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Enhancement of solubility of antifungal Ketoconazole By Co-crystallization and Microemulsion

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M.Sc. Thesis

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Enhancement of solubility of antifungal Ketoconazole By Co-crystallization and Microemulsion

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Dedication

This thesis is dedicated to:

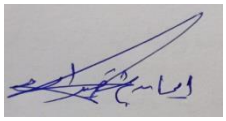
The sake of Allah, Our Creator and Master, the teacher of teachers, Prophet of humanity Mohammad -peace be upon him.

My husband, my mother, brothers and sisters, who support, and encourage me to the end of this great achievement, all thanks and gratitude to them.

Eman Ahmad Shqirat

Declaration

I certify that this thesis is submitted for the degree of Master graduation in applied industrial technology is my own research.

Signed: 

Eman Ahmad Jameel Shqirat

Date: 06/06 /2020.

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First and foremost praise be to Almighty Allah for giving me courage, determination and guidance in my life and never less in my challenging academic journey and for sending his Prophet Muhammad (PBUM), the teacher of teachers, and the mercy to the whole world.

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Abstract

This study aims to synthesize and characterize cocrystal of ketoconazole to improve solubility properties of ketoconazole. Ketoconazole cocrystal synthesized by the solvent crystallization method and characterized using FTIR spectroscopy,DSC, melting point and solubility analysis. Synthesized co-crystal have wonderful crystals. FTIR spectrum show hydrogen bonding in the co-crystal, which gives it better physical properties (such as solubility, and melting point) compared to single component. Melting point was 146.0°**C** - 148.0°**C** For the single API, after co-crystallization, it became (141.0 - 143.0 C) and (115.0-119.0 °**C**) for subreic acid and sebacic acid co-crystals respectively .The ketoconazole co-crystal resulted in an improved solubility compared to ketoconazole. This research demonstratesthat co-crystallization between ketoconazole and suberic acid, sebacic acid and different co-solvent were effectively formed and enhanced the solubility of the drug.

In addition, this research aims to prepare microemulsion of ketoconazole with a different formulations applied using the minimal amount of surfactant at ambient temperature by titration method.The effect of different percentages of co-surfactants &co-solvent on the phase behavior of the systems suggested at ambient temperature were also studied. The surfactant used in this research was Polyoxyethylene sorbitan monooleate (Tween 80). Short chain alcohol used as a co-surfactant was ethanol. The oil phase used was oleic acid. The aqueous phase was double distilled water with propylene glycol acting as co-solvent. The formulation which showed the area covered under the microemulsion was found to be larger in presence of higher amount of surfactant than co-surfactant.The surfactant /co-surfactant ratio (3:1) was found to be more efficient than the ratio (1:1) in formation of isotropic microemulsion region and produced liquid crystal region. The formulation which has propylene glycol as co-solvent gave large area of liquid crystal and microemulsion compared with the absence co-solvent.

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Abbreviations, Symbols and Terminology

DSC	Differential Scanning Calorimeter
3D	Three Dimension
HLB	Hydrophile Lipophile Balance
L1	Spherical normal micelle
L2	Reversed micelle
CMC	Critical Micelle Concentration
O/W	Oil in Water
W/O	Water in Oil
API	Active Pharmaceutical Ingredient
FTIR	Fourier Transform Infra-Red
PG	Propylene Glycol
USFDA	United States Food and Drug Administration
EAFUS	Everything Added to Food in the United States

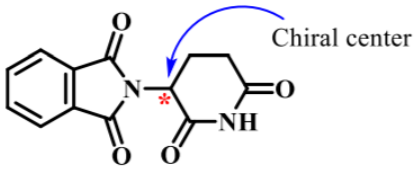
Chapter One

Introduction

1. Introduction

1.1. Structure-property relationship in chemistry

The very essence of chemistry, the periodic table, provides a systematic classification of all known elements into different periods and groups to help chemists in rationalizing the properties of these elements based on their structure at an atomic level. Moving from the atomic to the molecular level, a pioneering investigation conducted in 1947 on saturated hydrocarbons recognized that the properties of these compounds varied with molecular bulk and branching.[1],[2]One of the many examples of structure-property relationship in molecules comes from the enantiomers of thalidomide.



It turns out that one enantiomer has effective antidepressant properties while the other is teratogenic.[3]The property difference is directly connected to the chirality of the molecular structure.

For bulk materials, structure-property relations fall into two broad categories.[4]Figure 1.1. In the first, the property alterations are due to variations in molecular structure and molecular conformation. In the second category property alterations arise from differences in the spatial relationships between molecules in the crystal, such as intermolecular arrangement, interactions and packing of individual building blocks with respect to each other in a crystal lattice. Thus, in 2 order to manipulate the properties of bulk materials, either alterations of molecular structure and conformations or refinements in packing arrangements are required.

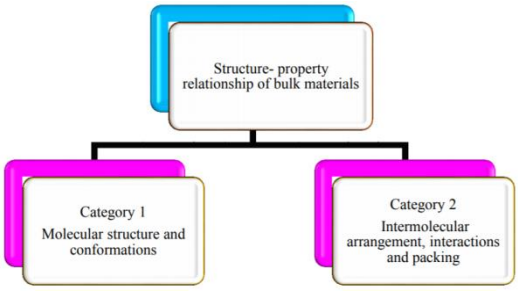


Figure 1.1 Categorization of structure-property relationships in bulk materials

The best examples which show property alterations under both above categories are polymorphs. A polymorph is defined as *"Different crystalline forms of the same substance"*.[5]Polymorphs of a substance can be attained in two ways (Figure 1.2), if the molecule can have different conformations and if the molecule is flexible, polymorphs can be attained by conformational changes (conformational polymorphism) and if the molecule is rigid, polymorphs can be attained by changing the packing arrangements (packing polymorphism).[6]Conformational polymorphs give rise to property alterations under category 1 and packing polymorphs under category 2, Figure 1.2.

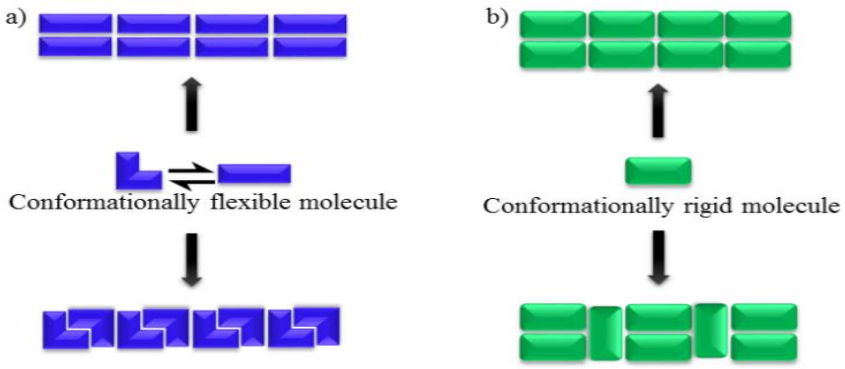


Figure 1.2 Schematic representation of a) conformational polymorphism and b) packing polymorphism

Ritonavir provides an example of conformational polymorphism where form 1 is comprised of beta like stacks of ritonavir molecules held together via hydrogen bonding while in form II, OH groups are participating in hydrogen bonding due to the change in conformation of ritonavir molecule. As a result, the solubility of the two formisdistinct from each other. Form 1 has 90 mg/mL of solubility in a 99:1 ethanol water mixture at 5 °C and form II has only 19 mg/mL of solubility under same conditions,Figure 1.3.

