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FORMULATION AND EVALUATION OF FAST DISSOLVING FILMS EMBEDDED WITH NANOPARTICLES OF MIRTAZAPINE HYDROCHLORIDE

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ABSTRACT

Objectives: The goal of this research was to produce fast dissolving film embedded with nanoparticles of mirtazapine hydrochloride. **Method:** Semisynthetic polymer (HPMC) is used to prepare orodispersible films which is prepared by solvent casting method and nanoparticles were formed by ion gelation method from chitosan and STPP. Fourier transform infrared spectroscopy and scanning electron microscopy demonstrated that all ODF constituents were compatible with one another. Different physico-chemical, mechanical characteristics, morphological examination and in vitro drug release investigations evaluation were used to assess and evaluate ODFs. **Results:** All formulations have disintegration times of less than 70 seconds. Mirtazapine hydrochloride content consistency in all ODFs ranged from 95.7% to 99.1%. These values revealed that the ODFs were strong and flexible enough. Tensile strength of oral films measured as 10.82 ± 1.8 to 28.31 ± 1.5 , and percent elongation was 3.23 ± 0.63 to 10.04 ± 0.27 . The ODFs had a smooth surface with uniform distribution of nanoparticles and all constituents, according to scanning electron microscopy. Mirtazapine hydrochloride were released from ODFs 20 to 25 minutes, according to in vitro drug release experiments. After stability tests, no significant differences in physicochemical and mechanical parameters were found in any of the formulations. **Conclusion:** The formulation of nanoparticles of mirtazapine hydrochloride were successfully developed and embedded on fast dissolving film, which was prepared, in vitro release of film was observed for specific time.

Keywords: Orodispersible films, Mirtazapine hydrochloride, Nanoparticles, Fast dissolving film, Solvent casting method, Ionic gelation method, Polymeric film

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INTRODUCTION

Nearly 40% of all newly discovered medications are currently water insoluble, resulting in low oral bioavailability, as a result their application is limited. The main purpose of pharmaceutical research is to discover innovative technological ways for improving the solubility of medications with low solubility in water. The reduction of Particle size to the nanometer level has been proposed as a feasible method for improving dissolving. Drug nanosuspensions are colloidal dispersions of drug particles in a dispersion media with a particle size in the nm range, often between 10 & 1000 nm, stabilized by polymers, surfactants, or a combination of the two. Apart from enhancing the solubility of drugs or dissolution rate of poorly water-soluble medications, nanosuspensions carries variety of other advantages, like high drug loading, reduced excipient side effects, and low cost [1].

Nanosuspension, on the other side, is a thermodynamically unsound system, and stability is serious criteria in ensuring pharmacological nanosuspensions' protection and efficacy [2]. Storage and transportation of drug nanosuspensions can cause several stability issues, including sedimentation, agglomeration, and crystal formation. Liquid formulations of nanosuspensions that are difficult to transport, limiting the nanosuspension delivery system's use and promotion. It is process of transforming the nanosuspension to a dry powder form for purposes of physical stability and patient ease [3]. Dried form of nanoparticles must be able to convert to their initial nanosuspension form after being reconstituted in water to enhance the oral bioavailability of drugs that are less water soluble. As a result, developing solid nanoparticles with

strong redispersibility for improving the oral bioavailability of weakly water-soluble medicines is a major task [4].

Because nanosuspensions are liquid preparations that are difficult to transport, the usage and promotion of this delivery technology is limited. It is important to convert the nanosuspension to powder form for reasons of physical stability and/or patient convenience.

Orodispersible films (ODFs) are postage stamp-sized strips of thin polymeric films formulated to disintegrate or dissolve virtually instantly when placed over the tongue. Because of different pharmaceutical and pharmacological characteristics, adaptability, non-invasiveness, and compliance of patient, the orally route for drug delivery is the safest and optimal route for drug delivery when compared to other routes of and nanosuspensions formulations which are in liquid form that are difficult to transport, limiting the application or marketing of the nanosuspension delivery system. drug administration [5]. Children, the elderly, nauseated patients, and persons with specific conditions (Parkinson's, dementia, dysphagia, and so on) may find the oral route challenging [6]. Transdermal drug delivery devices, microneedles, parenteral preparations, mucoadhesive buccal films (MBFs), orodispersible films (ODFs), orodispersible tablets (ODTs), suppositories, and other options are available in such situations [7]. Each of these drug delivery techniques has its own set of benefits and drawbacks. However, ODFs and ODTs are preferred over other systems for oral mucosal drug delivery dosage forms since they have fewer drawbacks. The medication in ODFs is absorbed by the oral mucosa, bypassing first-pass hepatic metabolism and entering the systemic circulation. The medication is then swiftly disintegrated and dissolved to allow for oromucosal absorption, or with formula tweaks, the quick-dissolving components are maintained to allow for gastrointestinal absorption when eaten. Unlike other quick release dosage forms currently available, the rapid films can be made with a technique that is comparable to that of normal tablets. ODFs are preferred over ODTs by infants, children, the elderly, and special populations due to noncompliance and the risk of choking. ODFs provides number of advantages, including accurate and convenient dosing, transportability, ease of acceptance for dysphasic, pediatric, and elderly patients, relatively quick absorption, higher

stability, and enhanced bioavailability due to bypassing first-pass metabolism [8].

ODFs are good option for solidifying drug nanosuspensions after further consideration. It is not only turns drug nanosuspensions into solid products, but it also solves the problem of ODFs being ineffective for delivering medications with low water solubility. A new ODF comprising drug nanoparticles was produced in this work Furthermore, they are not universally applicable to all medications. Therefore, the medications with low aqueous solubility constitute a serious difficulty in the creation of ODFs [9].

ODFs, on the other side, are not suited for the administration of medications with low water solubility, as these compounds have poor oral mucosa drug absorption. To increase the water solubility of weakly water-soluble pharmaceuticals, self-microemulsifying mouth dissolving films were produced. Furthermore, ODFs remove the disadvantages of oral disintegrating tablets, such as their brittleness and fragility, which necessitates specific packaging for storage and transit, and their preparation via the costly lyophilization process [10].

