water to prepare their respective 0.1 M solutions. The two solutions were mixed until pH of 7.9 was obtained. To this calculated amount of NaCl was added to get the desired molarity.

(B) Sucrose solutions: To prepare a 0.2 M solution 68.460 g of sucrose was dissolved in 1000 ml of double distilled water.

(C) The AChE assay mixture in 3 ml contained the following:

<table>
<thead>
<tr>
<th>Substance</th>
<th>Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acetylcholine</td>
<td>1 mM</td>
</tr>
<tr>
<td>DTNB</td>
<td>0.125 mM</td>
</tr>
<tr>
<td>Triton-X-100</td>
<td>0.05 %</td>
</tr>
<tr>
<td>Enzyme preparation</td>
<td>0.1 ml.</td>
</tr>
</tbody>
</table>
3.2.9.2 **Methodology:** Immediately after irradiation the animals killed by the cervical dislocation and livers removed in 0.2 M sucrose solution. The livers homogenized in PBS (pH 7.9) using a motorized tissue homogenizer. The homogenate centrifuged at 2000 \( x \) g for 30 min at 4 °C using a Heraeus RS 20 centrifuge. The resulting supernatant (enzyme preparation) subjected to the assay of AChE.

The assay mixture containing the enzyme preparation in a cuvette placed in a Shimadzu UV 150-02 spectrophotometer. The reaction followed by observing an increase in absorption (412 nm) at intervals of 30 sec for 3 min and the increase in absorption \( \text{min}^{-1} \) calculated. One unit activity of enzyme is defined as the amount of enzyme which consumes 1 M of the substrate at room temperature \( \text{min}^{-1} \).

3.2.10 **Data And Statistical Treatment**

The data presented in this thesis are mean±SD or SEM representing minimum of six to nine independent sets each consisting of at least three mice. In a few experiments, only one mouse constituted an independent set. Such experiments were independently repeated nine times. The controls were age-matched mice, sham-injected with PBS 30 min before irradiation. Student’s \( t \)-test was applied to calculate the significance of differences. All values with \( p \geq 0.01 \) have been taken as significant.

3.3 **RESULTS**

Administration of 2.5 mM, 5 mM or 10 mM MPG is equivalent to 10 mg, 20 mg or 40 mg kg\(^{-1}\) body weight, respectively, to each mouse. The doses of whole body \( \gamma \)-irradiation to mice were 1, 2, 4, 6 and 8 Gy. In all the cases, the MPG, either as free MPG or in liposome encapsulated MPG (LEM), was intraperitoneally administered to mice approximately 30 min before irradiation.

3.3.1 **Effect Of \( \gamma \)-Irradiation On Survival Of Spleen Cells**

Fig. 3-1 shows that the decrease of survival of the spleen cells was radiation dose dependent in the absence of MPG (curve i). The presence of free MPG significantly enhanced the percentage of surviving cells in all the cases (curve ii). The LEM further enhanced the survival of the cells at all doses (curve iii) and exhibited reversal of the trend for the dose of 20 mg Kg\(^{-1}\) body weight (panel B). For this dose of LEM, the relative protection afforded by MPG was highest at 8 Gy of \( \gamma \) irradiation.
3.3.2 Effect Of $\gamma$-Irradiation On Survival Of Bone Marrow Cells

Fig. 3-2 shows that the decrease of survival of the bone marrow cells was radiation dose dependent (curve i). The presence of free MPG significantly enhanced the percentage of surviving cells in all the cases (curve ii). Like spleen cells, the LEM protected BMC from radiation induced killing at all doses (curve iii). It again exhibited reversal of radiation dose dependent cell killing at the dose of 20 mg kg$^{-1}$ body weight of MPG (panel B). These results also show that the relative protection afforded by LEM at the dose of 20 mg kg$^{-1}$ body weight was optimum at 8 Gy.
FIGURE 3-2: Percentage survival of bone marrow cells following γ-irradiation at different doses (curve i) and that after administration of MPG (curve ii) and LEM (curve iii) 30 min prior to irradiation. Dose of MPG = 10 (panel A), 20 (panel B) and 40 mg kg⁻¹ body weight. No bar means the SEM was smaller than the thickness of the point.