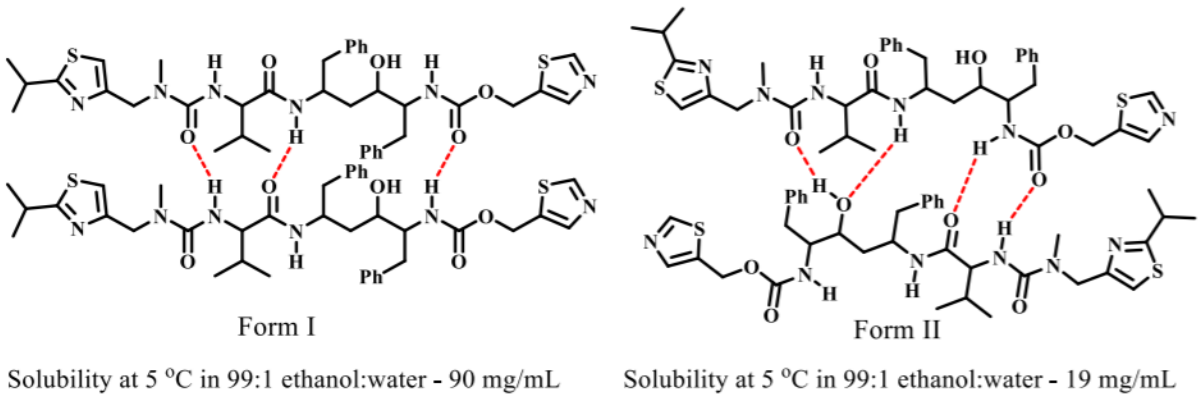


Figure 1.3 Conformational polymorphs of ritonavir and their solubilities

Polymorphic forms of aspirin provide an example of packing polymorphism. Both form I and II consist of layers of aspirin molecules with same conformation and similar interactions, and the subtle difference between the two forms is the position of the molecules in one layer relative to the molecules of the adjacent layer, Figure 1.4.[7]

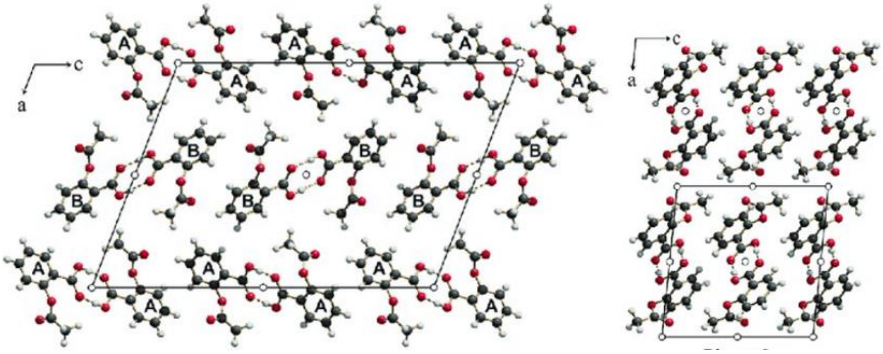


Fig 1.4 Packing polymorphism of aspirin form I and Form II

1.2 Supramolecular chemistry

A way to modify space arrangement relationships of a moleculeSupramolecular chemistry has been defined as"…the chemistry of the intermolecular bond, covering the structures and functions of the entities formed by association of two or more chemical species…" by Jean-Marie Lehn. [8] In simple terms, supramolecularchemistry refers to chemistry beyond molecules and focuses on chemical systems composed of molecular subunits brought together via intermolecular interactions[9],[10],[11]. One of the underlying goals of supramolecular chemistry is to control assembly and arrangement of individual components in a predictable way and thus to control macroscopic properties of materials. Because supramolecular chemistry can lead to modifications in the spatial relationships of a molecule of interest, it directly affects macroscopic properties.

1.2.1 Solubility

Solubility is a fundamental physicochemical property impacting various stages of drug discovery process, for example: target identification, lead optimization, preclinical and clinical trials. Solubility plays a unique function in the design and development of different

dosage forms for numerous routes of administration, such as oral, dermal, parenteral and pulmonary.

Poor water soluble drugs tend to have limited and variable bioavailability, Low solubility and dissolution rate affect many pharmaceuticals therapeutic efficacy, and significantly lower a drug's market value.In the design of new solids, particularly in the pharmaceutical field, multi-component crystals such as solvates, hydrates, co-crystals, salts play a major roleto enhance the solubility[12].

1.3 Co-crystal: A supramolecular entity

The definition of what a co-crystal is, remains under debate.[13]Thus, for the scope of this thesis, a co-crystal will be defined as, solids that are crystalline single phase materials composed of two or more different molecular compounds generally in a stoichiometric ratio. In simple terms, a co-crystal is comprised of two or more distinctly different molecular entities known as co-formers that are brought together via non-covalent interactions within the same crystal lattice. This is done without making or breaking any covalent bonds, thus the chemical integrity of each individual molecule is preserved. The end result is the formation of a heteromeric species via co-crystallization over the natural tendency to form homomeric species via re-crystallization. Figure1.5Thus, a co-crystal can be recognized as a supramolecular entity which enables the modification of physical properties of a material by changing the spatial relationships between molecules in the crystal.

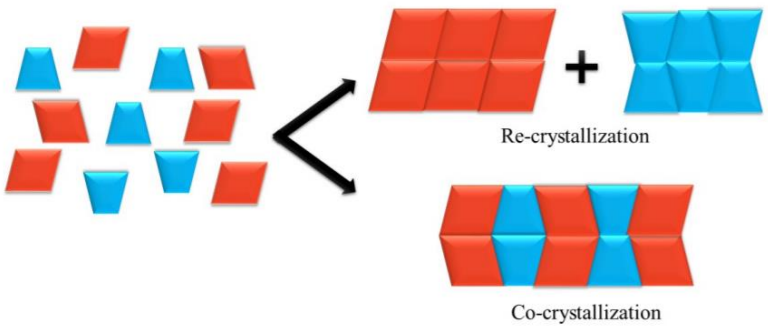


Figure. 1.5 Schematic representation of re-crystallization and co-crystallization

1.3.1 Intermolecular interactions: design of co-crystals

Co-crystals contain periodic assemblies of molecules connected via intermolecular interactions such as π-π stacking interactions[14],[15],[16],[17],dipole-dipole interactions[18],halogen bonds[19]and hydrogen bonds[20],[21],Figure 1.6 Knowledge of the intermolecular interactions and their effects on crystal packing allows for the engineering of co-crystals with desired properties.

