Research Article

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Buccal Mucoadhesive System Formed by Liquid Crystals of Rasagiline Mesylate For Non- Oral Parkinson Treatment

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ABSTRACT

The morning "off period" in individuals with Parkinson's disease (PD), characterized by challenges in receiving the appropriate dose for sleep, insufficient drug levels in the blood, and gastrointestinal issues, poses a significant challenge that impacts the overall quality of life for these patients. Based on research findings, these issues can be successfully addressed using non-oral treatments exclusively. Numerous mucoadhesive systems, such as lyotropic liquid crystals (LCs), have been identified[1–4]. These systems can form an oblique connection with the buccal mucosa, thereby extending the residence time and optimizing drug bioavailability. The objective of this study is to prepare rasagiline mesylate (RM), glyceryl monooleate/poloxamer 188 liquid crystal matrices (GMO/Plx188 LCs) to improve the bioavailability of RM. The structures of the prepared formulations were examined by polarized light microscopy and SAXS analyzed. These formulations were also evaluated for several parameter such as appearance, visualization, content uniformity, homogenity, viscosity, mucoadhesion. Obtained results demonstrates the prospective of liquid crytals as a viable and alternative approach for effective RM buccal delivery.

Keywords: Liquid Crystals, Rasagiline mesylate, Glyceryl Monooleate, Parkinson, Buccal delivery

1. Introduction

Parkinson's disease (PD) is the prevalent neurodegenerative movement disorder. The main motor symptoms seen in patients are tremor, rigidity, bradykinesia/akinesia and postural instability, but whose clinical manifestation also includes other motor and non-motor symptoms like sleep problems, constipation, anxiety, depression and fatigu. It is known to be a chronic and progressive disease, so the symptoms become worse in the course of time[5]. One of the important problems affecting the quality of life of PD patients is the "off period". Since patients cannot take the medication while they sleep at night, the level of the medication in the blood decreases, however, due to the gastrointestinal problems seen in the patients, patients experience off periods in the morning hours. It is thought that orally administered treatments may be insufficient due to these negativities occurring in the PD process. When standard therapy is insufficient, alternative routes of administration and treatments should be considered [5].

Buccal administration of drugs offers several advantages for therapy because this route has high blood flow and prevents hepatic first pass metabolism and/or gastrointestinal drug degradation by allowing systemic absorption. However, ingestion and dilution of the dosage form with saliva may result in reduced formulation residence time, which may result in poor performance, loss of formulation, and poor absorption of drugs. Therefore, it is preferable to use mucoadhesive systems, such as lyotropic liquid crystals (LCs), can create a slanted contact with the buccal mucosa, enhancing residence time to optimize drug bioavailability.. LCs are one of the most preferred carrier systems by researchers recently due to their unique microstructure and physicochemical properties, and they have attracted significant attention because they exhibit the physical properties of both crystals and liquids.

Rasagiline mesylate (RM) is an irreversible MAO inhibitor with high selectivity for the B form of the enzyme, so in this respect, it is analogous to selegiline, a drug that has been shown to be of benefit in the treatment of PD. RM is freely soluble drug, its biological half-life is 3 hours and dose is 0.5- 2 mg per day. However, it is more potent than selegiline on a weight and molar basis [6]. RM undergoes extensive metabolism in the liver via N-dealkylation and hydroxylation. Therefore, the absolute bioavailability of RM is 36%. RM is an effective active substance with low oral bioavailability and therefore is considered to be a very suitable candidate for transmucosal administration. In a study, buccal film was developed for intraoral application of Rasagiline mesylate. In a study, buccal film was developed for intraoral application of RM [7]. In a study, researchers developed HPMC buccal film to improve the bioavailability of RM [8]. In a study by Toksoy et al., they prepared and optimized RM loaded solid lipid nanoparticles (RM-SLNs) in a thermosensitive mucoadhesive gel. Their results show that the optimized formulation could be a potential drug delivery system for the treatment of Parkinson's disease[9].

The aim of this study is to prepare RM glyceryl monooleate/poloxamer 188 liquid crystal matrices (GMO/Plx188 LCs) to deliver of RM. We aimed with these matrices to improve the bioavailability and of RM. At the same time, controlled release of RM can be achieved with the development of this system.

2. Materials and Methods

2.1. Materials

RM was gifted from Ali Raif Pharmaceutical Industry, Turkey. GMO was gifted from Arkem Chemistry, Turkey, Poloxomer was purchased from BASF, Germany.

