examination of table 1 and table 2 reveals that the present method is very accurate and precise, when compared with the $KBrO_3$ method. Tablet excipients such as starch, talc, magnesium stearate etc. did not interfere in the determinations.

Owing to the simplicity, non-requirement of pH control and ability to use aqueous solutions in the determinations combined with high accuracy and precision, the present method appears to be better than the methods reported in the literature.

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Biodegradable Microspheres of Gentamicin Sulphate

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Gentamicin sulphate loaded albumin, chitosan and poly(dl-lactide- co-glycolide) microspheres were prepared. The *in vitro* dissolution studies showed that the release could be controlled for 2 weeks by the vial method. The stability of the drug was better by encapsulation. The nasal absorption of the drug from these microspheres was about 60 percent.

IN the recent years, extensive efforts are being made in various research laboratories for the development of novel and targeted drug delivery systems. The advantages of these newer systems include patient compliance, reduction in dose and frequency of dosage and reduction of first pass metabolism. Gentamicin sulphate (GS) is an aminoglycoside antibiotic. The drug profile is well

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Fig. 1: In vitro release of Gentamycin sulphate from microspheres

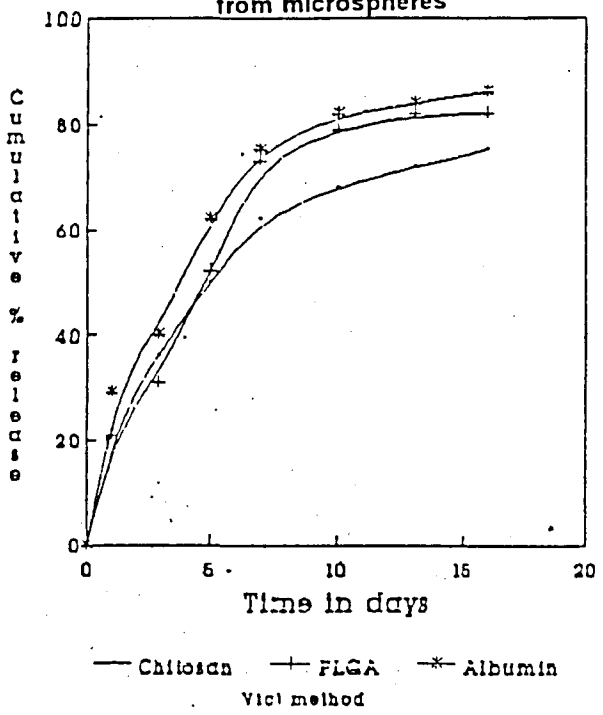
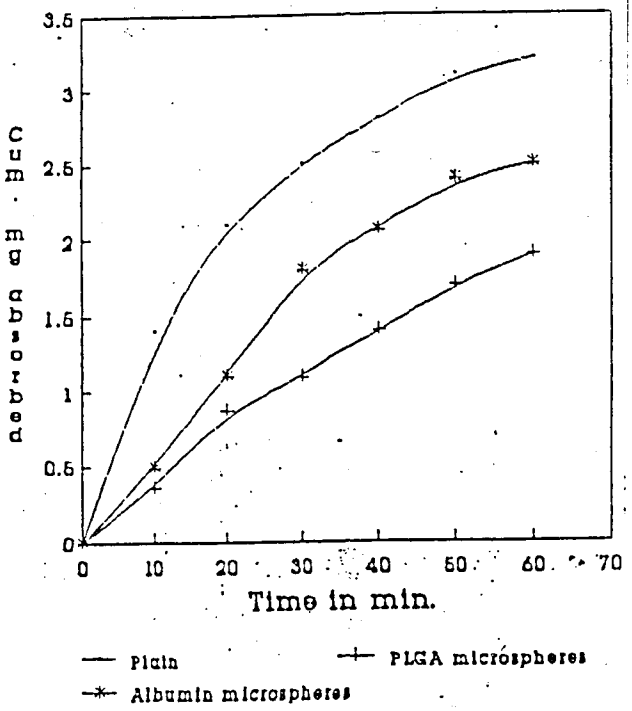


Fig. 2: Nasal absorption profile



reviewed by Gillman et al[1]. The biodegradable polymers have potential for long term unattended therapy and site specific drug delivery without the need for device retrieval. Extensive work on microspheres using chitosan[2,3], albumin[4,5] and poly (dl lactide-co-glycolide)[6,7,8] (PLGA) have been carried out. Extensive work on nasal absorption using various drug delivery systems have been performed[9-12].