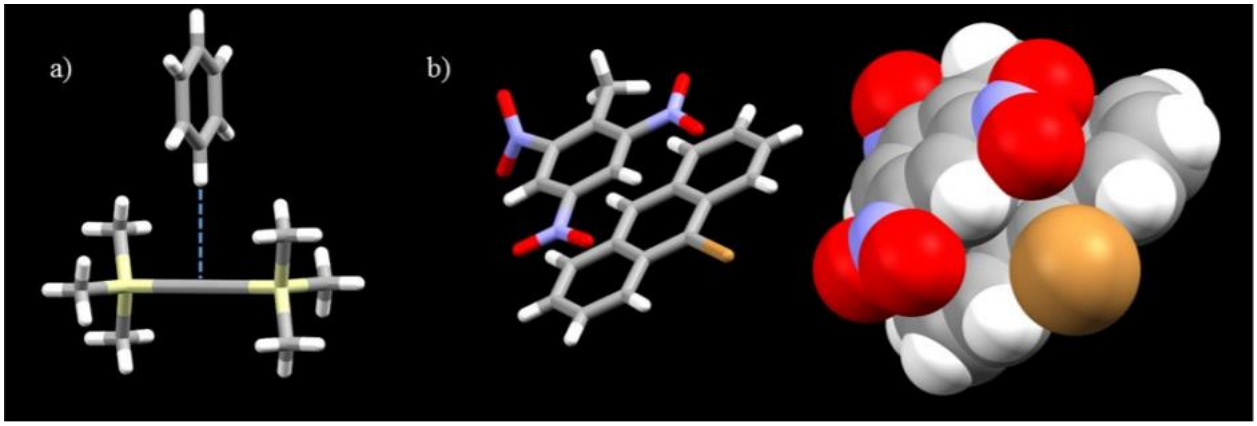


Figure 1.6 a) CH···π interactions in the co-crystal of bis(trimethylsilyl)acetylene with benzene[22],[23]and b) π-πinteractions in the co-crystal between trinitrotoluene (TNT) and 9-bromoanthracene[24]

1.3.2 Hydrogen bond based co-crystals

A hydrogen bond (H.B) is defined as "an attractive interaction between a hydrogen atom from a molecule or a molecular fragment, X−H···A in which X is more electronegative than H, and an atom or a group of atoms in the same or different molecule, in which there is evidence of bond formation"[25].

Figure 1.7 Schematic diagram showing the formation of a hydrogen bond (X-more electronegative atom than hydrogen; H-hydrogen; A-acceptor) .

Hydrogen bonds are useful tools for assembling supramolecular structures because they are both strong and directional and arguably the most widely used interaction for constructing cocrystals[26],[27],[28]Figure 1.4. In order to recognize the preferred connectivity patterns, the hydrogenbond rules proposed by Margaret C. Etter are very useful[29].

The general rules are:

1. All acidic hydrogens available in a molecule will be used in hydrogen bonding in the crystal structure of that compound[30].
2. All good acceptors will be used in hydrogen bonding when there are available hydrogen-bond donors[31].
3. The best hydrogen-bond donor and the best hydrogen-bond acceptor will preferentially form hydrogen bonds to one another[32].

These guidelines have provided the stage for important advances in co-crystallizations.

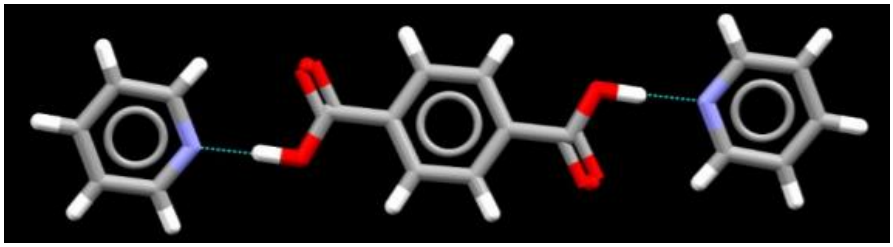


Figure 1.8 A binary co-crystal between pyridine and terephthalic acid formed via acid···pyridineheterosynthon[33]

A cocrystal is a crystal consists of more than one component which all components are solid stateat ambient conditions when in their pure form. Which all components accompanied as a stoichiometric ratio of a cocrystalformer,and depended on hydrogen bond or non-covalent forces between active pharmaceutical ingredient (API) and co-former , also it is used todescribe the hydrogen bonded molecular arrangements between two separate molecular entities .Cocrystals can increase solubility and dissolution rate of the drug as they are stable .[34],[35].

1.3.3 Co-crystallization

Co-crystalizationistheassociation between active Pharmaceutical ingredient (API) and coformer which are similar molecules (or homomers), and different molecules (or heteromers). Hydrogen bonds are the basis of molecular identification phenomena in pharmaceutical systems and are responsible for producing families of molecular networks in crystalline state with the same molecular components (single component crystals and their polymorphs) or with different molecular components (multiple component crystals or co-crystals)[36].

1.4 :Pharmaceutical co-crystal

Pharmaceutical co-crystal is an unique crystalline solid that contains two neutral molecules, one of which is an API and the other a co-crystal formers. Co-crystal former may be an excipient or another drug[37] . The cocrystalcomponents are neutral molecular reactants which are solids at ambient temperature. They present in definite stoichiometric ratio of solids that interact through noncovalent interactions, predominantly hydrogenbonds[38] which is used to define and create new formulation of drugs and offer a chance to increase the number of forms of an API. Pharmaceutical co-crystal formation improved the performance of a drug known to have poor solubility.[39]

A lot of cocrystals have been designed to improve physicochemical properties of APIs such as solubility/dissolution , chemical stability , and bioavailability.[40]

Co-crystal synthesis has been performed using a lot of methods for example: slow solvent evaporation, slurry conversion, solid-state and solvent drop grinding, melt and sublimation, electrospray technique .Most of these techniques have their own limitations because they often lead to crystallization of individual components, the requirement of complicated manufacturing process, specialized equipment, require large amount of raw materials, regulatory complications and are time consuming(30) .Solvent crystallization method

involves some other techniques for producing super-saturation beyond the large concentration ratios of co-crystal components of the solution.

1.5 :Co-former selection

Coformer is one of the components in pharmaceutical cocrystal which is considerable counter-molecules in cocrystallike pharmaceutical excipients. The API and coformer held together by hydrogen bonding .also Coformer play important role in cocrystal which is to provide stability for it. The coformer should be non-toxic, having no adverse effects and should be included on the USFDA ,EAFUS list.[41],[42]

Coformers are also chosen based on functional groups that are capable of providing complimentary hydrogen bonding with the API. Hydrogen bonds affect molecular recognition mainly because their direct interactions.

One of the biggest challenges in the production of cocrystal pharmaceuticals is the selection of co-formers compatible with a specific API.

Carboxylic acid is known as strong electron acceptors and donors of electrons, and are also well involved in supramolecular reactions. Carboxylic acids also fall under the GRAS (Generally Recognized as Safe) category as specified by the FDA (Food and Drug Administration). And are cocrystal formers to be combined with an API.

A general approach to coformer selection is by “tactless” cocrystal screening, whereby a predetermined library of pharmaceutically acceptable/approved compounds is used to attempt cocrystallization[43]

1.6 Chemicalstructure of ketoconazole

Ketoconazole , cis-1-acetyl-4-[4-[[2-(2,4- dichlorophenyl)-2-(1-H-imidazole-1-yl methyl)-1,3-dioxolan-4-yl] methoxy] phenyl] Piperazine .

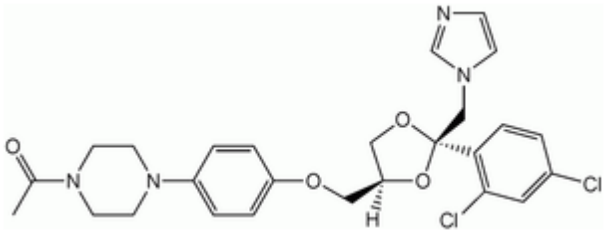


Fig. 2: Molecular structure of KTZ

is a broad-spectrum imidazole antifungal agent and administered by topical and oral . Being a BCS class II drug , the chemical structure of ketoconazole has a visible hydrophobic site and weak basicity that promotes to its poor aqueous solubility[44].