Mirtazapine hydrochloride is a novel antidepressant drug, with poor water solubility was used as a drug. Mirtazapine hydrochloride nanoparticles were prepared by ion Gelation method and then transferred into ODFs carrying Drug nanoparticles by solvent casting method. (SEM) was used to examine the prepared ODFs. The consistency and uniformity of the content were also examined.



Figure 1: Approaches to NDDS.

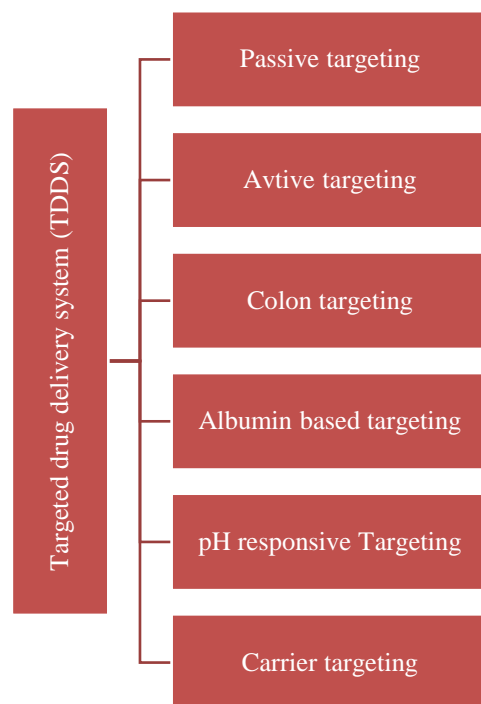


Figure 2: Targeted drug delivery system.

Objectives of Research

The main objective and purpose of this research was to design an ODF comprising the drug which has antidepressant activity. The more specific objectives and the goals of the research were, 1. To formulate and evaluate the ODFs containing nanoparticles of mirtazapine hydrochloride, 2. To improve the physico-chemical and mechanical properties of the ODFs, 3. To study the compatibility of the drugs with the excipients, 4. To highlight the effect of the ratio of polymer on the release drugs with time, 5. To study the stability of the prepared ODFs through different parameters.

MATERIALS AND METHODS

For formulation or characterization for fast dissolving films with embedded nanoparticles these chemicals are used. Chitosan, Acetic acid, Ethanol Absolute, Sodium tripolyphosphate (STPP), Sodium Hydroxide (NaOH), Potassium Dihydrogen Phosphate and Potassium Chloride were purchased from Sigma Aldrich, Germany. Hydrochloric acid 37% was purchased from AnalaR BDH Laboratory, England. Mirtazapine hydrochloride was received as gift sample from Mass Pharma Pvt. Ltd.

Preparation of Orally Dispersible Films

The preparation of orally dispersible film (ODF) was made using solvent casting method.

Preparation of ODFs

The polymer of each film formulation was dissolved or suspended in distilled water followed by the addition of other excipients like aspartame and PEG 400.

The temperature and time of the mixture was noted and stir it until it becomes a homogenized solution.

Nanoparticle's Preparation

Ion gelation was used to make nanoparticles. In step I, a chitosan (0.1 percent) solution was made by dissolving chitosan in an acidic solution (2 percent glacial acetic acid). Using a pH meter, set the pH with NaOH solution, the pH of the solution was changed from 4.6 to 5. The solution was filtered via Whatman filter paper to eliminate any contaminants or undissolved particles. In step-II Mirtazapine hydrochloride solution was prepared by dissolving Mirtazapine hydrochloride into ethanol. In step-III STPP (0.1%) solution which was prepared by dissolving in distilled water having a concentration of 1mg/ml.

In step-IV, in a 50 ml beaker, 6 ml of chitosan solution was taken on the magnetic stirrer and continue stirring at 650 to 750 rpm for 15 minutes. Then, add drug solution, which is Mirtazapine dissolved in ethanol, added dropwise by using a 1ml syringe and continue stir for 30 minutes. Then, in a 1ml syringe, 0.1 percent STPP solution was added dropwise to the chitosan solution very gently. Continue stirring for 45 minutes after adding the STPP solution to ensure complete crosslinking. A transparent solution (Nanosuspension) was formed after 45 minutes. The formulation was prepared by altering the drug concentration.

In step V, the nanosuspension was put into eppendorf tubes and centrifuged for 35 to

50 minutes at 12,000 rpm using an ultra-centrifuge equipment.

In step-VI, by draining the supernatant, at the bottom solid pellet of the eppendorf tube was taken. A minute amount of water was added up to the eppendorf tubes, vortexed them for 5 to 10 minutes using a vortex mixer to re-suspend the nanoparticles and transfer them in a petri dish. In step-VII, the materials were lyophilized for 6 to 8 hours, after which they were ground into a dry powder. At 4°C, the lyophilized substance was collected or stored.

Loading of Nanoparticles

Then nanoparticles of mirtazapine hydrochloride were loaded in film of polymer which was prepared by early. After addition of nanoparticles polymer film was placed to dry in room temperature for 24 hrs.

In vitro Evaluation of ODFs

Fourier Transform Infrared Spectroscopy

A study about drug-excipient interactions among distinct excipients of a formulation was conducted using the Fourier transform infrared (FTIR) technique. Whenever infrared rays were absorbed by various functional classes and chemical bonds inside a molecule, the result is a spectrum, which in FTIR is either transmittance or absorbance spectra [11]. The range of IR ray's spectrum wavelength range of 1000 cm⁻¹ to 3500 cm⁻¹.

Zeta Size, Potential and Polydispersity Index

When developing a dosage form, the zeta size and PDI of the nanoparticles are crucial aspects to consider. The polymeric nanoparticles should be minor enough (1 nm-1000 nm) to pass through biological membranes without difficulty. The zeta potential is the potential that exists between solid and liquid interfaces and governs the sustainability of colloidal suspensions (electrokinetic potential) [12].

