Chapter 10

EVALUATION OF BIO-COMPATIBILITY OF THE FORMULATIONS

10.1 Introduction

The toxicity testing of any formulation developed has to be done to ensure its safety when administered in the living being. The formulation developed in these investigation, were used for intra-articular injection. Thus, it is necessary to ensure that the formulation itself is not toxic to the synovium. The evaluation of the potential toxicity of the formulation to the synovium was done by histopathology studies of the rat joint.

10.2 Experimental

10.2.1 Materials

Formaldehyde solution (37%w/w) was purchased from s.d.fine chem. limited.

10.2.2 Apparatus

Rat cages, glass syringe with 26 guage needle, glass slides.

10.2.3 Selection of animals

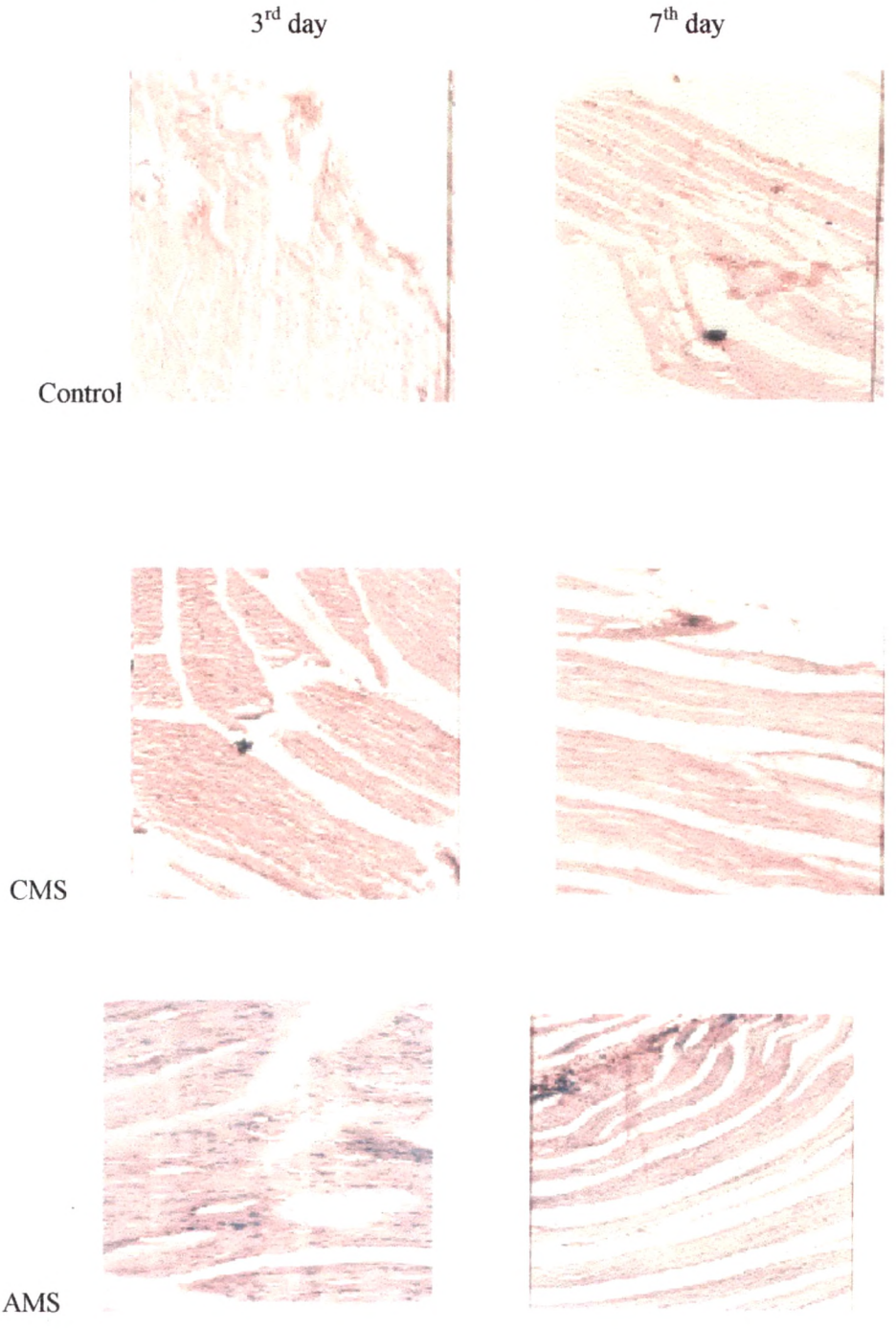
Male Sprague-Dawley rats weighing 300-350 gms were chosen for histopathology studies. No diet restriction was enforced prior to studies.