KTZ commercially available in several dosage forms for oral and topical administration. KTZ is a weak base and it can be soluble only under extremely acidic conditions. It is classified in the Biopharmaceutics Classification Scheme (BCS) as a Class II drug, since it has a high permeability but low solubility in water (0.087 mg/l at 25 °C).[45]

Besides the low, variable bioavailability, the risk of hepatotoxicity led to the re-evaluation of the oral drug for the risk/benefits effects followed by restriction of its therapeutic indications. An increase of the ketoconazole bioavailability would lower the dose required for the therapeutic effect, decreasing in the same time the risk of hepatotoxicity[46].

1.6.1 Solubility of ketoconazole

In **Table #1**which provides the solubility requirements according to the general United State Pharmacopeia.

Classification	**Parts of solvent required for one part of solute (between 15˚C and 25˚C)**
Very soluble	Less tha**n** 1part
Freely soluble	1- 10 parts
Soluble	10 – 30 parts
Sparingly soluble	30 – 100 parts
Slightly soluble	100 – 1000 parts
Very slightly soluble	1000 – 10000 parts
Practically insoluble	≥10000 parts

Table #1 United State Pharmacopeia solubility criteria

Ketoconazole is Practically insoluble in water; freely soluble in dichloromethane; soluble in methanol; sparingly soluble in ethanol (~750 g/l)(96 per cent) .

Solubility of ketoconazole in oleic acid and Tween80 (5.22,13, 88 respectively).

In this thesis; cocrystal formation of ketoconazole with acceptable pharmaceutical coformer, suberic acid and sebacic acid were evaluated.

1.7 Sebacic Acid and Suberic acid chemical structures

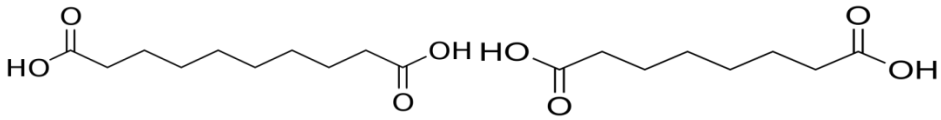


Fig. #3: a) Sebacic acid chemical structur b)Suberic acid chemical structure

Suberic andsebacic acids arecoformers , dicarboxylic acid. They have two hydroxyl groups that allow them to make hydrogen bonds with pharmaceutical drug, to producecocrystal, which improves the solubility of ketoconazole.

1.8 Microemulsion

Microemulsions are homogeneous, optically isotropic, transparent, thermodynamically stable dispersions of water and oil, stabilized by a surfactant, usually in combination with a cosurfactant (typically a short-chain alcohol) obtained by titration [47].[48].[49].

The term ―microemulsion means thermodynamically stable isotropically clear dispersion of two immiscible liquids, like oil and water, they are stabilized by an interfacial thin film of surfactant molecules[50] .[51],The dispersed phase usually included small particles, with a size range of 5 nm-200 nm, and has very low interfacial tension of (O/W). Microemulsions are transparent, with the reason of that the droplet size is less than 25% of the wavelength of visible light. Microemulsion can be produced readily and spontaneously, sometimes without need high-energy input.

Three types ofmicroemulsions could be formed:

1. Water in oil microemulsions or reverse microemulsion, the water particles are dispersed in the continuous oil phase, (W/O).
2. Oil in water microemulsions or direct microemulsion , the oil particles are dispersed in the continuous aqueous phase, (O/W).
3. Bi-continuous microemulsions, the micro ranges of oil and water are inter dispersed inside the system.

In all types of microemulsions, the interface is stabilized by using surfactants and/or co-surfactants as shown in Figure. #4

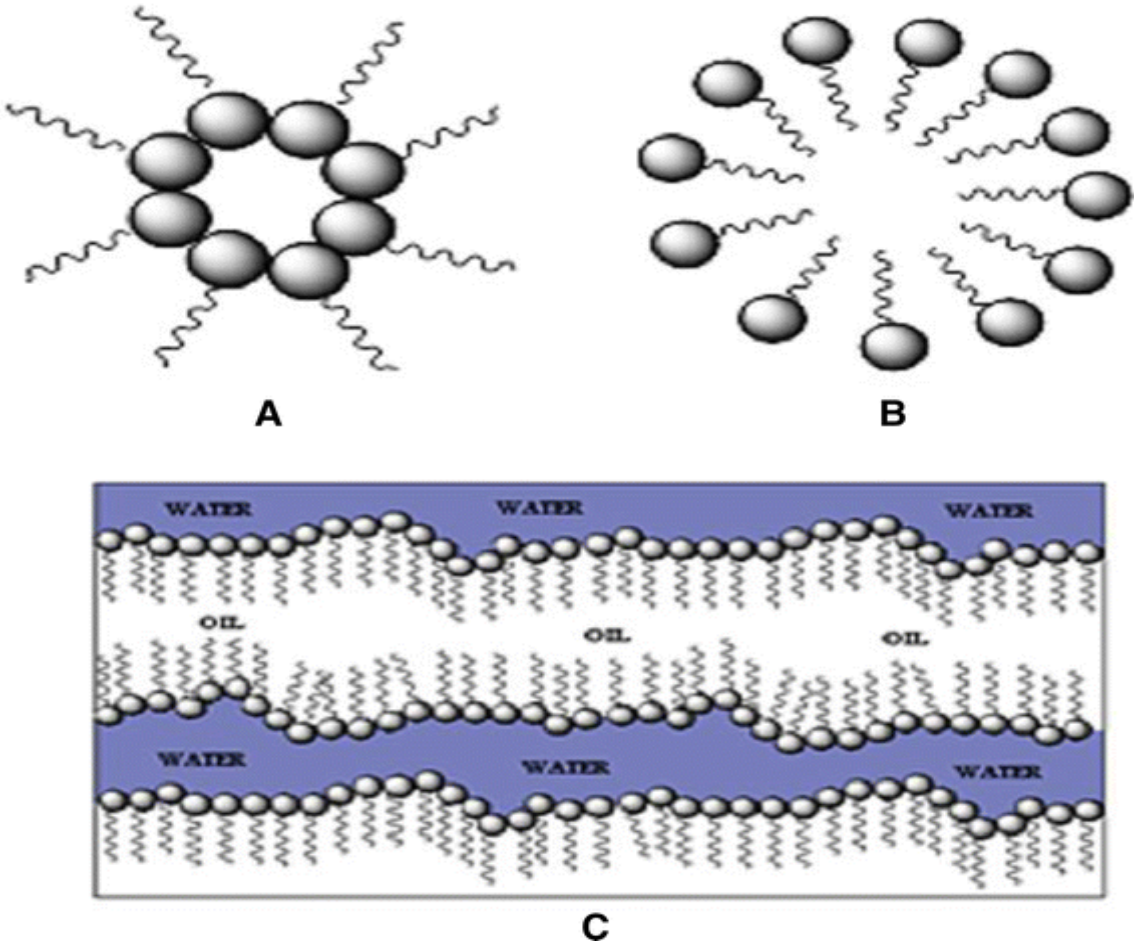


Figure.#4.Microemulsion structures: (a) reverse microemulsion ,(b) direct microemulsion and (c)bicontinuousmicroemulsion

