# Sodium carboxymethylcellulose/pectin microparticles as a new carrier for colon targeting of Progesterone

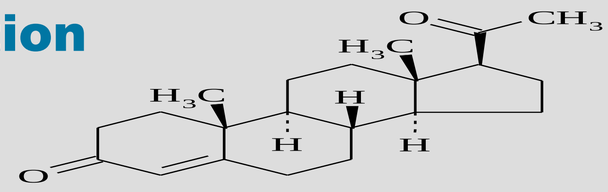
Ahmed M. El-Sayed, I. El-Gibaly, Fergany A. Mohamed, Ghareb M. Soliman and **Hytham H. Gadallah**

*Department of Pharmaceutics, Faculty of Pharmacy, Assiut University, Assiut, Egypt*

## Abstract

The colon is being viewed as a promising site of drug delivery owing to its long transit time (up to 78 hrs), which is likely to increase the time available for drug absorption. MP's were prepared using sodium carboxymethylcellulose (NaCMC)/pectin mixture as a biodegradable matrix, zinc acetate and aluminum sulfate as cross-linkers. A $2^4$ full factorial design was carried out to optimize the experimental conditions. The prepared MP's were investigated under conditions mimicking mouth-to-colon transit. The results obtained implied that, regardless the concentration of cross-linking agents, the polymer and drug concentrations exhibited the greatest influence on the drug entrapment efficiency (EE), which decreased as the drug concentration decreased from 1 to 0.5% w/v (95.13% and 83.88% respectively) and as the polymer concentration increased from 1.25 to 1.5% w/v (107.35% and 95.13% respectively). On the other hand, MP's prepared with 1% drug showed a significantly slower release rate than those prepared with 0.5% drug as indicated by the values of Mean Dissolution Time (MDT) & the release rate constant(k). This study confirms the viability of the prepared microparticles as a colon-targeted drug delivery system.

## Introduction



**Figure(1) Molecular structure of Progesterone**

Progesterone (PG, $C_{21}H_{30}O_2$ = 314.5) which is a natural hormone, is widely used as a hormonal contraceptive and in habitual or threatened abortion. It has a short elimination half-life (~19-95 min) and undergoes extensive first-pass hepatic metabolism which results in very low oral bioavailability (~25%). Thus, the aim of this work was to study the feasibility of preparing PG-loaded colon-targeted microparticles (MP's) to overcome the shortcomings associated with the oral administration of PG.

## Methods

### 1. Formulations design:

MP's were prepared based on $2^4$ full factorial design. The independent variables and their levels are shown in table(1). On the other hand, the release rate constant (K, $mg.h^{-n}$, SSIF (pH 7.4), Y1), the drug entrapment efficiency ratio (EE ratio, %w/w, Y2) and mean dissolution time (MDt (h), SSIF (pH 7.4), Y3) are the dependent variables (response parameters).

### 2. Preparation of MP's:

The ionotropic gelation technique was adopted to entrap PG in MP's as summarized in figure(2)

| Factor | Coded levels | | |
|---|---|---|---|
| | –1 | 0 | 1 |
| ($X_1$) Total polymers (CMC / pectin ratio 1:1) concentration (% w/v) | 1.25 | 1.375 | 1.50 |
| ($X_2$) Drug concentration (% w/v) | 0.50 | 0.75 | 1.00 |
| ($X_3$) Concentration of $Zn(CH_3COO)_2$ (M) | 0.035 | 0.0425 | 0.050 |
| ($X_4$) Concnetration of $Al_2(SO_4)_3$ (M) | 0.0125 | 0.0188 | 0.025 |

**Table(1) A $2^4$ factorial design independent variables and their actual levels**

### 3. Formulation characterization:

*3.1. Determination of the entrapment efficiency (EE) of PG in MP's:*

Known weight of MP's were broken in a mortar, extracted several times with ethanol and stirred for 1 h. The filtered solution was assayed spectrophotometrically at 242 nm

