**Référé 1**

**1.A./** 1. The first one question has not been answered at all ("The approach applied in choosing the process parameters, polymer/drug ration, type of polymer matrix is not systematical and it is quite difficult to follow",etc...) and is still difficult to follow the set up conditions since for each Lot both polymer/drug ratio and polymer/solvent ratio was changed. Please, read the comments more carefully before answering. Another part of the first one question has not been answered as well ("Also,
only for one microspheres preparation, CAB as polymer matrix was used with no explanation why the authors did not set up the condition with this polymer, too."). The authors explained just how they prepared this type of microspheres and write the preparing conditions, but the reason why did they not set up conditions for EC polymer matrix but just chosen one already used for another polymer matrix has not been explained. Please, review this part, too.

**Response:**

1. The purpose of this study is to optimize the experimental conditions to get high encapsulation efficiency and drug entrapment DL% by modifying and adjusting the process parameters. So, in this work, we tried to establish a relationship between the nature of matrix, the polymer/solvent ratio, the number of blades and the stirring speed by calculating the distribution of the prepared formulations; this is because, when the size distribution is minimized, a regular controlled release is achieved.

Two cellulosic polymers EC and CAB were used separately in order to study their influence on the release of the active agent in the simulated gastric medium. Their structural difference will directly affect the size and size distribution of the prepared formulations and thus on the release rate of the single active agent used. By comparing the two matrices EC and CAB, under the same operative conditions, we have obtained fast release from EC microspheres (Fig. 4).

After systematic analysis of the relevant bibliographies, and from the investigation of different literatures, it was found that each factor can be controlled in its effect on the microspheres properties. Then the effects of the selected process variables on the microparticles' characteristics, that is, the size, the drug content, and drug physico‐chemical properties, that is, the drug dissolution, have been elucidated and added in Results and Discussion section (see the text in red color).

For a given matrix (EC), the polymer concentration in a fixed volume of solvent and the drug/polymer ratio have a significant effect on microsphere properties.

-For MS1–MS3 microparticles, the d32 varied from 166 to 271 μm; here, the EC concentration and 2-ABZT:EC ratio were varied. The results demonstrated that the increase of polymer concentration increased the microparticles' size (Fig. 1 and Table I); the same remark has been reported in other drug/EC microspheres preparation. In fact, it has been proved in several papers using the same method that these parameters had a strong effect on the microparticle's size.20,23,24,41,42 Indeed, Raut et al. in the preparation of metoprolol succinate microspheres mentioned that the increased EC or other polymer (such as HPMC) in a fixed volume of solvent increases the viscosity of the medium, which might have diminished the shearing efficiency leading to increased droplet size and hence microsphere size.41 **Elbahri et al demonstrated that the parameters that have considerably affected the microspheres’ size are the polymer/solvent ratio and the stirring speed23.** Also, Sharma et al. reported that the mean particle size of repaglinide EC microspheres increases with increasing polymer concentration and viscosity42.

On the other hand, the drug/polymer parameter directly affects the drug entrapment %DL (DL–Drug Loading,%): The results showed that when the drug/polymer ratio is decreased from 1:2 to 1:4, the theoretical drug entrapment decreased and therefore the actual drug entrapment decreased from 24% to 18% (Table I). The same remark has been reported in other drug/EC microspheres preparation35. Also, some researchers reported that the drug loaded was increased by increasing initial drug/polymer ratio.23

However, for all the prepared microspheres MS1 and MS4-MS6 except the Formulations MS2 and MS3, the mean diameter *d*32 varied from 61 to 181 μm; in this part, only the carrier nature, the blade number and the stirring rate of emulsion were varied. Similar to other authors,35,37 we have remarked that the stirring speed had a strong effect on the microparticles' size; the mean diameter is effectively decreased when the stirring speed was increased. Moreover, the particle size distribution values (*δ*) were similarly decreased manner as these mechanical forces increased, supporting a more enhanced size-uniformity of the particles. Indeed, when comparing the lots MS1 and MS5 and where the blades number was varied, we remarked that this parameter has considerably influenced the particles size; in fact, increasing the blades number from four-bladed turbine to the sixth one yielded small microparticles.