Percentage Entrapment Efficiency (%EE)

An indirect method was used to calculate the polymeric nanoparticles' % EE. A suspension of nanoparticles was centrifuged at 12,000 rpm for 45 minutes. The nanoparticles in the form of solid pellet present at the bottom of the tubes. In the test tube, the supernatant remained separate out. The absorbance was assessed using a UV-spectrophotometer after the supernatant was taken & diluted by dilution factor. The following is the mathematical equation that was used to calculate the % EE:

$$\%EE = \frac{\text{Total drug loaded} \left(\frac{\mu\text{g}}{\text{ml}}\right) - \text{Free drug} \left(\frac{\mu\text{g}}{\text{ml}}\right)}{\text{Total drug} \left(\frac{\mu\text{g}}{\text{ml}}\right)} \times 10$$

Scanning Electron Microscope

Films surface morphology the was studied using a SEM (S-2380N, Hitachi, Japan). A tiny piece of the film was attached to a pad that was placed on an aluminium stub, and the Hummer sputtering system in a high vacuum evaporation system was utilized to sputter coat the film with gold.

Visual Inspection

ODF can be visually inspected to determine color, transparency, and uniformity.

Film Thickness and Weight Variation

Measurement of thickness was done with a micrometer screw gauge at three distinct sites, and the average was reported. By cutting the ODF in 3X2 cm², the weight variation of the film was investigated and using a weighing balance to calculate each film's weight. The film's thickness or weight uniformity are critical for establishing content uniformity since they are linked to dose accuracy in the ODF [11].

Table 1: Composition of orodispersible film.

| Sr No | Formulation code | Chitosan: STPP (W/W) | Mirtazapine HCl (mg) | HPMC E50 (mg) |
|-------|------------------|----------------------|----------------------|---------------|
| 1 | ODF1 | 6:1 | 15 | 20 |
| 2 | ODF2 | 6:1 | 15 | 40 |
| 3 | ODF3 | 6:1 | 15 | 60 |
| 4 | ODF4 | 6:1 | 15 | 80 |
| 5 | ODF5 | 6:1 | 15 | 100 |

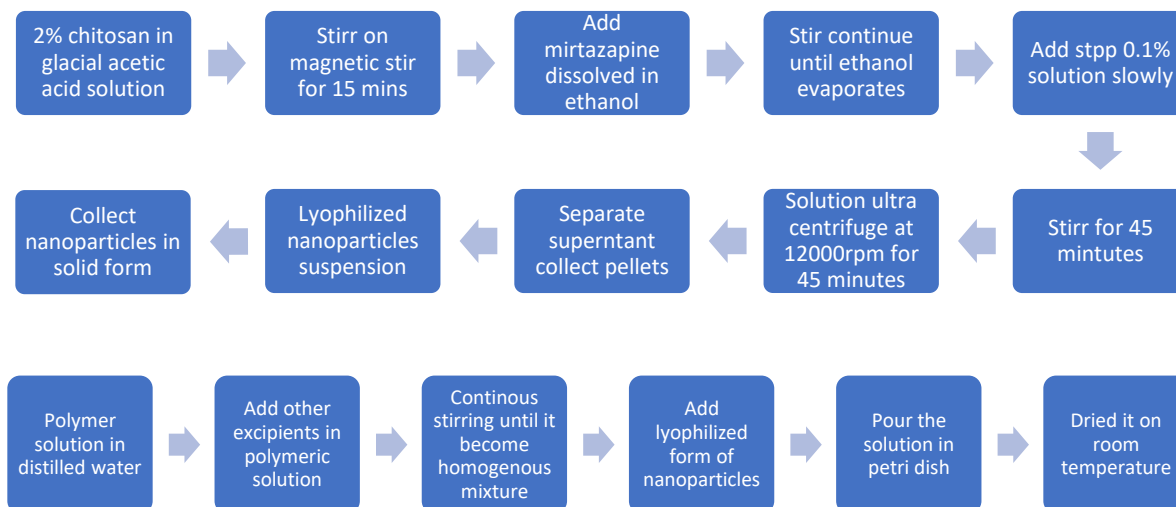


Figure 3: Scheme of preparation of nanoparticles and fast dissolving films.

Disintegration Time of ODFs

The film sample was held in a petri dish. Distilled water (2mL) was poured & gently stirred throughout. The time taken to disintegrate the film fully was recorded. The optimal period for disintegration of ODFs is < 3 minutes, which is also indicated in the European Pharmacopoeia [13]

Surface pH of the Film

Splattering distilled water (5 μ l) on the film's top and then putting the electrode of a pH meter to the film's surface were used to determine the pH. The pH of all formulations' surfaces was evaluated three taken as the average [14].

Content Uniformity

The prepared film's content uniformity is determined using the standard procedures specified in USP. Standard analytical procedures are used to check for content uniformity on twenty samples of films. According to the USP 27, content should be in the 85-115 percent range [15].

Drug Loading

Drug loading (DL) in the prepared ODFs was calculated through Eq.

$$DL (\%) = \frac{W_D}{W_{ODF}} \times 100$$

where, W_D and W_{ODF} are the weight of the drug in the ODFs and total weight of the prepared ODFs, respectively.

Entrapment Efficiency

The ODFs' entrapment efficiency (EE) was measured by Eq.

$$EE (\%) = \frac{W_D}{W_{ID}} \times 100$$

where, W_D and W_{ID} denote the weight of the drug in the ODFs and initial concentration of the drug added during the preparation of ODFs, respectively.

Moisture Uptake

The moisture uptake research of the produced film (2X3 cm²) was carried out by placing it in an environment with a relative humidity (RH) at 25 °C of around 75% for 7 days.

The weight gain of the films indicates moisture uptake and can be determined by Eq.

$$M_u = \frac{W_f - W_i}{W_i} \times 100$$

where, M_u is the moisture uptake percentage, W_f is final weight and W_i is initial weight of the film.