The aim of the present study was to prepare microspheres of GS using biodegradable polymers, to study the *in vitro* release profile and the *in situ* nasal absorption of the drug.

Poly(dl-lactide-co-glycolide) (PLGA) and chicken albumin were procured from Polysciences, USA. Chitosan was a gift sample from Fisheries research institute, Trivandrum. Gentamicin sulphate was a gift sample from Biovaccines Pvt. Ltd., Hyderabad.

Chitosan microspheres were prepared by dissolving chitosan (2%) in glacial acetic acid. Drug (100mg) was added to the chitosan solution and sonicated (20 KHz, 100 W for 30 sec). The contents were added dropwise into 25 ml of 1N sodium hydroxide solution containing gluteraldehyde (1 ml) (crosslinking agent). The microspheres formed were collected by filtration and dried at room temperature[13].

PLGA (200mg) was dissolved in 10 ml of acetonitrile. Fifty mg of the drug was added to the polymer solution. The contents were added slowly into a beaker containing 50 g of sesame oil and stirred at 1200 rpm till the organic phase evaporated. The microspheres were collected by filtration and washed with n- hexane and dried at room temperature[14].

2.5 g of albumin and 625 mg of the drug were dissolved in 25 ml of water. The contents were slowly added to a beaker containing 65 ml of sesame oil and stirred at 1200 rpm for 1 h. The temperature was raised to 40° for hardening process and was maintained for 25 minutes followed by slow cooling to room temperature. The microspheres were collected by filtration and washed with n-hexane and dried at room temperature[15].

The *in vitro* release studies were performed in triplicate by the vial method[14]. Microspheres containing known quantity of drug were placed in vials containing phosphate buffered saline pH 7.4 (PBS).

Table 1: Degradation rate constant data

Formulations	Days^{-1} Room Temp.	37°C	82% RH
Plain GS	0.0318	0.0343	0.0330
Chitosan. MIC.	0.024	0.0212	0.0302
PLGA. MIC.	0.0250	0.0221	0.0276
Albumin. MIC.	0.0244	0.0187	0.0255

At predetermined time intervals aliquots were withdrawn and replaced with the same amount. The drug was analysed spectrophotometrically at 568 nm after addition of ninhydrin and pyridine[16].

The formulations were evaluated for stability by exposing them to room temperature, 37° and 82% humidity at 25°.

Albino rats were anaesthized and the trachea exposed and cannulated with poly ethlyene tubing. Then the oesophagus was cannulated with second tubing towards the postering part of the nasal cavity. Perfusate (1 %, 50 ml), plain gentamicin sulphate dissolved in PBS or microspheres equivalent to 1% drug dispersed in PBS) at a flow rate of 2 ml/min was introduced using a volumetric infusion pump through this tubing after nasopalantine was sealed with an adhesive agent and collected through a funnel into a reservior for recirculation[17]. At predetermined time intervals the samples of the perfusate were withdrawn and replaced with fresh phosphate buffered saline pH 7.4.

All the three formulations were spherical without the evidence of aggregation. Drug entraped in chitosan, PLGA and albumin microspheres were 76, 54.4 and 43.4% respectively. The PLGA microspheres had a size range of 5 to 17 microns, albumin microspheres, 8 to 26 microns and chitosan microspheres, 975 to 1095 microns. One of the factors influencing the size of the chitosan microsphere is the molecular weight of the chitosan and the degree of deacetylation[18]. The molecular weight of the chitosan was ca 250 000, degree of deacetylation was ca 80%. The higher molecular weight and degree of deacetylation of the chitosan polymer used in the study may have resulted in larger microspheres. Bigger chitosan microsphere size has also been reported by many authors[13,18,19] using chitosan of high molecular weights. Two percent chitosan solution was used because at lower concentrations microspheres were not obtained and at higher concentrations the dropping process became difficult. The microspheres were prepared by dropping the chitosan solution containing the drug in sodium hydroxide. The interaction of positively charged chitosan molecules with the anionic counterion caused the formation of gelled beads[19].