*3.2. Equilibrium swelling studies of MP's:*

Swelling tests were separately carried out in enzyme-free simulated gastric fluid (SGF, pH 1.2), and enzyme-free simulated small intestinal fluid (SSIF, pH 7.4). The swelling ratio (SR) of the MP's was then calculated from the formula:

$$SR = (Wt - Wo) / Wo$$

*3.3. In vitro drug Release Studies:*

In order to simulate the pH changes along the GIT, the sequential pH change method is used in all release studies. MP's were tested in enzyme-free simulated gastric fluid (SGF, pH 1.2) for 2 h, followed by enzyme-free simulated small intestinal fluid (SSIF, pH 5.5) for 0.5 h and then enzyme-free simulated small intestinal fluid (SSIF, pH 7.4) for the subsequent hours.

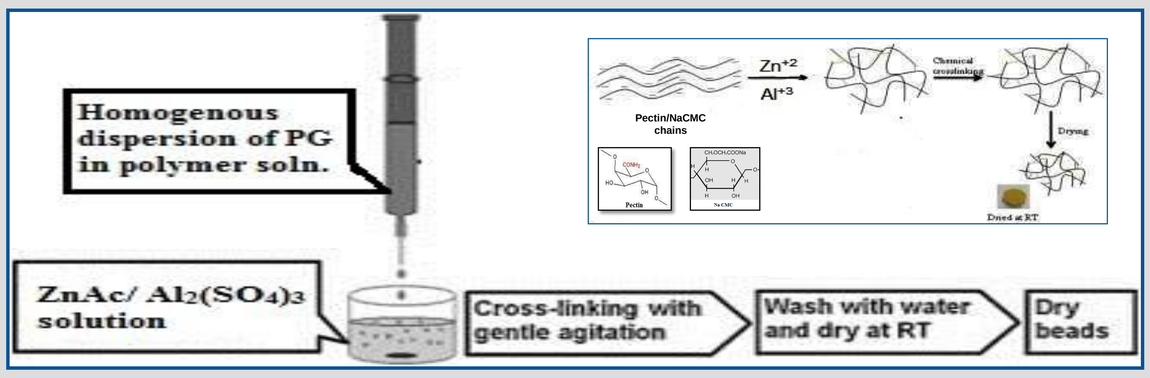
*3.4. Scanning electron microscopy (SEM):*

Morphological examination of the dried MP's was conducted using scanning electron microscope.

*3.5. Compatibility studies of MP's:*

Differential scanning calorimetric (DSC) analyses were performed on the solid samples of MP's.