Loss on Drying

It is determined by taking the weight of the film earlier and after putting the film in oven at 105 °C for 1 h. The difference in weight of film was evaluated using the following formula, Eq

$$LOD = \frac{W_f - W_i}{W_i} \times 100$$

where, LOD is measured by W_f is final weight and W_i is the weight of the film at the start of the experiment.

Tensile Strength

The tensile strength of a film is measured by determining the maximal force required to break it. It was successful in determining the film's strength [16] From the applied stress on the film until the film ruptures over the cross-sectional area, a test can be developed and determined by Eq

$$TS = \left(\frac{L}{T} \times W_d \right)$$

where, TS for tensile strength, L used for load at failure, T represents strip thickness and W_d is strip width.

Percent Elongation

Strain is defined as the stretching of a film as a result of a stress. The ratio of a film's variation in length to its beginning length is called strain. The

concentration or amount of plasticizer supplied during the production of the film is measured in percent elongation. Increasing the plasticizer concentration resulted in enhanced film elongation.

$$E = \frac{L_f}{L_i} \times 100$$

Folding Endurance

Folding endurance would be another approach to evaluate a film's mechanical qualities. This is accomplished by repeatedly folding the film at the same location. The procedure was completed till the film cracked. The number of folding endurances is the maximum times in which the film can be folded without breaking. The film sample's mechanical strength will improve as the value rises.

In vitro Drug Release Study

The release of drugs of the generated ODF was investigated using the USP dissolving device II, with paddle speed set at 100 rpm. KH_2PO_4 (12 mM), CaCl_2 (1.5 mM) and NaCl (40 mM) were used to make simulated saliva. NaOH was used to adjust the pH of the resulting solution to 6.8.

Simulated saliva was used for dissolution study maintaining at 37 °C. sample was withdrawn after regular intervals of time (1,2,3,4,5,10,15 and 30 min), filter it and diluted (if required) and scanned through UV-visible spectrophotometer at 290nm for mirtazapine. Dissolution media was immediately replaced with new simulated saliva in an equal volume after the sample was taken [17].

Drug Release Kinetics

The drug's dissolution kinetics were determined using a model-based technique. These models use mathematical equations to describe the drug's dissolution characteristics. There are some models:

Zero-order Model

The model illustrates the release of drug from a non-disintegrated dosage form, which is exceedingly sluggish. A mathematical equation can be used to explain this:

$$Q_t = Q_0 + K_0 t$$

$Q_t = 't'$ is the amount of medication that has been dissolved

$Q_0 =$ The initial concentration of the medication in the solution

$K_0 =$ Constant of zero order rate

$t =$ Time of Release

The model depicts the drug release, which is independent of its initial concentration.

First-order Model

This model illustrates how release of drug from a designed dosage form is altered by the original drug concentration, the greater the initial concentration, the greater the release rate, and vice versa. The best way to describe the first-order release is to use the following mathematical equation:

$$\log C = \log C_0 - K_t / 2.303$$

$\log C_0 =$ Initially drug Concentration

$K =$ Constant of first-order release

$t =$ Drug release time

This first-order release model best describes the release kinetics of aqueous pharmaceuticals confined in a porous matrix structure. Nanoparticles releases the drug which was plotted by cumulative drug stays in the system vs time using this graphical equation.

Higuchi Model

This model depicts medication release from dosage forms that have been developed using a matrix approach. The Higuchi model can be justified using the following mathematical equation:

$$Q_t = K_h \times t^{1/2}$$

$Q_t =$ % of the drug's release

$K_h =$ Higuchi release constant

$t =$ Drug release time

The model depicts the pores that form in the matrix system, as well as the drug release via diffusion. The release of drugs from the formulation is described by Fick's law of diffusion.

Hixson-Crowell Model

Hixson and Crowell proposed an equation that characterizes particle's area as being directly proportional to the cubic root of the volume. The Hixson-Crowell release model is described by the following mathematical equation:

$$W_0^{1/3} - W_t^{1/3} = K_{HC} t$$

$W_0 =$ Initially drug concentration

$W_t =$ The drug's concentration remains in dosage form.

$K_{HC} =$ Rate constant of Hixson-Crowell

$t =$ Drug release time

This model revealed release of drug caused by changes in the diameter and particle surface area of the dose form. The percent cubic root of release of drug vs time was plotted in the graphical representation of this model's release experiments.

Korsmeyer-Peppas Model

The drug release kinetics from a polymeric system were described by Korsmeyer and Peppas.

The following is a mathematical equation that represents this model:

$$\frac{M_t}{M_\infty} = K_{kp} \times t^n$$

$M_t/M_\infty =$ At a certain time 't', the drug release fraction

$K_{kp} =$ Rate constant of Korsmeyer-Peppas

$n =$ Drug release Exponent

In this model, the release of drug process is described by the drug exponent 'n'.

RESULT AND DISCUSSION

ODFs Preparation using Solvent Casting Method

The ODF's prepared by solvent casting method. In this method a polymeric solution is prepared and then nanoparticles were prepared by ion gelation method. Solid form of nanoparticles of drug mirtazapine hydrochloride were embedded into polymeric film. Then dried form of prepared film was cut down and peeled of pieces of 3x2 cm² having 15mg of drug in every piece of film Fig 4. Then these prepared films are further characterized.

Particle Size Analysis using Zeta Sizer

The size of the nanoparticles is 266.4nm, PDI 0.549 and zeta potential is 10.5 ± 3 . The stability of the formulation is influenced by the zeta potential of the nanoparticles; the higher the zeta potential value, the more stable the formulation will be as the repulsion between the nanoparticles increases.

The size of nanoparticles and zeta potential is same as reported in the literature, the prepared nanoparticles were evaluated for physicochemical properties, percent entrapment efficiency [18].