For PLGA microspheres, oil phase system was used because gentamicin sulphate is water soluble and the use of aqueous phase would result in the diffusion of the drug from the organic phase to the outer aqueous phase which may reduce the entrapment. For water soluble drugs, oil phase system result in better entrapment of the drug in the microspheres[20].

The *in vitro* release profile is shown in Fig. 1. Burst effect was observed which may be due to the drug adsorbed on the surface of the microspheres. The release could be controlled for a period of 2 weeks. The mechanism of drug release from PLGA

microspheres may be explained on the basis of medium entering the polymer and forming pores and channels through which drug diffusion occurs. The increase in drug release with time caused by degradation and medium uptake compensated for the decrease in release caused by the increase in the distance with time which the drug has to travel to be liberated and so an essentially constant rate of drug release as exemplified by the steady state release region. Similar results have also been reported by other investigators[21-23]. The release from chitosan and albumin microspheres can also be explained on similar basis, i.e. the channels and pores being formed through which the drug release occurs[11,16]. The release of drug from these microspheres is influenced by factors like drug loading, amount of crosslinking agent and time for hadening process[13,15,18,19].

The degradation rate constants are shown in table 1. The drug was more stable in the formulations. This may be due to the polymer coating affording some degree of protection to the drug which is entraped inside[24]. In *in situ* nasal absorption showed that the amount of drug absorbed through the nasal route was 65.2, 57.8 and 60.6 percent for plain GS, GS entraped PLGA and albumin microspheres. Since the chitosan microsphere size are very big, they were not evaluated for nasal studies.

Gentamicin sulphate microspheres were prepared with chitosan, PLGA and albumin polymers. The release studies showed that they have the potential for controlling the drug release. The drug in the microspheres was more stable and the nasal absorption was satisfactory.

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Development and Evaluation of Lipospheres of Diclofenac Sodium

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Lipospheres of diclofenac sodium were prepared by melt dispersion technique using triple pressed stearic acid. Free flowing lipospheres were obtained by congealing the microemulsion. The amount of water, Tween 20 (surfactant) and butyl alcohol (co- surfactant) were identified as the key variables affecting the formation of discrete spherical lipospheres. More than 70% of the isolated lipospheres were of the size range 180-250 μ. The amount of drug entrapped in the lipospheres was found to be dependent on the lipid to drug ratio and the drug loading was further increased by using caranuba wax coated particles of diclofenac sodium. The *in vitro* drug release study was conducted in phosphate buffer (pH 7.2). Dissolution of the entrapped drug was greatly retarded. The results of the F-statistics revealed that the drug was released by anomalous diffusion.

DICLOFENAC sodium is frequently prescribed for the treatment of rheumatoid arthritis, osteoarthritis and ankylosing spondylitis. Its biological half-life has been reported as 1-2 h. Gastrointestinal side effects such as bleeding, ulceration or perforation of intestinal wall are commonly seen[1]. Thus, a controlled release dosage form of diclofenac sodium is required to be formulated to minimize the damage to the gastro-intestinal mucosa and to reduce the frequency of dosing.

A survey of the lipid materials contained in drug products marketed in the United States showed that stearic acid or its salts are widely used in dosage forms[2]. The low cost, low toxicity and ease of fabrication have been stated as the major advantages of lipids by Kabwvichii[3]. Studies have been reported on the formulation development and dissolution testing of wax microspheres of different drugs[4-7]. The aim of the present investigation was to develop controlled release lipospheres of diclofenac sodium using melt dispersion technique, which does not require organic solvents.

Diclofenac sodium (J.P.) was received as a gift sample from Sharda Drugs. Triple pressed stearic acid, Tween 20 and butyl alcohol were purchased from local market.

Lipospheres were prepared from microemulsions as reported by Dino and co-workers[8]. The formulation of different batches is depicted in Table 1. Briefly, triple pressed stearic acid was melted on a water bath maintained at 70-72°. Finely powdered drug particles (90#) were dispersed in the molten wax. Aqueous phase was prepared by heating a blend of water and Tween 20 (surfactant, HLB 16.7) to 70-72°. Butyl alcohol (co- surfactant) was