## Results

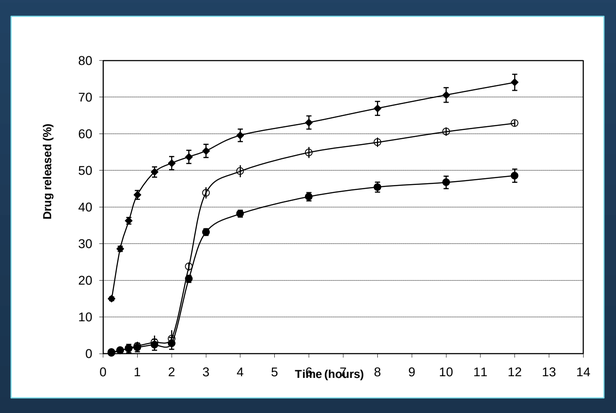



**Figure(2) The ionotropic gelation method**

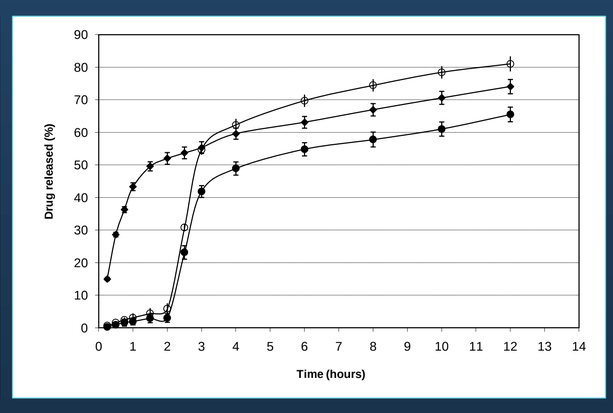
• The mean diameter of the prepared drug-loaded pectin/NaCMC gel microparticles after drying ranged between 0.9 and 1.5 mm (depending on the drug and polymer concentrations).

| Experiment code | $X_1$ | $X_2$ | $X_3$ | $X_4$ | Release rate constant (K,$mg.h^{-n}$) | Entrapment efficiency (EE, % w/w) | Mean dissolution time x $10^4$ (MDT,hr) |
|---|---|---|---|---|---|---|---|
| M1 | 1.25 | 0.5 | 0.035 | 0.0125 | 1.78 ± 0.422 | 102.50 ± 1.22 | 77.691 ± 0.011 |
| M2 | 1.50 | 0.5 | 0.035 | 0.0125 | 2.32 ± 0.570 | 92.71 ± 1.008 | 16.72 ± 0.00256 |
| M3 | 1.25 | 1.0 | 0.035 | 0.0125 | 1.428 ± 0.416 | 108.36 ± 6.4 | 298.94 ± 0.0262 |
| M4 | 1.50 | 1.0 | 0.035 | 0.0125 | 1.473 ± 0.181 | 101.27 ± 2.46 | 128.57 ± 0.0111 |
| M5 | 1.25 | 0.5 | 0.050 | 0.0125 | 1.436 ± 0.148 | 97.85 ± 5.81 | 237.2 ± 0.0152 |
| M6 | 1.50 | 0.5 | 0.050 | 0.0125 | 2.981 ± 0.391 | 82.33 ± 3.67 | 2.45 ± 0.000334 |
| M7 | 1.25 | 1.0 | 0.050 | 0.0125 | 1.187 ± 0.0654 | 110.77 ± 1.19 | 473.5 ± 0.023 |
| M8 | 1.50 | 1.0 | 0.050 | 0.0125 | 1.468 ± 0.1804 | 101.14 ± 5.82 | 139.0 ± 0.00911 |
| M9 | 1.25 | 0.5 | 0.035 | 0.0250 | 1.913 ± 0.595 | 94.39 ± 3.97 | 159.97 ± 0.0258 |
| M10 | 1.50 | 0.5 | 0.035 | 0.0250 | 1.98 ± 0.293 | 83.84 ± 5.35 | 25.9 ± 0.855 |
| M11 | 1.25 | 1.0 | 0.035 | 0.0250 | 1.311 ± 0.238 | 105.70 ± 2.36 | 374.33 ± 0.0415 |
| M12 | 1.50 | 1.0 | 0.035 | 0.0250 | 1.372 ± 0.145 | 98.36 ± 2.85 | 215.53 ± 0.0182 |
| M13 | 1.25 | 0.5 | 0.050 | 0.0250 | 2.767 ± 0.660 | 92.4 ± 3.94 | 1.894 ± 0.000244 |
| M14 | 1.50 | 0.5 | 0.050 | 0.0250 | 2.724 ± 0.867 | 83.88 ± 2.15 | 5.78 ± 0.00083 |
| M15 | 1.25 | 1.0 | 0.050 | 0.0250 | 1.163 ± 0.311 | 107.35 ± 2.00 | 1083.0 ± 0.0884 |
| M16 | 1.50 | 1.0 | 0.050 | 0.0250 | 1.50 ± 0.135 | 95.13 ± 3.91 | 137.83 ± 0.00593 |

**Table(2) Matrix of the prepared formulations and their response parameters. Results represent mean ± SD of 3 observations**