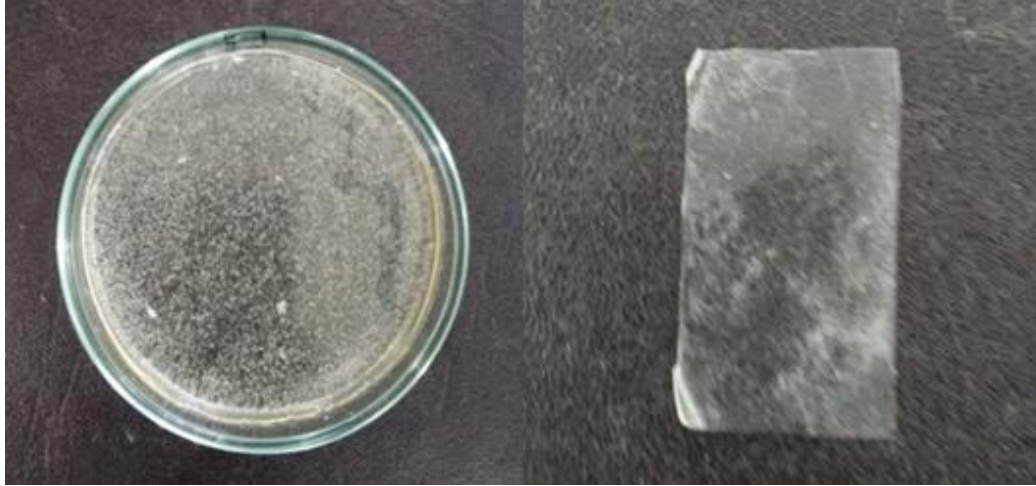


Figure 4: Orodispersible film (a); 3x2 cm² piece of film (b).

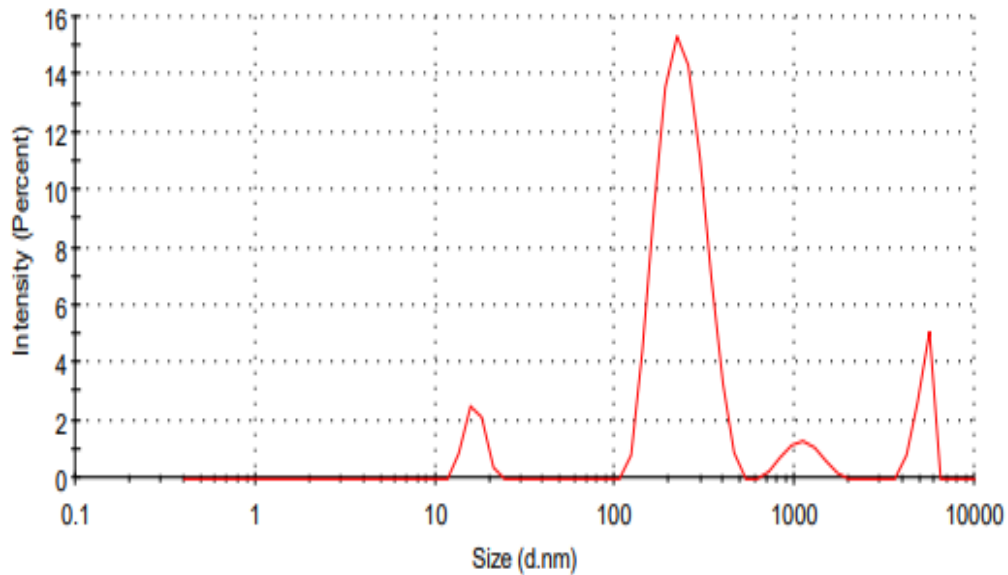


Figure 5: Particle size distribution of formulation.

Percentage Yield and Percentage Entrapment Efficiency (%EE)

The % yield of nanoparticles was calculated by the given formula

Actual yield / Theoretical yield

While the %EE of mirtazapine hydrochloride nanoparticles was calculated by using the indirect method. The entrapment efficiency of nanoparticles was 72.53%.

FTIR Study

The FTIR spectra of all ODF components, including drugs, polymers, and excipients, as well as various formulations, were obtained using the KBr pellet technique and are displayed in Figure. The FTIR spectrum of optimized formulation revealed that none of the constituents in the developed formulation had any physical or chemical interactions. The FTIR spectra of formulation with the drugs and respective polymers can be observed in (Fig. 6). Similarly the FTIR spectra of active and inactive ingredients and all formulations are shown in for better representation. After examining the FTIR spectra of these formulations, it was determined that the proposed technique of preparation had no physical or chemical interactions between the formulation's components, and that all substances were compatible.

The most relevant IR bands [(O-H stretching, CH₃ stretching, and pyridine ring stretching vibrations)] were allocated to the mirtazapine hydrochloride FT-IR and Raman experimental data with the goal of discovering the coordination effects. This study looked at mirtazapine's theoretical infrared spectrum (between 1600 and 1000 cm⁻¹) [19]. The FTIR spectra of HPMC E50 have shown the specific bands of -CH₂ at 2900 cm⁻¹, -OH from 3500-3300 cm⁻¹, -CO from 1650-1600 cm⁻¹, methoxy group from 1500-1450 cm⁻¹ and -C-O-C from 1300-1250 cm⁻¹. All the bands in the spectra are very similar to the carbohydrate area [20]. PEG 400 and aspartame FTIR spectra are like those previously reported in the literature. In FTIR spectrum of aspartame At 1720 cm⁻¹, a significant absorption band appeared, which is a characteristic vibrational stretching of the carbonyl moiety's ester. C-N stretching of secondary amide is the band that appeared at 1552 cm⁻¹. C-O and the stretching of C-C are regarded as peaks that arose between 1200 and 1400 cm⁻¹. The 687 cm⁻¹ peak corresponds to the C-H of an aromatic ring. Aryl ring distortion is the band that appears around 990 cm⁻¹. At 3428 cm⁻¹, vibrational stretching of NH₂ is visible [21]. The chitosan powder was scanned at wavelengths ranging from 4000 cm⁻¹ to 500 cm⁻¹. The amide NH group's symmetric

vibration is indicated by the band peak at 3369cm⁻¹. The characteristic vibrations of the C-H bond can be seen in the peak value of 2927 cm⁻¹. The bending vibration of NH₂ is demonstrated by the chitosan's sharp peak absorption at 1591 cm⁻¹. At 1339 cm⁻¹, the vibrational peaks of C-H can be seen. The peak of saccharides within the chitosan structure is at 1154 cm⁻¹ and 895 cm⁻¹, according to the structure of saccharides within the chitosan structure. The larger peaks at 1080 cm⁻¹ are shown by the stretching vibration of C-O [22]. The STPP pure powder was scanned at wavelengths ranging from 4000 cm⁻¹ to 500 cm⁻¹. The functional group PO has a characteristic stretching vibrational peak at 1248 cm⁻¹, while the functional group PO₂ has a peak at 1162 cm⁻¹. The vibrations of functional group PO₃ can be seen in the stretching peaks at 1079 cm⁻¹. The STPP structure has a distinct sharp peak at 885 cm⁻¹ implies the presence of P-O-P bonds the functional division.

