The aim of present work was the formulation of particulate drug delivery systems containing antineoplastic agents (5-fluorouracil and methotrexate) & their in-vitro characterization. We have selected there biocompatible and biodegradable polymers (egg albumin, chitosan, PLGA) for preparation of microspheres. Microspheres were prepared with an objective of preparing a formulation that can be used for size dependent targeting of 5-FU & methotrexate entrapped in microspheres to organs like lungs. Targeting to organs like lungs can be achieved by selection of appropriate size of microspheres. In order to achieve the above mentioned objectives, it is essential that microspheres possesses following in vitro attributes —

1. Small size of the microspheres.
2. narrow particle size distribution
3. Discrete particles (i.e. disaggregated particles)
4. Higher drug content

It is also essential that the product prepared for achieving above mentioned objectives should have following attributes from a stability perspective:

- Particle size & particle size distribution should not alter during storage period,
- Aggregation of particles should not take place during storage,
- To decide storage conditions for recommendation on the label (room temperature or refrigerator),
Physical / chemical stability of the drug in the product,

Whether such product should be marketed in suspension form or dry powder form?

So the specific aim of the work was –

I. To formulate-
   a. Cross-linked egg-albumin microspheres of 5-FU and methotrexate,
   b. Cross-linked chitosan microspheres of 5-FU and methotrexate, and
   c. Poly (D, L-lactide-co-glycolide) microspheres of 5-FU and methotrexate.

II. To optimize formulation/process parameters for preparation of above mentioned microspheres with respect to in-vitro attributes listed above.

III. To carry out stability evaluation of microsphere products listed above.

Following approach was adopted to achieve above mentioned objectives:

1. Selection of a method of preparation based on available literature

2. Identification of process parameters

3. Optimization of the selected process parameters

4. Evaluation of product

1) Identification of independent (process) parameters:

   Every method of preparation has some parameters related to the process which may have significant bearing on various characteristics associated with a delivery system. These parameters may be important, if when the product's characteristics are dependent on them. At the same time, there may be some parameters that may not be important as they don't have significant effect on
important attributes of the delivery systems. Based on the literature survey, parameters were selected for each delivery system which has been found to have significant effect on characteristics of the microspheres of the polymers selected. Parameters were also selected based on the results of preliminary study for the formulation of the product with desired characteristics. Preliminary studies were conducted in order to incorporate suitable changes in a selected method, mentioned in the literature, due to use of different instrument/reagent than mentioned/used in the original method.

2) **Optimization of the selected independent (process) parameters:**

Once the process parameters were selected as per the above discussion, levels (or various values) of these parameters were decided. Using the selected number of process parameters and levels for each parameter, total number of batches to be prepared was calculated as per the Factorial Design (in case of optimization of two or more than two process parameters). Batches were prepared and subjected to evaluation with respect to selected pharmaceutical attributes.

3) **Evaluation of dependent parameters of the product:**

Product obtained from each batch was evaluated for following dependent parameters which play a major role in efficacy of controlled release of drug from the delivery systems:

1. Mean particle size
2. Particle size distribution
3. Entrapment efficiency (in case of drug loaded microspheres only)
Further, significance of effect of change in independent process parameters on the dependent parameters (i.e. product characteristics) was evaluated by using suitable statistical tool.

**STABILITY STUDIES:**

The best batch for each delivery system (based on some predecided factors as indicated in annexure-5) was planned to be subjected to stability studies at two conditions: (i) 25°C/60% RH, and (ii) 40°C/75% RH. Samples drawn at various time intervals were subjected to following evaluation studies:

1. **Physical stability of the product**

   Physical stability of the product subjected to accelerated stability studies was determined by determination of:

   a) Mean Particle size

   b) Particle size distribution

   c) Drug content (in case of products subjected to accelerated test in solid state)

   d) Drug leaching (in case of products subjected to accelerated test in suspension state)

   e) Morphology of the product by taking scanning electron micrograph of the products.

2. **Chemical stability of the product**

   Chemical stability of the product subjected to accelerated stability studies was determined by qualitative determination of purity of drug using some method as mentioned in literature.
IN-VITRO RELEASE STUDY:

Study of release pattern of the best batch was done. From the pattern of release of drug from the selected batch of microspheres for each category of formulation, further investigation regarding the mode of release of drug from the system was investigated.