2.2. Preperation of LCs

Generally, these systems are designed by mixing water, oil and surfactant to self-assemble in liquid crystalline mesophases. GMO: Plx 188 and distilled water were used for our formulations. Firstly, Plx 188 (1.5%) was dissolved in the calculated amount of water. Then RM was added to the water phase and dissolved. The glyceryl monooleate was melted at 50°C and the water phase brought to the same temperature was added and mixed in the homogenizer until it cooled down. It was then allowed to equilibrate at controlled room temperature for 24 hours. Six formulations were prepared in the GMO:water system, containing 8%, 12%, 16%, 20%, 25% and 30% water [10, 11]associated with high mortality. Some factors may intensify its aggressiveness, for example, the overexpression of the epidermal growth factor receptor (EGFR.

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2.3. Characterization of Mucoadhesive LCs

2.3.1. Polarizing Optic Microscope (POM)

The system which is used to observe samples of anisotropic liquid crystals is basically composed of Olympus BX-50P polarizing microscope, interference and optic filters and Olympus C-5060 digital camera as well as of the specified heating thermostat and a temperature- controlling system. The samples to be studied were sandwiched in between 200m-width glasses and hermetically sealed. Between the securely closed glasses, the samples were placed on the microscope stage attached to the specified temperature system by which to change the temperature and examined. Those hermetically sealed samples were put on the microscope stage coupled with the specific temperature system and studied by changing the temperature[12, 13].

2.3.2. Small Angle X-ray Scattering (SAXS)

Small angle X-ray scattering (SAXS) measurements of liquid crystal samples were performed by means of RigakuUltima 4 X-ray diffractometer. To obtain X-ray, the standard CuK source with a wavelength of 1.54 A was employed. The scattered X rays were detected by the scintillation counter-type detector. The experiments were conducted in a way that 2θ range was 0.08-4-degree scanning range 0.01 degree and scanning rate 0.2 degree/ minute. The samples to be examined were sandwiched in between the standard quartz glasses of 1mm in width. While the trials were all being performed in transmission mode at ambient temperature, the correction was made for the background caused by the medium as well. In the analysis of the samples, one employed Reference Intensity Ratio (RIR), which is a general method used to implement a quantitative phase analysis by means of scaling all other diffraction data with a given diffraction one. The graphics were drawn by employing the origin program and performing a logarithmic arrangement[13].

2.3.3. pH measurement

0.5 g of the formulation was dissolved in 15 mL distilled water and the pH was measured using a calibrated pH meter, Mettler Toledo, USA. All measurements repeated three times.

2.3.4. Viscosity examination

The viscosity of the liquid crystal formulations was determined at 25ºC with a rheometer device (TA TX Discovery HR1). The studies were carried out using a 40 mm diameter steel probe, a fixed pitch of 0.3 mm, and the shear rate varied between 10 and 2000 1/s. While the shear rate varied between these values, 40 shear stress measurements were taken. Shear stress shear rate graphs were drawn, and flow curves were obtained. For each sample, the study was performed with at least three replications [14].

2.3.5. Evaluation of the mucoadhesive properties

Mucoadhesion assessments were conducted using a texture profile analyzer (TA-TX Plus, Stable Micro System, UK) at 37 ºC. During the studies 500 g load cell and P10 Perspex probe were used. Filter paper soaked with mucin gel (8%) was used. LC formulation was placed to the cylinder probe. Then, formulation and mucin gel were put in contact with a preload of 6000 mN during 300 s. The cylinder probe was moved upwards at 0.10 mm/s up to the complete separation of the surfaces (porcine mucin gel-LCs). The force-distance graph was evaluated and the maximum breaking force was found. The mucoadhesion was determined by calculating the area under the force-distance plot (n=6) [15, 16]mechanical and mucoadhesive properties of thermoresponsive, binary polymeric systems composed of poloxamer (P407).

2.3.6. Content uniformity

The prepared liquid crystal formulations (2g) were weighed in eppendorf tubes. Then, 40 mL of ethanol:water (50:50) was added to the eppendorfs (n=3). Eppendorfs were left to be extracted at 100 rpm at room temperature for 24 hours. At the end of 24 hours, 1 mL of the samples in each eppendorf was taken and analyzed with validated HPLC method[17].

3. Results and Discussion

This study demonstrated that GMO serves as an effective surfactant for creating liquid crystals with water. It successfully generated stable lyotropic liquid crystals with both distilled water and a 1.5% Plx188 solution.