1.B./- Another part of the first one question has not been answered as well ‘(’Also, only for one microspheres preparation, CAB aspolymer matrix was used with no explanation why the authors did not set up the condition with this polymer, too. ).

**Response**:

**1.B/** All microspheres loaded by 2-ABZT were produced using emulsion‐solvent evaporation technique. Firstly, using the selected EC matrix, four process parameters have been varied, namely, the polymer concentration, drug/polymer ratio (only for MS2 and MS3, for the other lots, the ratio 1:2 was already used (Table I)), the stirring speed and the blades number. In fact, it has been proved in several papers that these parameters had a strong effect on the microparticle's size.20 Secondly, CAB was used as polymer matrix to produce microparticles in the same operative conditions of lot MS1 formulation. In fact, the nature of matrix can affect the microspheres characteristics such as the release rate.

So, after the preparation and the optimization of the operative conditions of the EC microspheres and based on these results, the MS1 formulation was chosen for compare it with CAB polymer. This MS1 formulation has both a high drug entrapment DL% of 24% and a better size distribution δ = 1.22 (Table I) favorable for an extended release. Therefore, we studied under the same conditions of the lot MS1, the effect of the matrix nature on the release rate by replacing EC by CAB as a matrix. Thus, a single preparation of CAB microspheres was studied under the same operative conditions of the EC microspheres MS1 allows to compare the two polymer matrices and to derive the effect of the nature of the polymer matrix on microspheres characteristics (Table I).

* In the Table 1, third column was changed into a polymer concentration as recommended by Reviewer A. Further, the polymer concentrations were corrected in the text (in red color).
* The polymer concentrations are very close to each one, so it need to be explained why the authors expected to see difference in EE% and particle sizes by applying very similar concentrations.

**Response:**

We have obtained a difference in EE% and particle sizes by applying concentrations of 3% 5% and 6%. Thus, we have noted that the EE% increased from 73% to 89% when we **doubled** the EC polymer concentration from 3 to 6%. Such phenomenon is also suggested by Elbahri et al. working on 2,4-dichlorophenoxyacetic acid microspheres based on EC polymer23 and Raut et al. in the preparation of metoprolol succinate microspheres41. This may be explained by the fact that increase in polymer phase viscosity, typically caused by higher concentration of the polymer, could restrict the migration of the drug to the continuous phase and thus improve its entrapment41,42.

Also, we have seen that the Sauter mean diameter (*d32*) of the obtained lots of microparticles increased from 166 to 278 μm by doubling the polymer concentration. This result can be explained by the viscosity of the polymer EC, i.e. the increased EC in a fixed volume of solvent increases the viscosity of the medium, which might have diminished the shearing efficiency leading to increased droplet size and hence microsphere size.41 Indeed, many researchers reported the same observation.23,24,41,42 **For example, Elbahri et al. have studied the influence of the polymer concentration in a fixed volume of solvent (32 g) on the particle sizes and EE% of EC microspheres by applying polymer concentrations of 2.34% and 4.68% and obtaining the same observation23.**

Regarding the Fig. 1, SEM analysis demonstrate a spherical shape with a variation of the surface structure and particle size for microspheres prepared at different EC concentrations. So, in this study, the obtained difference in particle sizes and EE% by applying different concentrations of polymer was confirmed by SEM and Optical Microscopy.

2. The second one question was responded: The drug and the polymer matrixes chemical structures were given. The Scheme 1 with the desired resolution of structures has been replaced into Results and Discussion section and mentioned in the text.

3. In the Materials and Methods section, the infrared spectroscopy and the XRD analysis the information was added, please, see page 4 (text in red color).

4. The analysis of the FTIR spectrum of both CAB and EC was given in the Results and Discussion section and explained in detail.