DSC Analysis

The thermogram also indicated a very high degradation temperature and transition temperature which proved the thermal stability of these prepared formulation (Fig. 7). The presence of endothermic peaks in formulations can be observed distinctively which indicated that all components of a formulation are compatible with each other [23-24].

Morphological Studies of ODFs

Visual inspection for any imperfections and shape or distribution of the components, as well as SEM, were used to examine the morphology of the prepared ODFs. Photographs of all oral film formulations are shown in (Fig. 8). The surface of ODFs appears to be smooth in these images, with no obvious flaws or imperfections. The reasonably smooth surface of the films also reveals an equal, homogeneous, and uniform distribution of all constituents.

SEM image of orodispersible formulation (HPMC E5, a semisynthetic polymer) formulation is shown in (Fig. 4) which also showed a relatively smooth surface of the film. It is a polymer with good properties which keeps all the components smoothly and joined together. Because the nanoparticles which are made with chitosan and STPP were cross-linked with one another with time due to the charge on their surface charges, SEM showed unevenly shaped particles. With the passage of time, the STPP gets crystallized, altering the shape of the nanoparticles. These nanoparticles became linked to one another as a result of the ions, forming aggregates of nanoparticles that produce an uneven pattern under SEM.

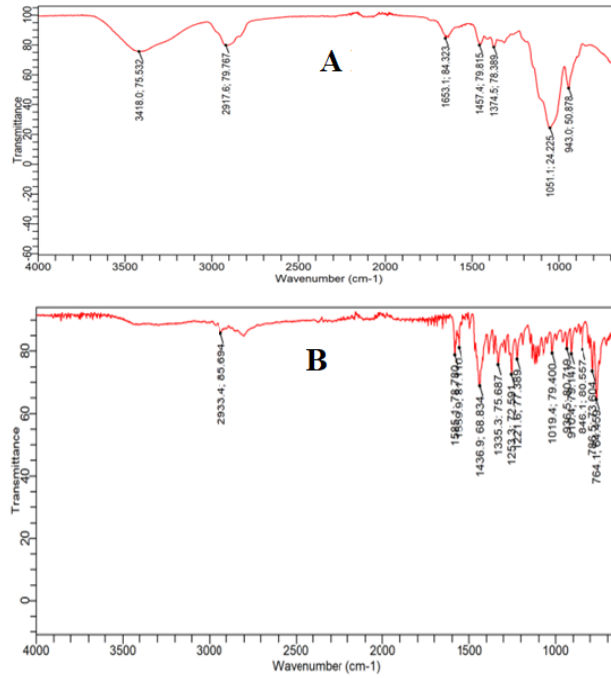


Figure 6: FTIR spectra of formulation.

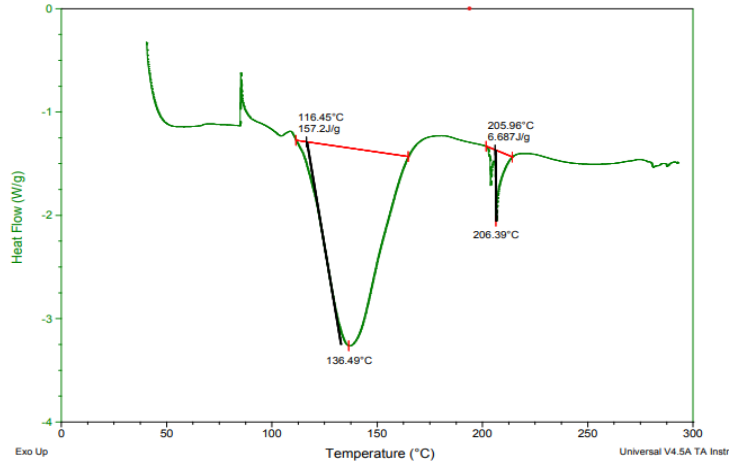


Figure 7: DSC thermogram of formulation.

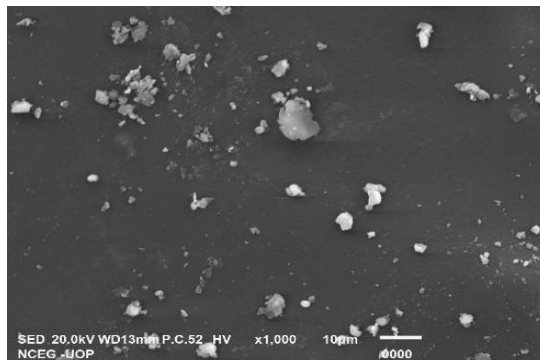


Figure 8: Surface morphology of orodispersible film using SEM.

Physico-chemical Properties of ODFs

Film thickness, disintegration time, weight and pH of the film surface were used to describe ODFs. All essential parameters of ODFs are demonstrated in (Table 2). Thickness and weight of all ODFs are in the range of 70- 156 μm and from 54-134 mg respectively. Disintegration time was observed and noted. All formulations disintegrated in less than or equal to 70 seconds, within 3 minutes, the orodispersible formulation should disintegrate. It was observed that when the concentration of polymer increases the percentage of drug release and disintegration time also increases, but in low concentration of polymer the film is not showing the good flexibility [15]. As a result, the disintegration time values recorded for all ODFs were within the normal range. which is comparable with ODF standards and published literature [13]. The pH of

ODF's surface ranges from 6.33 to 7.4, which is reported in early studies [14].