3.1. Polarizing optic microscope

LC mesophases with anisotropic characteristics exhibit different microphotographs called textures under a polarizing microscope[13]. Each mesophase shows a typical texture unique to itself. The ternary samples prepared and composed of GMO/Poloxamer 188/water were studied by Olympus BX50 polarizing microscopy through the sandwich type cells. Including %20, %25 and %30 water, the three samples of isotropic nature was obtained images of polarizing microscope (Figure 1-3). When these images compared with literature, results were found compatible with the literature [18, 19] 8F-B2ES, which has 2-[2-(butyloxy)]

Since phase separation was observed in the formulations with waiting and no liquid crystal image could be obtained under the optical microscope, characterization studies were continued with formulations containing only 25% and 30% water.

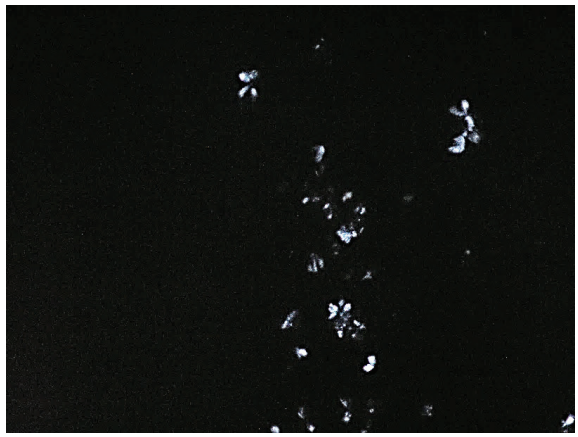

Figure 1. 20% water content

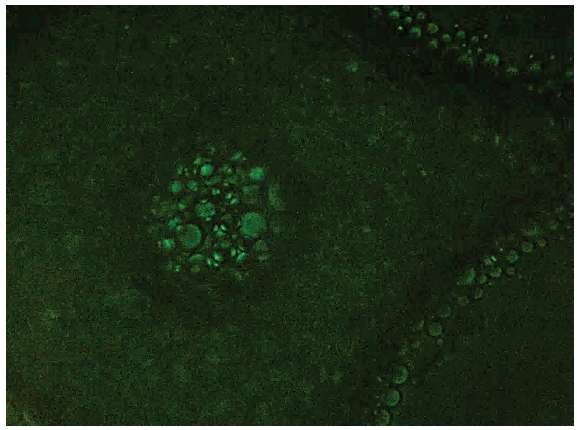

Figure 2. 25% water content

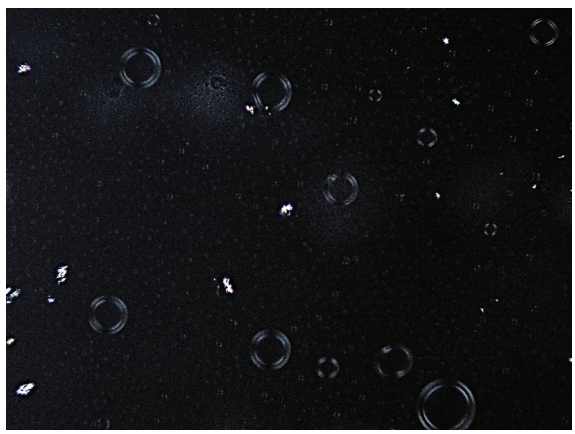

Figure 3. 30% water content

3.2. Small Angle X-ray Scattering

SAXS analysis was conducted to validate the findings from polarized light microscopy and offer additional quantitative insights into the nanostructure of the LCs[4].

As a result of literature research, it has been observed that the GMO/water binary system containing 25% and 30% water shows bicontinuous cubic phases of Gyroid type (G)[13, 18, 19]. However, with the addition of Poloxamer 188 as the third component to the bicontinuous system prepared within the scope of our study, it was observed that the geometry of the cubic phase changed and showed a Diamond type phase. Poloxamer 188 added to the medium is thought to affect intermolecular forces.

3.3. pH Measurement

The pH difference between the applied formulation and the buccal mucosal surface may cause tissue irritation. The pH of formulations was examined to establish its appropriateness for buccal use, thereby avoiding any sort of sensitivity or allergic reactions. The lowest pH value among the formulations was 6.15 and the highest pH value was 6.84. This is an indication that formulations were applicable to the buccal mucosa with a pH value of 6.8. In a literature, Salehi et. al. developed buccal film by using HPMC, polyvinyl alcohol (PVA), polyethylene oxide (PEO) as a polymer. The pH values of the formulations were found to be in the range of 6.54±0.04 to 6.98±0.01 for all formulations which are within the range of normal buccal pH [20].