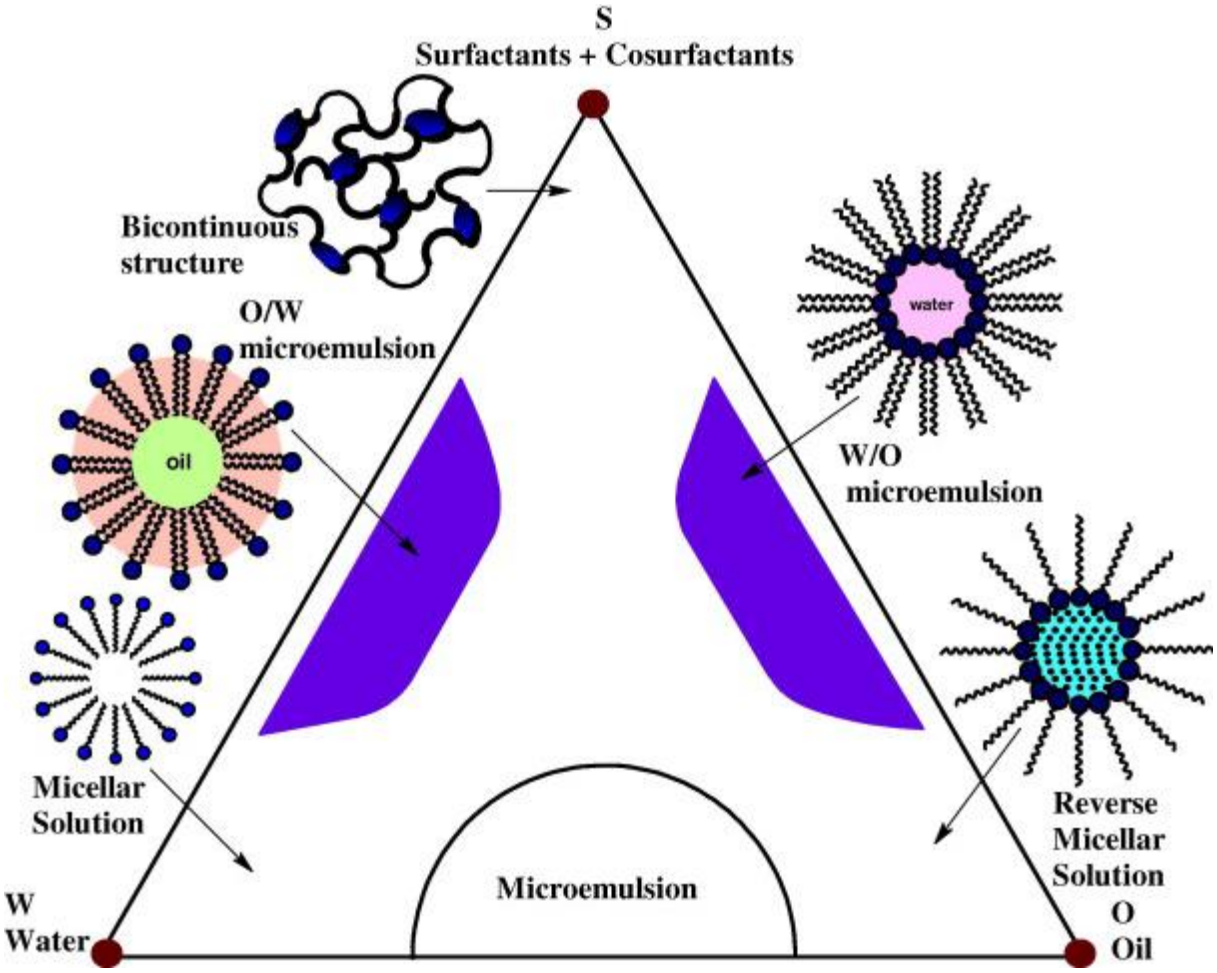


Fig.#5: Hypothetical phase regions of microemulsion systems [52]

1.8.1 Advantages of microemulsion for the transdermal delivery of drugs

1- Due to its high solubilizing ability, a significant amount of drug can be introduced into the formulation, with increased thermodynamic activity towards the skin.

2- the permeation rate of a drug from microemulsion may be increased, meanwhile the attraction of the API to the internal phase in microemulsion can be easily changed, to favor splitting into the stratum corneum, using different internal phases and changing the composition of the microemulsion.

3- the surfactant and cosurfactant used in the microemulsion may reduce the diffusional barrier of the stratum corneum by acting as penetration enhancers.[53]

1.8.2 More benefits of Microemulsion compared with other dosage types

1- Increasing the rate amount of absorption.

2- Decreasing variability in absorption.

3- Powerful in solubilizing lipophilic drug.

4- Adding an aqueous dosage form for water insoluble drugs.

5- Increasing bioavailability.

6- High effectivity in penetration of the drug moiety.

7- helpful in taste masking.

8- Protect oil-phase drugs from hydrolysis and oxidation in the O / W microemulsion.

9- Reducing the required amount of energy formulation

10- The drug can be administered through various routes, such as tropical, oral and intravenous[54].

1.8.3 Microemulsion preparation techniques

- Phase titration method:

Microemulsions are prepared by the process of spontaneous emulsification (phase titration process) and can be represented using phase diagrams. The Phase diagram design is a useful approach to studying the dynamic set of interactions that can occur when different components are mixed, Microemulsions are formed together with general structures of association (such as emulsions, micelles, lamellar, hexagonal, cubic, and various gels and oily dispersion) which depends on the chemical composition of each component and its concentration. At construction of phase diagram, each corner of the diagram represents 100% of the individual component. The region can be divided into w/o or o/w Microemulsion simply by taking into consideration the composition which is whether it is rich in oil or water.[55]

- Phase inversion method:

  Phase inversion of microemulsions occurs upon addition of excess of the dispersed phase or in response to temperature. During phase inversion dramatic physical changes occur including changes in particle size that can influence on drug release both in vivo and in vitro.

These methods make use of changing the spontaneous curvature of the surfactant. For non-ionic surfactants, this can be achieved by changing the temperature of thesystem, forcing a transition from an o/w microemulsion at low temperatures to a w/o microemulsion at higher temperatures (transitional phase inversion).[56]

1.8.4 Components of microemulsion formulations

A lot of oils and surfactants are obtainable which can be used as components of microemulsion system but their toxicity, irritation potential and unclear mechanism of action limit their use, to select components of the formulation that must be are biocompatible, non-toxic, and clinically acceptable.