Mechanical Parameters of ODFs

(Table 3) shows all of the mechanical parameters of ODFs like Tensile strength, % elongation, folding endurance, moisture uptake, moisture loss, loss on drying, and content uniformity of all formulations. The mechanical strength of the film sample will increase as the value increases. Folding endurance is the direct relation with mechanical strength. Tensile strength of different formulations measured as 10.82 ± 1.8 to 28.31 ± 1.5 and percent elongation was measured as 3.23 ± 0.63 and 10.04 ± 0.27 , as the HPMC ratio increases in the formulations it exhibited high tensile strength which shows that relatively hardness and brittle nature of formulations [25].

Table 2: Different parameters of prepared orodispersible films.

| Parameters | ODF1 | ODF2 | ODF3 | ODF4 | ODF5 |
|----------------------------|-----------------|-----------------|------------------|-------------------|-----------------|
| Weight (mg) | 54.5 ± 0.55 | 76.23 ± 0.7 | 97.9 ± 3.2 | 116 ± 1.2 | 134 ± 0.16 |
| Thickness(μm) | 70.66 ± 2.4 | 86 ± 2.64 | 100.67 ± 2.1 | 127.66 ± 2.51 | 155.2 ± 3.1 |
| Disintegration time | 45 ± 2.5 | 48.0 ± 1.52 | 55.7 ± 3.02 | 60.3 ± 1.2 | 70.0 ± 2.8 |
| pH of film surface | 6.33 ± 0.15 | 6.98 ± 0.13 | 7.01 ± 0.2 | 7.07 ± 0.15 | 7.4 ± 0.2 |

All values are expressed as mean \pm SD.

Table 3: Mechanical properties of ODFS.

| Parameters | ODF1 | ODF2 | ODF3 | ODF4 | ODF5 |
|-------------------------------------|------------------|------------------|------------------|------------------|------------------|
| Tensile strength (N/m^2) | 10.82 ± 1.8 | 14.62 ± 2.1 | 20.9 ± 2.3 | 26 ± 1.9 | 28.31 ± 1.5 |
| % elongation (%) | 3.23 ± 0.63 | 5.34 ± 0.54 | 7.94 ± 1.5 | 8.76 ± 0.32 | 10.05 ± 0.27 |
| Folding endurance | <20 | <20 | <20 | <20 | <20 |
| Content uniformity (%) | 97 ± 2.5 | 98.3 ± 1.52 | 97.7 ± 3.02 | 98.3 ± 1.2 | 99.01 ± 2.8 |
| Drug loading (%) | 15.03 | 15.3 | 15.14 | 14.9 | 15.4 |
| Entrapment efficiency (%) | 95.7 | 98.5 | 97.5 | 98.2 | 99.1 |
| Moisture uptake (%) | 1.97 ± 0.01 | 2.01 ± 0.04 | 2.11 ± 0.2 | 2.76 ± 0.33 | 3.26 ± 0.3 |
| Moisture loss (%) | 0.97 ± 0.01 | 1.13 ± 0.15 | 1.41 ± 0.07 | 1.73 ± 0.05 | 2.82 ± 0.10 |
| Loss on drying (%) | 0.023 ± 0.02 | 0.026 ± 0.04 | 0.033 ± 0.02 | 0.046 ± 0.02 | 0.141 ± 0.04 |

These values are in line with industry standards or those described in the literature. The drug loading and entrapment efficiency of mirtazapine hydrochloride in the constructed ODFs are shown in the table below. Drug loading in the range of 14.9 to 15.4 mg and the entrapment efficiency of the mirtazapine hydrochloride was determined as 95.7% and 99.1%.

In vitro Drug Release Studies

The graph shows the release studies of mirtazapine in the simulated saliva used as dissolution media. It was shown that nearly total drug release from all formulations was achieved in < than 25minutes. Mirtazapine was estimated using a UltraViolet spectrophotometer by measuring absorbance at 288nm in a pH 6.8 phosphate buffer, & this method was used in the study which is reported in literature mirtazapine show its absorbance at same wavelength. Different formulations which show their release at 0,5,10,15,20,25, and 30 minutes sample was taken and analyze with UV spectrophotometer, which shows the similar release in all formulations. Drug releases from the orodispersible film is between 15 to 20 minutes according to the studies [15].

Drug Release Kinetics and Mechanism

For mirtazapine hydrochloride nanoparticles, the values of different release kinetic models are expressed in (Table 4). The drug release data was evaluated using DDSolver. In the Korsmeyer-Peppas model for mirtazapine hydrochloride the

maximum value (1) of regression coefficient (R²) was noticed, indicating that the first order and Higuchi models were the best fit for representing the drug release kinetics of drug from ODF formulations.

The values of diffusion coefficient (n) for mirtazapine hydrochloride calculated from Korsmeyer-Peppas equation was found as n 0.553 ,0.584, 0.624, 0.635 and 0.634. The non-Fickian diffusion of drug from all ODFs was directed by these values of n, i.e., drug was released through swelling and diffusion mechanisms [11].

Histopathological Evaluation of ODFs

When provided the optimal formulation, i.e., ODF5 at 1 g/kg body weight of the animal, histopathological investigations of the vital organs demonstrated that newly produced formulations are safe. With respect to control group, essential organs tissues of treated animals showed no signs of deterioration or lesions. (Fig. 10) shows photographs of the rabbit's vital organs after administration of the formulation. Histopathology of vital organs is shown in (Figure 10). This study revealed that the newly synthesized formulations and with its ingredients are totally safe and don't have any significant toxicity. There are two groups group I as a controlled group and group II as experimental group, and no remarkable difference was found between both groups of animals.