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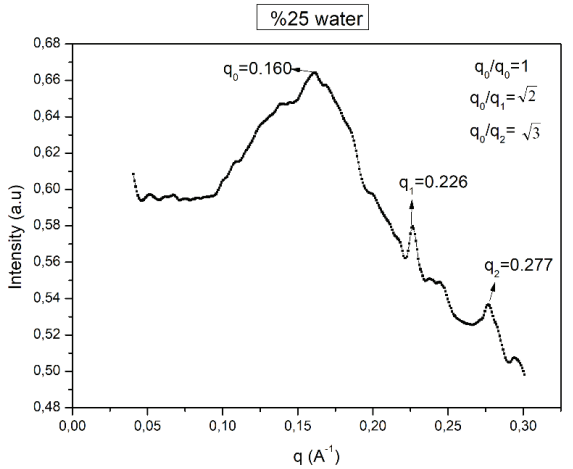

Figure 4. Diffraction peaks of 25% water content LC

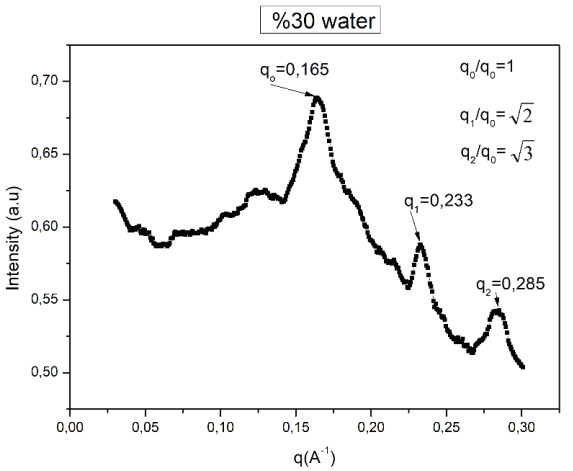

Figure 5. Diffraction peaks of 30% water content LC

3.4. Rheological examination

The viscosity curves of the LCs are shown in Figure 6. Selected formulations (25% and 30% water) showed pseudoplastic flow (Figure 3), indicating that the viscosity of the LC formulations decreased as they were mixed, with shear thinning behavior demonstrated.

When the results of the rheological analyzes were examined, it was observed that the viscosity of the formulations decreased with increasing shear rate. This will decrease the viscosity of the formulations when force is applied during application and facilitate their spreadability and applicability. With the disappearance of the force, an increase in viscosity is observed, which facilitates adhesion and prevents removal of the formulation from the buccal mucosa. In a study conducted by Marlow et al., it was examined how the viscosity of liquid crystal formulations changes with shear rate. As a result of the study, it was observed that the viscosities of the samples decreased with increasing shear rate[21].

3.5. Evaluation of the mucoadhesive properties

The results of the mucoadhesion studies, expressed as the maximum separation force (Fmax), are presented in Figure 4. The F1 formulation with the lowest water content and the F6 formulation with the highest water content show the highest and lowest Fmax values, respectively. Our findings confirm reports that mucoadhesion of GMO-based liquid crystal systems may be related to mesophase and water uptake capacities. GMO absorbs water and changes mesophase when in contact with excess water. Therefore, formulations with high GMO content were found to have higher Fmax values. Our results confirm that the mucoadhesion of the liquid crystal systems prepared with GMO can be associated with their mesophase and water uptake capacity. GMO absorbs water and undergoes phase transitions from the lamellar to the cubic phase when in contact with water[3, 22, 23]. The statement illustrates why mucoadhesion increases with an increased amount of GMO. The Fmax values of all the formulations we prepared were found to be suitable for buccal administration.

3.6. Content Uniformity

Results of quantification study were shown in Table 1. When the results of the study are examined, it can be said that the formulations are homogeneous due to the low standard deviation.

4. Conclusion

This research revealed that glycerol monooleate is a suitable surfactant for the formation of liquid crystals. Stable lyotropic liquid crystals were formed with GMO:distilled water (75:25) and (70:30) ratio and 1.5% plx188. Lyotropic Liquid Crystals can be recognized by different methods such as small angle X-ray scattering (SAXS), polarizing optical microscopy (POM), and transmission electron microscopy (TEM). POM and SAXS were also used in this study. It was observed by organoleptic studies that the formulations using Plx 188 were more stable than the

