

EFFECTS OF SPHERE SIZE, POLYMER TO DRUG RATIO AND PLASTICIZER CONCENTRATION ON THE RELEASE OF THEOPHYLLINE FROM ETHYLCELLULOSE MICROSPHERES

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تم تحضير كريات دقيقة طويلة الإتاحة من عقار ثيوفيللين باستعمال طريقة الإضافة بعدم استخدام المذيب. ولقد درست الإتاحة العملية للعقار من الكريات الدقيقة المحضرة وذلك باستعمال كريات مختلفة الأحجام. وقد تمت دراسة تأثيرات استعمال نسب مختلفة من البوليمر إيثايل سيلليولوز إلي العقار وكذلك تأثيرات استعمال تركيبات مختلفة من الملدن المستعمل في الدراسة وهو داي إيثيل فيثالات علي الإتاحة العملية للعقار. ولقد وجد أن إتاحة العقار العملية نقصت نقصاً ملحوظاً بزيادة نسبة البوليمر وكذلك قلت نسبة تحميل الدواء علي البوليمر. ولقد نقص معدل إتاحة العقار أيضاً بزيادة نسبة الملدن من 10% - 30% مع عدم اختلاف في الإتاحة العملية من الكريات التي لا تحتوي علي الملدن والكريات المحتوية علي 10% ملدن.

Theophylline sustained release microspheres were prepared by applying the non-solvent addition method. The in-vitro release of the drug from the prepared microspheres of different size ranges (≤ 300 , 300-600, 600-800, 800-1000 and 1000-1250 μm) was studied. The effects of different ethylcellulose: theophylline ratios (1:2, 1:1 and 2:1) and different plasticizer (diethylphthalate) concentrations (10, 20 and 30% v/w) on theophylline release were also investigated. At different ethylcellulose: theophylline ratios (1:1, 2:1 and 1:2), the release of the drug decreased significantly when the polymer content was increased and the drug loading was decreased. Also, the release was significantly decreased ($P < 0.05$) with increasing plasticizer concentration from 10 to 30%, with no significant difference between un-plasticized and 10% plasticizer.

Key words: Ethylcellulose microspheres, diethylphthalate concentration, theophylline release

Introduction

Preparation of sustained release dosage forms is one of the main objectives in drug formulation. Many sustained release dosage forms are designed to release the drug at slow rates maintaining uniform selective therapeutic drug levels for an extended period of time. These dosage forms have the advantages of reducing the frequency of dosing, lowering adverse effects and improving patient compliance (1).

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The successful formulation to control drug release for the required duration of time with optimum release pattern depends on various factors such as the physicochemical properties of the drug, nature of drug-carrier, ratio of polymer to drug, type of the dosage form and route of administration (2-8).

Polymeric microspheres and microcapsules have received much attention as drug delivery systems in recent years and used to modify and retard drug release (9). Their preparation results in the coating of the individual drug particles by inert polymeric materials, through which the drug would diffuse at a controlled and predictable rate to the surrounding medium (10).

There are several techniques used to produce polymeric microspheres drug delivery systems, which include physicochemical processes (such as sol-

vent evaporation method or phase separation method), mechanical processes (such as spray drying), and a non-solvent addition process (11-17).

Theophylline is the drug commonly used in management of asthma. It has a relatively short half-life and a narrow therapeutic index, with 5-20 µg/ml serum concentrations. Thus, sustained release theophylline formulations that can produce more uniform serum drug concentrations with less fluctuation in peak-through levels would be useful (18).

The aim of this study was to prepare sustained release microspheres of theophylline, using ethylcellulose as a release retarding material, by applying the non-solvent addition technique, and study the effects of spheres size, ethylcellulose theophylline ratio (EC:theo) and the plasticizer diethylphthalate concentration on the release characteristics of the prepared microspheres.

Experimental

Chemicals:

Theophylline powder was obtained from Sigma Chemicals (St. Louis, MO USA). Ethylcellulose, with 2.42 to 2.53 degree of substitution and viscosity of approximate 14 cp for 5% solution in 80:20 toluene:ethanol by weight, was obtained from Fluka Chemie AG (Switzerland). Petroleum ether and toluene were purchased from BDH Chemicals Ltd. (Poole, England). Diethylphthalate was obtained from Riedel-DE Haenag (Germany).

Methods

Preparation of Theophylline Microspheres:

Theophylline microspheres were prepared by using a non-solvent addition method. The calculated amount of theophylline (2.5 gm) was dispersed in 50 ml of EC polymeric solution in toluene (5% w/v) and stirred at 700 rpm for 15-minutes hours using Lightnin Lambaster paddle stirrer (T52010, USA). Diethylphthalate was added to the EC polymeric solution in a concentrations of 0, 10, 20 or 30 % of EC content, as a plasticizer and stirred for 15 min. Then, 100 ml petroleum ether was added slowly with continuous stirring for 2 hours at 25°C. After decantation and washing the produced microspheres with 50 ml petroleum ether in portions, to remove the excess toluene, the formed microspheres were filtered off and then allowed to dry in open air. Microspheres containing different concentrations of

plasticizer (0, 10, 20 and 30% v/w), and different EC:Theo ratios (1:2, 1:1 and 2:1), were prepared.