1.8.4.1 Oil phase

This is an essential carrier after water because its properties to solubilize lipophilic drug molecules and enhance the absorption through lipid layer present in body.Also has sole property of penetrating cell wall and therefore very useful for lipophilic active drug delivery. Swelling of tail group region of the surfactant is effect by oil phase. Penetration has greater extent in short chain alkanes as compared to long chain alkanes. Oil phase can consist ofsaturated fatty acids: lauric, myristic and capric acid, unsaturated fatty acids: oleic acid, linoleic acid and linolenic acid or fatty acid esters: ethyl or methyl esters of lauric, myristic and oleic acid.[57,58]

1.8.4.2 Surfactants

Surfactants are abbreviation of surface active agents which are organic compounds with at least one lyophilic ('solvent- loving') group and one lyophobic (‘solvent-fearing’) group in the molecule If water or an aqueous solution is the solvent in which the surfactant is to be used, then The respective terms ' hydrophilic ' and ' hydrophobic ' are used. The surfactant containsat least one non-polar group and one polar (or ionic) group which is represented in a somewhat stylized form shown in Figure #6

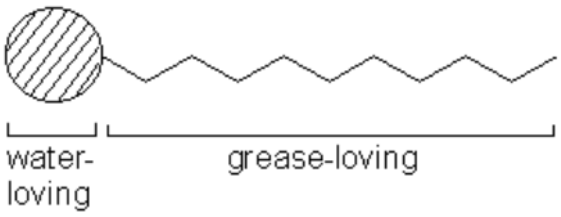


Figure #6: Schematic illustration of surfactant molecule .[59]

The amount of surfactant in emulsions is very low, 0.1% to 1.0% of the overall emulsion weight,

Selection of a suitable surfactant is crucial to the formation of any microemulsion. When we prepare w/o microemulsion will select suitable hydrophobic surfactants and choose good hydrophilic surfactants to form o/w microemulsion.In industrial applications, cheap ionic surfactants are widely used but ionic surfactant toxicity restricts their usage in milk, medicinal, and cosmetic applications. Nonionic surfactants are very often used in pharmaceutical microemulsion formation. Tweens (ethoxylatedsorbitan esters) are well known and widely used. They are water-soluble and have high HLB values and, therefore, are used mainly for making o/w microemulsions.[60]

1.8.4.3 Classification of surfactant

The most useful chemical classification of surfactant is based on the nature of the hydrophilic part (head), with subgroups based on the nature of the hydrophobic part (tail). The four basic classes of surfactants are[61] :Anionic in which the hydrophilic part is a negatively charged

group, cationic in which the hydrophilic part is a positive charge, nonionic the hydrophilic has no charge, but derives its water solubility from highly polar groups such as sugars and amphoteric groups (zwitterionic). It is not surprising that the selection of a suitable surfactant for a given application can become a significant problem. Thus an empirical numbering system has been developed to enable the correct type of surfactant to be chosen. The system is called the 'hydrophile–lipophile balance' (HLB). Therefore polysorbate 80 (Tween 80) is selected in this project preparation to achieve the drug product micro emulsion formation since it had an HLB=15.0

1.8.4.4Cosurfactant-

Co-surfactants play a very important role in microemulsions, they reduce the interfacial tension to very small values to obtain thermodynamic stability.Co-surfactants are used as breakers of liquid crystal structure. They lower efficiencycompared to surfactants with unsaturated (double) bonds in their tails. But they are especially essential when the surfactant has a saturated tail. In general, the co-surfactants are short-chain as ethanol, and medium-chain as propanol, both decrease the interfacial tension and increase the fluidity of the interface.[62]

1.8.4.5Aqueousphase

This represents the aqueous phase of the microemulsion. Usually used agents are water, alcohol etc. Used for drugs that cannot be formulated as an aqueous solution, emulsions and microemulsions have typically been cost effective and provided for ease of administration[63] . Co-solvents which are organic solvents like ethanol, propylene glycol (PG), and polyethylene glycol (PEG) can also be added to dissolve relatively high concentrations of surfactants including lipid soluble drugs.[64,65]

1.8.5 Factorsaffecting the microemulsion

1.8.5 a Packing ratio

The HLB of surfactant determines the type of microemulsion through its effect on molecular packing and film curvature,withthe analysis of film curvature for surfactant association's leadings to the formation of microemulsion.[66]

1.8.5 b Property of surfactant, oil phase and temperature

The kind of microemulsion depends on the type of surfactant, and that Surfactant consist of two parts, hydrophilic head group and lipophilic tail group, which are a degree of the differential affinity of water to increase of head part volume and oil to enlarge the tail area, which playsan important role to calculate approximately the surfactant HLB in a specific system. When weuse a high concentration of the surfactants, degree of dissociation of polar groups becomes lower and the type of microemulsion producedis w/o type. Diluting with water may increase dissociation and leads to an o/w system. Ionic surfactants are strongly influenced by temperature. It mainly causes increased surfactant counter-ion dissociation.

The oil component also influences curvature by its ability to penetrate and therefore enlarge the tail group region of the surfactant monolayer. Short chains oils penetrate the lipophilic group region to a great extent and results in increased negative curvature.

Temperature is extremely important in determining the effective head group size of nonionic surfactants. At low temperature, they are hydrophilic and form normal o/w system. At higher temperature, they are lipophilic and form w/o systems. At an intermediate temperature, microemulsion coexists with excess water and oil phases and forms bicontinuous structure.

1.8.5 c The chain length, type and nature of cosurfactant

In microemulsions alcohol is commonly used as a cosurfactant. Adding shorter cosurfactant chains provides a favorable curvature effect when alcohol swells in the head region more than tail region so, it becomes more hydrophilic and o/w type is favored, whereas longer chain cosurfactant favors w/o type,by alcohol swelling more in chain region than head region.[67]

Chapter Two

- **Literature review**
- **Problem**
- **Research objectives**

2.1 Literature review

2.1.1 Introduction

On reviewing the available literature, it is evident that extensive research has been carried out to address various aspects ofEnhancement of solubility of antifungal Ketoconazole By Co-crystallization and Microemulsion ,With this in mindin an attempt to simplify and delimit the subject to some extent, this chapter will focus primarily on the work that has been carried out on the synthesis and characterizations the cocrystals of ketoconazole and coformer also on microemulsion and phase diagram .

The primary objective of this chapter is to provide an insight into synthesis the cocrystals then characterized them. Characterization of melting point, FTIR , DSC and solubility test . These cocrystals are very important because they enter in pharmaceutical applications such as antifungal drugs.

2.1.2 Synthesis and characterization of the cocrystal of ketoconazole

IndraIndraa, et al.(2019)[38], cocrystal of ketoconazole and ascorbic acid was synthesized by the slurry method to improve solubility properties of ketoconazole. and characterized by using Thermogram (Differential Scanning Calorimetry), FTIR spectroscopy and solubility analysis. Ketoconazole cocrystal of the slurry method showed increased solubility by 50 times compared to ketoconazole alone.

R. N. Kamble , et al.(2017)[40],ketoconazole (KTZ) cocrystals, including oxalic acid, furamic acid and nicotinamide, were prepared using a solvent crystallization method . Prepared co-crystals have been distinguished by their thermal properties, prepared co-crystals have demonstrated improved solubility.

Stevanus Hiendrawan, et al.(2015)[68], ketoconazole cocrystals including oxalic acid (OXA) and fumaric acid (FUMA) were prepared by slurry conversion method. Differential Scanning Calorimetry (DSC) techniquewas employed to investigate the crystallinity,

melting point cocrystal. KTZ co-crystal was evaluated further for their solubility. The result from DSC analysis confirms the formation of cocrystal of KTZ with OXA and FUMA.

Flavia A. Martin, et al.(2013)[46]ketoconazole and three co-crystals with fumaric, succinic and adipic acids were synthesized by dissolving the ketoconazole with coformer in suitable solvent and were determined by single-crystal X-ray diffraction .All solid forms were stable and showedsignificant solubility improvement compared to ketoconazole alone ,with a 100-fold enhancement in the case of the co-crystals withfumaric and adipic acids .