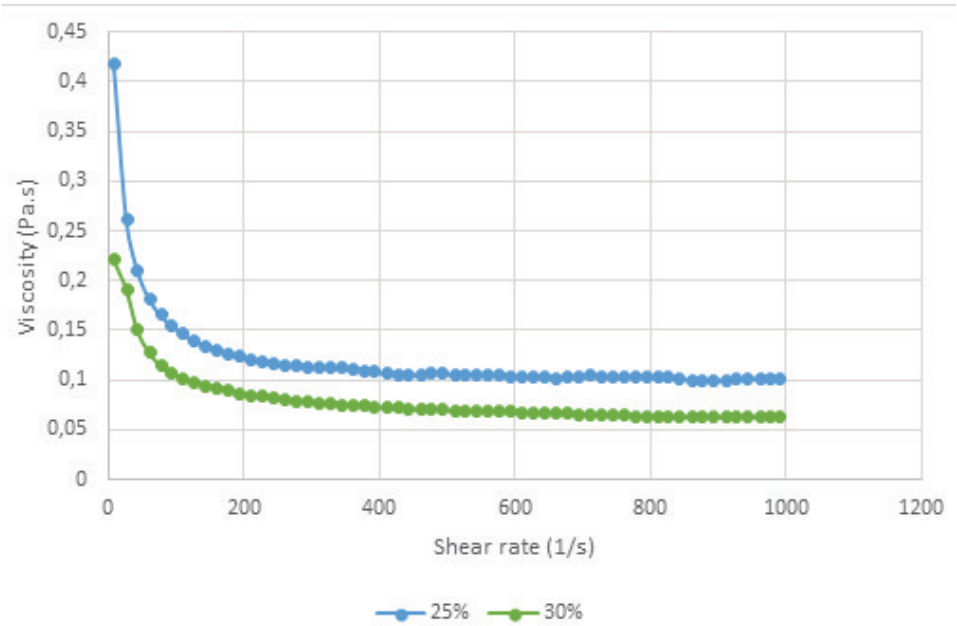

Figure 6. Viscosity/shear rate curve of formulations

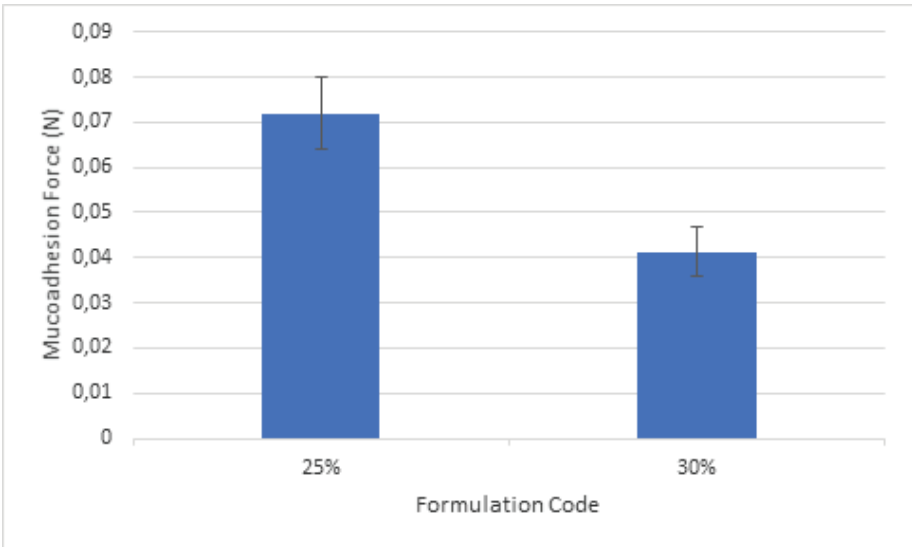

Figure 7. Mucoadhesion force of 25% and 30% water content LC

Table 1. Results of quantification

Formulation code	RM %
%25	98.19±1.25
%30	101.43±1.01

ones not used, and no phase separation was observed. LCs were visualized by polarized light microscopy. In addition, the pH, viscosity and mucoadhesive properties of the formulations were also evaluated and found to be suitable for buccal application.

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Conflict of Interest

The authors declare no conflicts of interest, financial or otherwise.

Statament of Contribution of Researchers

Concept – M.G., S.Y.K., Ö.M.; Design – M.G., S.Y.K., Ö.M.; Supervision – S.Y.K., Ö.Ö.; Resources S.Y.K., Ö.Ö., Ö.M.; Materials –M.G., G.T., S.Y.K., Ö.M.; Data Collection and/or Processing – M.G., G.T., S.Y.K., Ö.M.; Analysis and/or Interpretation – M.G., G.T., S.Y.K., Ö.M.; Literature Search – M.G., G.T., Ö.M.; Writing – M.G., G.T., S.Y.K., Ö.M.; Critical Reviews – M.G., S.Y.K., Ö.M.

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