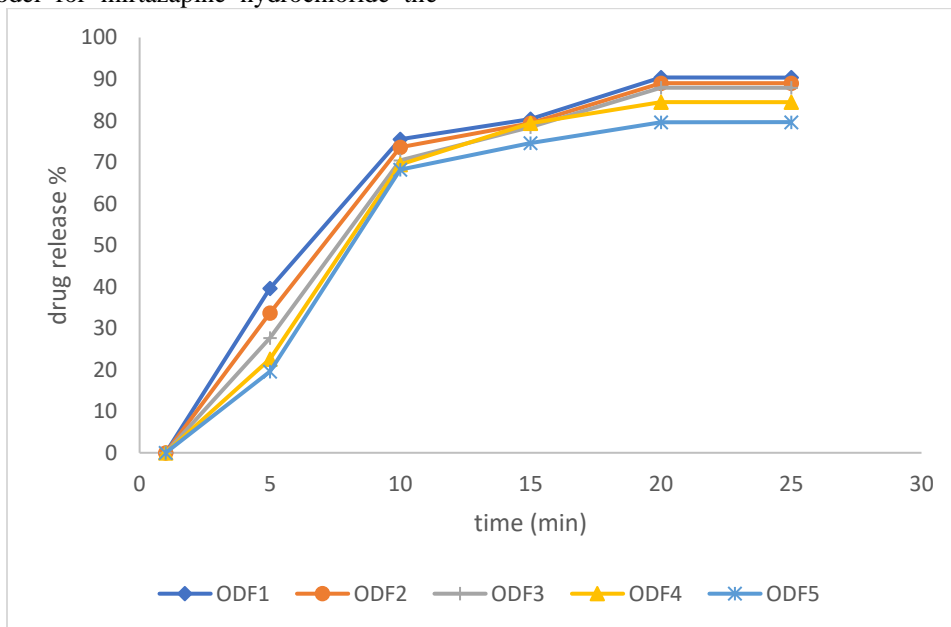


Figure 9: Percent drug release in all formulations.

Table 4: Kinetic release models for different formulations.

| Models | Key values | ODF1 | ODF2 | ODF3 | ODF4 | ODF5 |
|------------------|-----------------|--------|--------|--------|--------|--------|
| Zero order | R ² | 0.7741 | 0.7546 | 0.7684 | 0.7497 | 0.7423 |
| | K _o | 4.513 | 4.416 | 4.321 | 4.275 | 3.961 |
| First order | R ² | 0.9685 | 0.9632 | 0.9563 | 0.9368 | 0.9204 |
| | K ₁ | 0.141 | 0.104 | 0.096 | 0.090 | 0.081 |
| Higuchi | R ² | 0.8965 | 0.8922 | 0.8599 | 0.8588 | 0.8849 |
| | K _H | 19.660 | 19.187 | 18.767 | 18.584 | 18.332 |
| Hixson-Crowel | R ² | 0.9602 | 0.9573 | 0.9568 | 0.9345 | 0.9102 |
| | K _{HC} | 0.030 | 0.028 | 0.026 | 0.025 | 0.022 |
| Korsmeyer-Peppas | R ² | 0.9011 | 0.9027 | 0.9051 | 0.8815 | 0.8640 |
| | K _{kp} | 16.940 | 15.190 | 13.729 | 12.558 | 11.974 |
| | N | 0.553 | 0.584 | 0.624 | 0.635 | 0.634 |

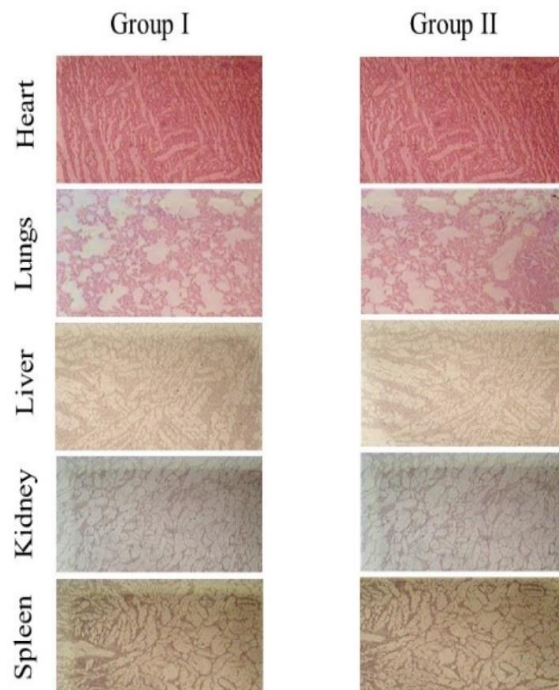


Figure10: Histopathology of vital organs.

CONCLUSION

Orodispersible film (ODF) embedded with nanoparticles of mirtazapine hydrochloride has been successfully developed using HPMC with different concentration ratios by solvent casting method. Nanoparticles were formed by ion gelation method and then embedded on Polymeric film. After

addition of nanoparticles in polymeric solution the film is dried and then cut down into 2x3cm². The disintegration time of fast-dissolving film was in water within 70s with reconstituted nanosuspensions particle size of optimized formulation was 266.4±11.57nm, indicating that the ODFs comprising mirtazapine hydrochloride

nanoparticles had good redispersibility. Smooth, free of rough, without beads, and non-porous surfaces were visible on the prepared fibers. Overall, the prepared of ODFs has the potential to be a suitable alternative to traditional oral dosage forms. The FTIR of optimized formulation shows the good compatibility of components of film, while SEM analysis shows that the dispersion of nanoparticles in polymeric film. The subsequent ODFs shows that far faster dissolution rates, the release profile of drug within 20 to 25 minutes. In vitro drug dissolution investigations demonstrated the behavior of

nanoparticles and the release of drug contents at simulated pH levels. Different release kinetic models that describe the method and the regarding drug release from nanoparticles were applied to dissolution data. Histopathology of vital organs does not show any toxicity, formulation and its components are safe to use. According to the findings, ODFs containing medication nanoparticles may present a possible option for converting drug nanosuspensions into solid dosage forms while also enhancing oral absorption of poorly water-soluble drug's bioavailability.

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