Amit Kumar Aggarwal, et al.(2011)[45]. Solid dispersions of ketoconazole were prepared by the melting method using nicotinamide as carrier.These solid dispersions were characterized by differential scanning calorimetry (DSC), Fourier transform infrared (FT-IR) spectroscopy, Solubility studies indicated that nicotinamide increased significantly the solubility of ketoconazole in water.

Ali Shayanfar ,etal.(2014)[69]. Ketoconazole (KTZ)cocrystal with4-amino benzoic acid (PABA) and nicotinamide were prepared through solvent evaporation method,and characterized by differential scanning calorimetry, powder X-ray diffraction and Fourier transform infrared spectroscopy. Dissolution profile and apparent solubility of the cocrystal have been increased in a non-acidic condition because of higher solubility of the coformer.In this research, the higher coformer solubility than the drug has shown that the cocrystal is less stable but more soluble.

FatemehKeramatnia, etal.(2016)[70]. Ketoconazole (KTZ) (synthetic imidazole antifungal drug), which is a practically insoluble drug, citric acid (CA), and tartaric acid (TA) were chosen as counter-ions for preparing the ionic liquid were prepared applying solvent evaporation method with methanol as the solvent. Characterization by different instrumental analysis methods such as DSC, These results could be utilized in overcoming the disadvantages of KTZ solid form and increasing its solubility.

2.1.3 Constructionof phase diagram

Mrunali R. Patel , etal .(2010)[71].The solubility of KTZ was determined in various oils and surfactants, the oils employed were isopropyl myristate (IPM), ethyl laurate (EL), wheat germ oil(WO),ethyl oleat(EO), arachis oil (AO)and oleic acid (OA). The surfactants used werepolysorbate80(T-80), polysorbat40 (T-40), polysorbate20 (T- 20), span80(S-80) . Drug powder of ketoconazole (KTZ) was added in excess to each of the oils and the solubility of KTZ was determined by analyzing the filtrate spectrophotometrically using double beam, Among the nonionic surfactants studied tween 80 (T-80)led to the highest solubility of KTZ, Also T-80 is known to be less affected by pH and ionic strength changes and acts as a solublizing agent. On the other hand, there was no significant difference in the solubility of KTZ among the various oils tested except arachis oil (AO) and oleic acid (OA) which exhibited low solubility as compared to other oils.

Nisha Tiwari ,et al .(2018)[72],Microemulsion was formulated using rose oil as oil phase, Tween 20 as surfactant and water as continuous phase.Themicroemulsions for drug loaded and unloaded were prepared by titrating oil- surfactant mixture with water, until clear oil in water microemulsion was formed .The worked out optimized formulation was achieved through oil: surfactant: water (5:20:70) which was subjected to physiochemical analysis. The rate of drug release was checked for drug loaded microemulsion compared to conventional drug (Ketoconazole).

2.2 Problem

Pharmaceutical cocrystal described as solid, a crystalline material consisting of two or more molecules in the same lattice of crystal , a Many cocrystals were intended to improve the physico-chemical properties of APIs such as solubility / dissolution.

Ketoconazole is a broad-spectrum imidazole antifungal agent and administered by topical and oral.Being a BCS class II drug, the chemical structure of ketoconazole has a visible

hydrophobic site and weak basicity that promotes to its poor aqueous solubility,so the co-crystallization method that used to increase solubility of ketoconazole .

On other hand, as possible drug delivery systems microemulsion has attracted increasing attention,Either as vehicles for topical use or as bioavailability enhancers for poorly water-soluble pharmaceutical active ingredients (API),The existence of microemulsion of different polarity within the same single-phase solution enables both hydrophilic and lipophilic materials to be solubilized.

The main benefits of the topical delivery system is its ability to distribute medications to a particular location more selectively,thermodynamic stability, optical clarity, ease of preparation.We must use these benefits for microemulsion and work to develop productivity in order to improve the "Palestinian pharmaceutical industry‖ that increases the patient satisfaction and compliance. We need to find such industries support from the Palestinian ministries to adopt such these projects.

2.3 Researchobjectives

The main purpose of my research is to prepare ketoconazole co-crystal by using different diacids in various solvents by using solvent crystallization method ,and characterize the new cocrystal by using different method .the second aim of this research is study the stability and phase diagram of ketoconazole in a therapeutic product, to produce stable, biocompatible microemulsion with ketoconazole and a sugar/nonionic surfactant like Tween 80, as well as a co-solvent such as ethanol,propylene glycol and water. The topical microemulsion therapeutic products are important in drug delivery systems and therapy, which becomes more widespread.

To obtain this plan there are many objectives needed to be achieved in this present work:

- To prepare cocrystal and microemulsion to improve the drug solubility of KTZ
- To estimate phase diagram of the best components concentration for microemulsion preparation
- To use the crossed polarizers instruments to identify, approve and justify the output results
- To use the following instruments in identifying the co-crystal output: FTIR,DSC and melting point range .

Chapter Three

Instrumentation and Methodology

3. Instrumentation & Methodology

3.1 InstrumentationEquipments &

Analytical balance, Vortex, culture tubes sealed with Viton lined screw caps,sonicator, crossed polarizers, FTIR,DSC(PerkinElmer, jade DSC) , Melting point apparatus(Designed in UK,assembled in PRC,SMP10.), Hot plate, glass ware .

3.2 Materials

Material which is needed for preparation cocrystal of ketoconazole with diacid (suberic acid &sebacic acid) :

Ketoconazole (KTZ) (al-Quds Pharmaceutical Company, Piramal Enterprises Limited) , methanol 99%(sigma Aldrich),acetone 99% (chemistry lab at Al-Quds university), suberic acid 98% (Mw:174.19, mp:139-146 °C),sebacic acid 99%(Mw: 202.25 , mp:133-137 °C (lit.) (sigma Aldrich).

Materials which is needed for preparation microemulsion :

polysorbate 80 (Tween 80), double distilled water (Chemical Analysis Laboratoryatal-Quds university), oleic acid, KTZ ,ethanol, and propylene glycol (PG).All components used as supplied and tested in al-Quds university .

3.3 Co-crystal Methodology

Ketoconazole is considered as active pharmaceutical ingredients (API) in my work. The used solvent ismethanol(99%), and co-solvent is Acetone, they used in the co-crystal technique (solvent crystallization).

Ketoconazole co-crystals were prepared by using solvent crystallization method:

Weigh each diacid with the API according to **Table #2**which shows the molar ratio between the API and each diacid in mg which is (1:1), mixing and grinding them by mortar and pestle,then, they were dissolved in minimum amount of solvent (methanol:aceton) (1:1) ,

which the (1:1) cocrystals of KTZ and the dicarboxylic acid coformers were prepared by solvent crystallization method at room temperature.The crystals wereseparated by filtration through a membrane (0.45 μm) and dried at room temperature for 24 h, The dried crystals were placed in airtight container and stored in a desiccated environment and the crystals to be testedfor FTIR spectroscopy were collected individually and compared with the pure diacids and the API spectrum that were tested before the beginning, also, testing the melting point for the produced crystals, and testing the solubility in water .and testing the DSC .