3.3.3 Viability Modification Factor Of Free MPG or LEM

Fig 3-3 shows the viability modification factor (VMF) of MPG, either as free MPG or LEM, for spleen cells after 1 Gy, 2 Gy, 4 Gy, 6 Gy and 8 Gy of γ-irradiation.

The VMF afforded by free MPG (curve i) administered at the dose of 10 mg kg⁻¹ body weight (panel A) was essentially invariant for all doses of radiation. When the same dose was administered as LEM (curve ii), the initial high protection at 2 Gy leveled off at higher doses (curve ii, panel A). The situation was not very different when either free MPG (curve i) or LEM (curve ii) was administered at the dose of 40 mg kg⁻¹ body weight (panel C). However, free MPG at the dose of 20 mg kg⁻¹ body weight exhibited a clear and statistically significant radiation dose dependent increase in VMF (panel B). The VMF was highest for LEM at this dose (panel B).
FIGURE 3-3: VMF calculated for spleen cells following different doses of γ-irradiation to mice after administration of free MPG (curve i) or LEM (curve ii) 30 min prior to irradiation. Doses of MPG = 10 (panel A), 20 (panel B) and 40 (panel C) mg kg⁻¹ body weight. No bar means the SEM was smaller than the thickness of the point.

Fig 3-4 depicts the VMF afforded by free MPG (curve i) or LEM (curve ii) for BMC at the dose of 10 (panel A), 20 (panel B) and 40 mg kg⁻¹ body weight. (panel C) of MPG or LEM 30 min prior to irradiation. While the dose of 10 mg kg⁻¹ body weight did not show clear radioprotective gains for the range of radiation dose used (panel A), both 20 mg kg⁻¹ body weight (panel B) and 40 mg kg⁻¹ body weight (panel C) groups of mice exhibited significant increases in VMF after administration of MPG by either way. The affordable radioprotection, especially for the LEM treated group was very clear and statistically highly significant for its dose of 20 mg kg⁻¹ body weight (panel B).
3.3.4 Effect Of γ- Irradiation Or Release Of Liver AChE Into Supernatant

The radiation induced damage to liver was monitored by observing the activity of membrane bound acetylcholine activity in the supernatant fraction at different doses γ radiation either in the presence or absence free MPG or LEM at the dose of either 10, 20 or 40 mg kg⁻¹ body weight. Fig. 3-5 shows the results obtained from mice treated with free MPG or LEM at 10 mg kg⁻¹ body weight 30 min prior to irradiation. Except for the radiation dose of 2 Gy, all other doses showed that LEM prevented the release of AChE into supernatant. When 20 mg kg⁻¹ body weight dose of MPG or LEM was administered prior to irradiation (Fig. 3-6), both MPG and LEM offered radioprotection was statistically similar to the possible exception of that at 1 Gy. As depicted in Fig. 3-7, the dose of 40 mg kg⁻¹ body weight of MPG, either in free form or as LEM, offered no radioprotection as more AChE was assayed in the supernatant fraction.
FIGURE 3-5: Liver AChE activity in the supernatant fraction following whole body \( \gamma \)-irradiation of mice at different doses (filled) and that after administration of 10 mg kg\(^{-1}\) body weight of MPG (slashed) or LEM (dotted) 30 min prior to irradiation. No bar means the SEM was too small to be visible on this scale.