Particle size analysis:

Fractions of ≤ 300 , 300-600, 600-800, 800-1000 and 1000-1250 µm of the prepared theophylline microspheres were separated by shaking using a set of standard sieves (British Standard). Selected fractions were used for in-vitro dissolution.

Determination of drug content:

A weighed quantity (100 mg) of microspheres was extracted with 50 ml ethanol using sonication. The drug content was spectrophotometrically assayed at 272 nm [16] using Philips Pu 8620 spectrophotometer (England, UK). Each determination was carried out in triplicate. The incorporation efficiency was calculated as percentage of the theoretical content. Drug free microspheres, prepared exactly with the same manner, was used as reference.

In-vitro release studies:

The release measurements were performed using USP dissolution apparatus 2, at 50 rpm (Caleva Ltd., Model 85T) using a continuous automated monitoring system which consists of an IBM computer PK 8620 series and PU 8605/60 dissolution test software, Philips VIS/UV/NIR single beam eight cell spectrophotometer Model PU 8620, Epson FX 850 printer, and Watson-Marlow peristaltic pump. In each flask, 0.9 Liter phosphate buffer pH 7.4 was used as a dissolution medium. The temperature was maintained at 37 ± 0.5 °C. An accurately weighed amount of the prepared microspheres equivalent to 50 mg of the drug was added to each flask. For each formula, release was run in triplicate and absorbance was recorded automatically at 272 nm up to 8 hr. The percentage of drug released was determined as a function of time.

Statistical analysis:

The significance of difference between the dissolution of different studied patches with various variables was evaluated using the analysis of variance (ANOVA) followed by Tukey-Kramer multiple comparison test. The differences were considered significant at $p \leq 0.05$. These statistical calculations were performed using the Graph Pad InStat computer program (1990-1993; Graph Pad Software, V2.04, San Diego, CA).

Results and Discussion

Ethylcellulose has been reported as one of the important polymeric materials used in microspheres and microencapsulation formulations (19-26). This is due to its high safety, good stability, easy fabrication and cheapness (16).

Theophylline microspheres were successfully obtained in a size ranging from ≤ 300 -1250 μm . The drug content in the prepared microspheres ranged from 95 to 102 % of the theoretical content, indicating high incorporation efficiency. The total yield value was not less than 70% in all batches.

Fig. 1 shows the percent theophylline released from microspheres of different particle size ranges (≤ 300 , 300-600, 600-800, 800-1000 and 1000-1250 μm). The results showed that the release from microspheres of the size range ≤ 300 μm was significantly ($P < 0.05$) higher than that from all other studied ranges (300-1250 μm). This presumably is due to the much larger surface area available for release, and the shorter path length necessary for drug to diffuse through (27). There was no significant difference ($P > 0.05$) in the release between the sizes of 300-600 μm and those of size ranges 600-800 and 800-1000 μm . The slowest release was from the largest particle size range 1000-1250 μm .

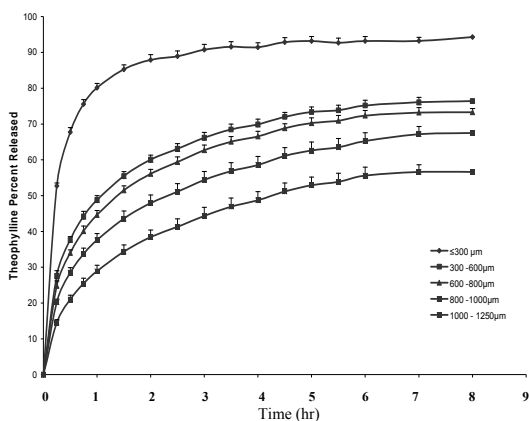


Fig. 1 Percent theophylline released from different particle size ranges (≤ 300 , 300- 600, 600-800, 800-1000 and 1000-1250 μm) microspheres of 1:1 EC:Theo ratio and containing 10 % plasticizer (n= 3).

Fig.2 shows the release of theophylline from microspheres containing 10 % plasticizer and various EC:Theo ratios of 1:1, 2:1 and 1:2. The results showed that the release from microspheres of 1:1

ratio was significantly ($P < 0.05$) higher than that from microspheres of 2:1 EC:Theo ratios. While, it was not significantly ($P > 0.05$) different from that of 1:2 EC:Theo ratios. Also, the release from microspheres of 1:2 EC:Theo ratios was significantly ($P < 0.05$) higher than that of 2:1 EC:Theo ratios. These results emphasized the fact that as the polymeric thickness increases (2:1) and the drug loading decreases, the release of the drug decreases from the microspheres. It has been reported that the thickness of the polymeric membrane barrier determines the lag time and duration of drug release (28).