6. The figure caption for the Figure 4 is properly written (Please, see the corrected Figure 4 in the text).

7. The *in vitro* release studies were performed in duplicates. We have corrected this part as recommended by the reviewer.  After the evaluation of the *in vitro* study in duplicates, the obtained results were presented in graph (Results and Discussion section, *In vitro* *release study*).

By plotting the two repeated kinetics for a single batch, the values obtained are close and therefore the two graphs are identical. The standard deviation values have been calculated and given on the following tables separately:

|  |  |
| --- | --- |
| t (min) | MS1 StandardDeviation SD  |
| 2 | ±0,012 |
| 4 | ±0,024 |
| 6 | ±0,010 |
| 8 | ±0,650 |
| 10 | ±0,092 |
| 12 | ±0,060 |
| 15 | ±0,270 |
| 18 | ±0,311 |
| 20 | ±0,024 |
| 25 | ±0,016 |
| 35 | ±0,036 |
| 45 | ±0,042 |
| 60 | ±0,022 |
| 75 | ±0,010 |
| 90 | ±0,030 |
| 105 | ±0,049 |

|  |  |
| --- | --- |
| t (min) | MS2 Standard Deviation SD |
| 2 | ±0,019 |
| 4 | ±0,017 |
| 6 | ±0,0025 |
| 8 | ±0,014 |
| 10 | ±0,030 |
| 12 | ±0,025 |
| 15 | ±0,041 |
| 18 | ±0,018 |
| 20 | ±0,027 |
| 25 | ±0,045 |
| 35 | ±0,051 |
| 45 | ±0,064 |
| 60 | ±0,011 |
| 75 | ±0,030 |
| 90 | ±0,013 |
| 105 | ±0,010 |

|  |  |
| --- | --- |
| t (min) | MS3 StandardDeviation SD |
| 2 | ±0,018 |
| 4 | ±0,049 |
| 6 | ±0,007 |
| 8 | ±0,53 |
| 10 | ±0,071 |
| 12 |  ±0,044 |
| 15 | ±0,013 |
| 18 | ±0,055 |
| 20 | ±0,036 |
| 25 | ±0,024 |
| 35 | ±0,046 |
| 45 | ±0,014 |
| 60 | ±0,027 |
| 75 | ±0,007 |
| 90 | ±0,011 |
| 105 | ±0,023 |

|  |  |
| --- | --- |
| t (min) | MS4 StandardDeviation SD |
| 2 | ±0,014 |
| 4 | ±0,021 |
| 6 | ±0,016 |
| 8 | ±0,010 |
| 10 | ±0,024 |
| 12 | ±0,030 |
| 15 | ±0,019 |
| 18 | ±0,018 |
| 20 | ±0,023 |
| 25 | ±0,042 |
| 35 | ±0,014 |
| 45 | ±0,021 |
| 60 | ±0,014 |
| 75 | ±0,029 |
| 90 | ±0,011 |
| 105 | ±0,015 |

|  |  |
| --- | --- |
| t (min) | MS5 StandardDeviation SD |
| 2 | ±0,010 |
| 4 | ±0,144 |
| 6 | ±0,069 |
| 8 | ±0,021 |
| 10 | ±0,026 |
| 12 | ±0,050 |
| 15 | ±0,010 |
| 18 | ±0,014 |
| 20 | ±0,010 |
| 25 | ±0,042 |
| 35 | ±0,017 |
| 45 | ±0,014 |
| 60 | ±0,010 |
| 75 | ±0,015 |
| 90 | ±0,057 |
| 105 | ±0,029 |

|  |  |
| --- | --- |
| t (min) | MS6 StandardDeviation SD |
| 2 | ±0,022 |
| 4 | ±0,025 |
| 6 | ±0,028 |
| 8 | ±0,026 |
| 10 | ±0,010 |
| 12 | ±0,030 |
| 15 | ±0,010 |
| 18 | ±0,021 |
| 20 | ±0,014 |
| 25 | ±0,011 |
| 35 | ±2,142 |
| 45 | ±0,010 |
| 60 | ±0,015 |
| 75 | ±0,021 |
| 90 | ±0,040 |
| 105 | ±0,015 |