FIGURE 3-6: Liver AChE activity in the supernatant fraction following whole body \( \gamma \)-irradiation of mice at different doses (filled) and that after administration of 20 mg kg\(^{-1}\) body weight of NPG (slashed) or LEM (dotted) 30 min prior to irradiation. No bar means the SEM was too small to be visible on this scale.
3.4 DISCUSSION

The biodegradable nature of liposome and its capability of encapsulating a target molecule (Gregoriadis, 1988; Allen and Cleland, 1980) are the main reasons for the selection of liposome as a drug delivery system. Reverse phase evaporation method was selected for the encapsulation of MPG into liposomes as the method is simple and reproducible with high entrapment efficiency (Sharan et al., 1992). This method produces large unilamellar vesicles that are suitable as a carrier for MPG. The liposome preparation had the molar ratio of DPPC:Chol:DCP as 1:0.25:0.9 and the lipid/MPG ratio as 5.21. The percentage entrapment of MPG was found to be 54.54±4.95 when 5 mM MPG solution was used (chapter 2). Administration of 0.5 ml of 5 mM MPG aqueous solution or 1 ml of LEM, therefore, delivered 0.408 mg MPG equivalent to each mouse. This is quantity of MPG per mouse shall deliver a dose of 20 mg MPG kg⁻¹ body weight that is the effective radioprotective dose of MPG (Ayene and Srivastava, 1985; Saini and Uma Devi, 1979, 1980; Sugahara and Srivastava, 1976;
Upon administration free MPG migrates to different tissues including the reticuloendothelial system i.e. the spleen cells, bone marrow cells and the liver (Chiba, 1973; Toshioka et al., 1970; Carlsson et al., 1990). MPG molecules are subjected to metabolic alterations depending on the physiological state of the subject. The present finding on BMC shows the protection of pronormoblasts and normoblasts by MPG (Saini and Uma Devi, 1980). The behavior of LEM differs from that of MPG as liposome preferentially migrates to the liver, spleen cells, and bone marrow cells that is rich in reticuloendothelial cells and fenestrated capillaries (Gregoriudis, 1988). At 5 mM there is an increase in protection with the increase in radiation up to 8 Gy. At 10 mM the protection by MPG and LEM remains to almost the same whereas for spleen at 8 Gy the protection by LEM drops below that of MPG. In each case LEM shows a higher viability over the respective free MPG. The encapsulation of the drug in the liposomes delays the metabolic alteration of the entrapped drug (Papahadjopoulos et al., 1991) and so LEM offers a significantly higher radioprotection than that of free MPG to spleen cells and bone marrow cells.

**Acetylcholine esterase activity on liver** shows a supporting trend with LEM. Spleen cells and bone marrow cells show a higher viability with increasing doses of radiation in LEM treated groups but not so in the cases of free MPG. Since free MPG and LEM were injected in the mice 30 min before radiation, a part of free MPG could have been metabolically altered (Chiba, 1973; Toshioka et al., 1970; Carlsson et al., 1990) making it a less effective radioprotector. The release of the enzyme acetylcholine into supernatant signifying the radiation induced membrane damage was significantly reduced by MPG and LEM shows a higher tendency of protection. The dose of MPG was far below the toxic dose and so it is assumed that the drug did not involve in the metabolism or the survival of the treated animals. LEM as compared to free MPG afforded greater protection for all doses of radiation statistically highly significant (p≤0.001) in the case of BMC and at lower levels of statistically probability (0.12p≥0.0001) in the case of spleen cells (Sharan et al., 1995).

On the other hand metabolic alteration of MPG is reduced to the minimum level in the case of LEM and so it remains in its protective form. The encapsulation of MPG also prevents the circumstantial interaction of MPG with Fe+2 which causes radiosensitization effects (Ayene and Srivastava, 1985; Wary and Sharan, 1988). It is possible to assume that MPG has leaked out at higher doses of radiosensitization into the cell membranes and thus a higher protection. Undesirable radiosensitization by liposome encapsulation is prevented and a higher radioprotective effect of MPG in the experimental conditions. In an attempt to reasons for it Gabizon have reported that liposome encapsulated DOX, an anti tumor drug, to be more effective in compared to free form. On liposome encapsulation efficiency of radioprotection is
enhanced of the drug. Liposome can successfully be a vehicle for other drugs in radiotherapy whose use is limited to high toxicity and metabolism problems.

3.5 REFERENCES