10.2.4 Histopathology

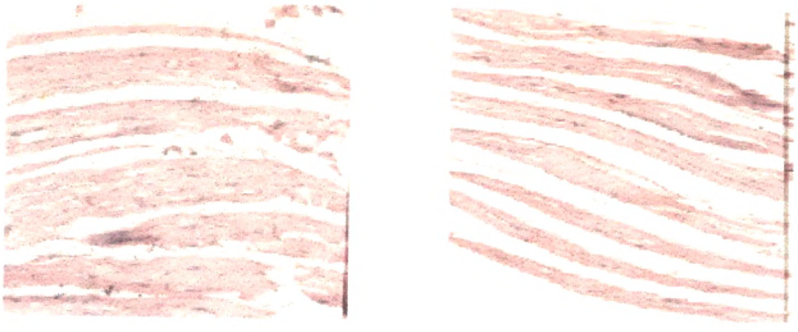
The potential toxicity of the microspheres to the synovium was evaluated by histopathological studies. 8-10 mg of plain microspheres were suspended in 0.2 ml saline and injected into the left knee joint, 0.2 ml saline being injected into the right knee joint as control. Three days and seven days after the injection, the rats were sacrificed and the joints were isolated. They were fixed in buffered formalin and embedded in paraffin wax. Sections (5μm) were cut, stained with eosin and haemotoxylin and evaluated for the inflammatory changes. Photographs were taken using Olympus microscope at a magnification of 40x.

Figure 10.1: Histopathology of the joints

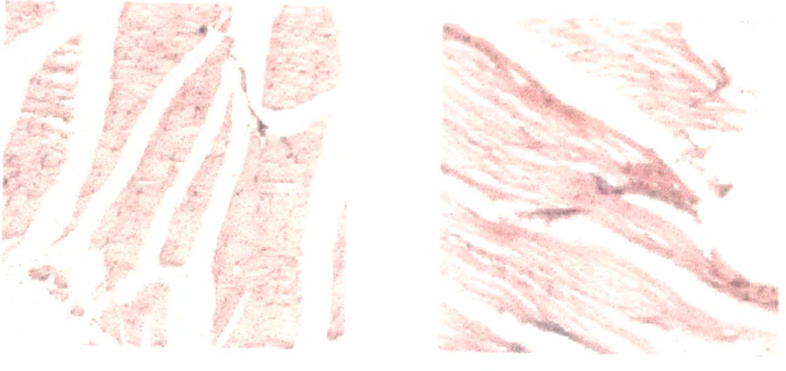
CMS=Chitosan microspheres **AMS=Albumin microspheres**
SLN=Solid lipid nanoparticles **GMS=Gelatin microspheres**



GMS



SLN



10.3 Results and discussion

Histopathology has been used by the previous workers for evaluation of biocompatibility of the microspheres with the synovium (Ratcliffe et al, 1984, Brown et al, 1998). Microspheres prepared using different polymers were injected intra-articularly and the toxicity was evaluated by morphological as well as histopathological studies. As reported by earlier workers (Ratcliffe et al, 1984) albumin microspheres were found to be most acceptable to the tissues of the knee joint. Our results indicated that the prepared microspheres are not highly toxic to the synovium. There were no inflammatory infilterates observed in case of chitosan microspheres, gelatin microspheres and the solid lipid nanoparticles. However, albumin microspheres exhibited some inflammatory infilterates in the synovium. Similar results were obtained by previous workers (Bogdansky, 1990) where repeated injections of albumin microspheres in the knee joints of rabbits caused pronounced rapid joint swelling, while with gelatin microspheres no swelling occurred. The inflammatory potential of the albumin microsphere may have been responsible for not decreasing the inflammation on injection of the celecoxib loaded albumin microspheres in the arthritic rats.

10.4 References

Brown KE, Leong K, Huang CH, Dalal R, Green GD, Haimes HB, Jimenez PA, Bathon J. Gelatin/chondroitin 6 sulphate microspheres for the delivery of therapeutic proteins to the joint. Arthritis Rheum.1998; 41: 2185–2195.

Bogdansky S. Natural polymers as drug delivery systems in: "Biodegradable polymers as drug delivery systems", 1990, Chasin M., Ed., Marcel Dekker, Inc., New-York, pp-249.

Ratcliffe JH, Hunneyball IM, Smith A, Wilson CG, Davis SS. Preparation and evaluation of biodegradable polymeric systems for the intra-articular delivery of drugs. J. Pharm. Pharmacol. 1984; 36: 431-436.