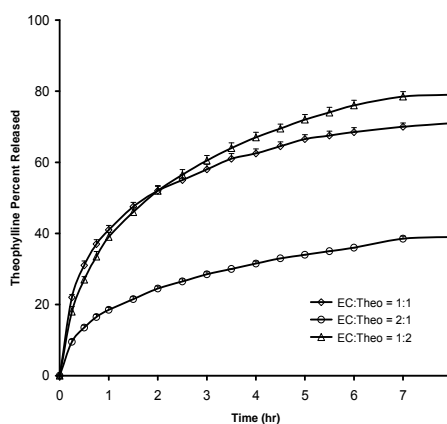


Fig. 2 Percent theophylline released from microspheres prepared using different EC:Theo ratios (1:1, 2:1 and 1:2) and containing 10% plasticizer (n=3).

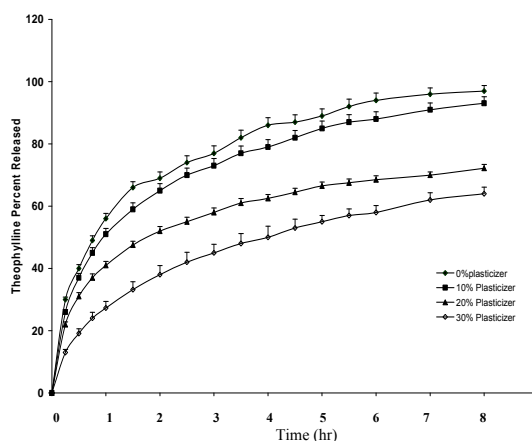


Fig. 3 Effect of different plasticizer concentrations (0, 10, 20 and 30%) on percent theophylline released from microspheres of 1:1 EC:Theo ratio (n=3).

The effect of different plasticizer concentrations (0, 10, 20 and 30%) on release from the chosen particle size range (600-1000 μm) and EC:Theo ratio (1:1) of the microspheres is shown in Fig.3. The results indicated that the drug release from microspheres decreased as the plasticizer concentration increased. No significance ($P > 0.05$) difference was observed for those microspheres containing 0 or 10% plasticizer, while the drug release was significantly decreased ($P < 0.05$) upon increasing plasticizer concentrations from 10-20 % and from 20-30%. In the preparation of microspheres, the plasticizer is used to render the wall material more elastic and flexible and never get fragile or ruptured under pressure (29). Diethylphthalate is hydrophobic in nature and supposing, if the plasticizer is more hydrophobic than the wall forming material (ethyl-cellulose), the release of the drug through the ethyl-cellulose wall membrane is reduced (29), which is confirmed in these results.

From the above mentioned results, it was obvious that the optimum release was from microspheres of size ranges 600-800 and 800-1000 μm prepared using EC:Theo ratios of 1:1 and containing 10% diethylphthalate as plasticizer. 10% plasticizer is the most common ratio of plasticizer used in microencapsulation for other polymers (28).

Table 1. Comparison of correlation coefficient (r) from *in-vitro* release data of theophylline microspheres (600-1000 μm) containing 10% plasticizer fitted to various release models.

Release Order	r values for EC:Theo Ratios		
	1 : 1	1 : 2	2 : 1
Zero	0.937	0.954	0.963
First	0.973	0.990	0.972
Higushi diffusion model	0.983	0.990	0.995
n	0.375	0.502	0.364

In order to obtain meaningful information for the release, the drug release data were fitted to various kinetic models. Table (1) summarizes the correlation coefficient (r) for the different release kinetic models of theophylline microspheres. Models with higher r values were judged to be more appropriate models for the release data. The linear relationship between

the logarithm of the percentage drug remained to be released from the microspheres as well as the relationship between the amount of theophylline released and square root of time, indicated that the drug release appeared to fit either first order or Higushi diffusion model. The n values were determined to verify release mechanism using the following equation;

$$Mt/M_{\infty} = K.t^n$$

Where:

Mt/M_{∞} is the fraction released by the drug at time t
K is a constant incorporating structural and geometric characteristic and
n is the release exponent characteristic for the drug transport mechanism.

It is known that, when $n = 0.5$ fickian diffusion is observed and the release rate independent on t, while $0.5 < n < 1.0$ indicate anomalous (non fickian) transport and when $n=1$, the release is zero order.

Higher r values for Higushi diffusion model were observed in all tested ratios. The value of n was equals 0.502 in case of 1:2 ratio, confirming fickian diffusion mechanism of theophylline release for the prepared microspheres.

In conclusion, the proper dissolution rate from theophylline microspheres was achieved by the preparation of the microspheres with 1:1 EC:Theo ratio, particle size range from 600- 1000 μm , and containing 10% diethylphthalate as plasticizer.

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