**Figure(3)** Effect of drug concentration on drug release.[O, 0.5% w/v drug (M2); ●, 1% w/v drug (M4) ♦; free drug]




**Figure(4)** Effect of cross-linking agent concentration on drug release.[O, $Al_2(SO_4)_3$ conc.: 0.0125 M (M6); ●, $Al_2(SO_4)_3$ conc.: 0.025 M (M10); ♦, free drug]

• The EE of PG decreased as the drug concentration decreased and as the polymer concentration increased. This was attributed to the low mass transfer with the more viscous medium in presence of higher polymer concentration (Table 2)

• The main effects and interactions with their magnitude and direction of the response parameters were obtained from the predictor polynomial equations:

$$EE (\%\ w/w) = 97.37 - 5.047\ X_1 + 6.146\ X_2 - 1.01\ X_3 + 1.092\ X_2X_3 - 2.234\ X_4 - 1.049\ X_2X_3X_4$$

$$K (mg.h^{-n}) = 1.8 + 0.177\ X_1 - 0.437\ X_2 + 0.103\ X_3 - 0.136\ X_2X_3 - 0.124\ X_1X_4 + 0.094\ X_3X_4 + 0.133\ X_1X_2X_4$$

$$MDt (h) = 0.0211 - 0.0127\ X_1 + 0.0145\ X_2 - 0.00739\ X_1X_2$$

• All formulations remained intact with no visible swelling or erosion for the first 2 hours in S.G.F. with negligible drug release (< 6%).

• It was observed that MP's prepared with a high drug level (1% w/v, formulations M4) have slower in-vitro release profiles than those prepared with lower drug level (0.5% w/v, formulation M2).This may be due to the higher amount of encapsulated drug (fig.3)

• Regardless of $Zn^{2+}$ ions concentration, increase in $Al^{3+}$ ions concentration from 0.0125 M (M6) to 0.025 M (M10), markedly decreases the percentage of drug release. This findings may be attributed to the fact that the cross-linking reaction involves competition on $Al^{3+}$ ions between NaCMC and pectin (fig. 4)

• Scanning electron micrographs showed typically spherical MP's with a smooth surface and no deep fissures or cracks. Drug crystals were markedly embedded in the surface of MP's (fig. 5)

• DSC thermograms of the selected formulations showed no physical or chemical interaction between the drug and polymers used.


**Figure(5) Surface morphology of drug-loaded pectin/NaCMC gel microparticles as appears under SEM**

## Conclusion

The obtained results showed that PG, as a model drug, was successfully encapsulated in the MP's reflecting the efficiency of the preparation procedure. The prepared MP's had good morphological and release characteristics, preventing premature drug release in the upper GIT. This can reduce the GI side-effects and allow for extended duration of action and thus, Improving the oral bioavailability and tolerability of PG. This system, which hasn't been reported in the literature before, seems to be promising for delivery of water insoluble drugs to the colon providing the majority of drug release only upon breakdown by the bacterial microflora in the colon. In vivo studies of the prepared formulations and their degradation behavior in presence of pectinolytic enzymes are currently in progress.

## References

- Ibrahim El-Gibaly, *Int. J. Pharm.* 232, 199-211, (2002).
- Anna T. Reza, Steven B. Nicoll, *Acta Mater.* 6, 179-186, (2010).
- P.G Paterakis, E.S Korakianiti, P.P Dallas, D.M Rekkas, *Int. J. Pharm.* 248, 51-60, (2002).
- Pornsak Sriamornsak, Ross A. Kennedy, *Eur. J. Pharm. Sci. 29, 139-147, (2006).*
- Ali Assifaoui, Odile Chambin, Philippe Cayot, *Carbohydr. Polym.* 85, 388-393, (2011).
- Ellen Hagesaether, Ragnar Bye, S. Arne Sande, *Int. J. Pharm.* 347, 9-15, (2008).