Table #2 The molar ratio between KTZ(531.43 mg, 1 mmol) and diacids weight (1:1 mmol)

#	Diacid	Formula	Molar mass	1:1 mmol (molar ratio)
1	Suberic acid	$C_8H_{14}O_4$	174.2	17.4 mg
2	Sebacic acid	$C_{10}H_{18}O_4$	202.2475	20.2mg

3.3.1 Solubility of Co-crystals

Weighing 10 mg of each new co-crystal (Suberic acid-KTZ ,Sebacic acid- KTZ), then put in clean tubes, then, added dropwise of water , with stirring, until dissolve the cocrystal.

3.3.2 Differential scanning calorimeter test

Thermal properties of drug alone and co-crystals were

investigated using differential scanning calorimetry (DSC). Approximately 5–7 mg of sample was hermeticallysealed in an aluminum pan with a hole and heated at a constant rate of 10 °C/min over a temperature range of RT –200 °C.

3.3.3 Melting point byMel-Temp Apparatus

The result of melting point achieved by using Mel-Temp Apparatus.

3.4 Phase diagram Methodology

3.4.1 Constructing of phase diagram

The phase behavior of the systems consisting of water (with or without Co-solvent ,propylene glycol),oleic acid with ketoconazole , surfactant (Tween 80), may be described

on a phase tetrahedron whose apexes respectively represent the pure components, 1g mixture consisting of oleic acid with ketoconazole and surfactant (Tween 80) at different weight ratios were prepared in culture tubes sealed with Viton lined screw caps and stirred by vortex until clear solution was obtained. These samples were titrated with water (with or without Co-solvent (propylene glycol)) which was added drop wise until its solubilization limit was reached. Vigorous stirring followed after additions on a vortex mixer. The time for equilibration between each addition was

Typicallyfrom a few minutes up to 24 hours (therefore the reading will be taken after 24 hours). Each phase diagram was investigated at ambient temperature, and detecting the number of phase by bare eye, the anisotropy by cross polarizer. The anisotropic lamellar liquid crystal and hexagonal liquid crystal are determined by the cross polarizer and polarizing microscope. Detect the boundary of single phase. Finally draw the phase diagram using specified computer software.

Table 3: Phase diagram1 components **Table 4** Phase diagram 2 components

Tube #	**Oleic acid (g) + KTZ**	**Tween80 (g)**
19	0.9006	0.1006
28	0.8030	0.2002
37	0.7040	0.3036
46	0.6023	0.4012
55	0.5024	0.5106
64	0.4021	0.6018
73	0.3013	0.7036
82	0.2025	0.8034
91	0.1014	0.9033

Tube #	**Oleic acid (g) +KTZ**	**Ethanol/Tween80(g)(1:1)**
19	0.9004	0.1011
28	0.8001	0.2005
37	0.7011	0.3002
46	0.6014	0.4039
55	0.5001	0.5010
64	0.4002	0.6003
73	0.3004	0.7002
82	0.2006	0.8028
91	0.1008	0.9010

Table 5 Phase diagram3 components Table 6 Phase diagram 4 components

Tube #	Oleic acid (g) + KTZ	Eth /Tween80 (1:2)(g)
19	0.9004	0.1005
28	0.8002	0.2006
37	0.7012	0.3018
46	0.6002	0.4013
55	0.5047	0.5039
64	0.4003	0.6006
73	0.3002	0.7011
82	0.2006	0.8000
91	0.1004	0.9000

Tube #	Oleic acid (g) +KTZ	Eth/Tween80 (1:3)(g)
19	0.9008	0.1004
28	0.8013	0.2001
37	0.7001	0.3000
46	0.6009	0.4000
55	0.5007	0.5000
64	0.4001	0.6000
73	0.3002	0.7019
82	0.2004	0.8000
91	0.1020	0.9001

Table #7: Phase diagram5 components (double distilled water+PG)

Tube #	Oleic acid (g)	Tween80 (g)
19	0.9007	0.1017
28	0.8000	0.2033
37	0.7000	0.3009
46	0.6019	0.4040
55	0.5000	0.5031
64	0.4001	0.6000
73	0.3010	0.7026
82	0.2000	0.8021
91	0.1000	0.9004

Note: The tube # shows the amount of surfactant and oil components in each tube. For example, tube # 91 contains 0.9g of oil and 0.1g of surfactant and co-surfactant

3.4.2 Titration amount required in grams for each percentage

Table #8 Titration percentage versus weight.

%	Total added weight (g)	Net weight Each interval (g)
4	0.0417	0.0417
8	0.0870	0.0453
12	0.1364	0.0494
16	0.1905	0.0541
20	0.2500	0.0595
24	0.3158	0.0658
28	0.3890	0.0732
32	0.4707	0.0817
36	0.5626	0.0919
40	0.6668	0.1042
44	0.7859	0.1191
48	0.9233	0.1374
52	1.0836	0.1603
56	1.2730	0.1894
60	1.5003	0.2273
64	1.7776	0.2773
68	2.1249	0.3473
72	2.5715	0.4466
76	3.1670	0.5955
80	4.0007	0.8337
84	5.2077	1.207
88	7.2910	2.0833
92	11.4577	4.1667
96	23.9577	12.500

3.4.2.1 Preparation of contents for pseudo phase diagram

- Preparation of Tween 80:ethanol (95%) (1:1).
- Preparation of Tween 80:Ethanol (95%) (2:1).
- Preparation of Tween 80:Ethanol (95%) (3:1).
- Preparation of double distilled water: propylene glycol (1:1).
- Dissolving the KTZ in oil phase (oleic acid).

3.4.2.2 Choosing a suitable surfactant and oil phase

To develop microemulsion (ME) system for topical delivery of KTZ, the suitable oil and surfactant has to be chosen, Among the nonionic surfactants studied T-80 led to the highest solubility of KTZ, Also T-80 is known to be less affected by pH and ionic strength changes and acts as a solublizing agent, which oleic acid (OA) exhibited low solubility as compared to other oils.

Chapter four

Results and discussion

4. Results and discussion

4.1 Cocrystal results-

4.1.1 Solvent crystallization method

The diacids (sebacic acid, suberic acid) with KTZ and the used solvents (methanol and acetone) produced white cocrystals successfully, the hydroxyl group in these diacids will be differentiated and fluctuation of hydrogen bonding will invite, and leading to bonding with hydrogen bonding in the API and forming stable co-crystal. After that, we tested them individually by FTIR spectroscopy, and compared the result with the pure samples spectrum for each diacid and KTZ that was tested before the beginning of crystallization**.**

4.1.2 Co-crystal solubility in water

The most important goal in co-crystallization method is to improve the solubility of the ketoconazole in the pharmaceutical drugs. We tested the solubility in water for each co-crystal

We weighed about 10 mg from each co-crystal into large test tube then added dropwise of purified water with stirring then increased the amount of water gradually to dissolve all the solid of co-crystal after dissolve them recorded the volume of water which needed to dissolve the solid .

The solubiltyofsuberic acid-KTZcocrystal is 10mg\22 mL,sebacicacicd-KTZcocrystal is 10mg\24 mL

product	Solubility mg/100ml
suberic acid-KTZ	46
sebacicacicd-KTZ	42

Table #9 solublity of cocrystal-KTZ

Accordingly, the efforts to co-crystalize the ketoconazole with diacids resulted in enhancement of the solubility of the API.

The dosage given to adults is 200 mg daily, and for infants is between 3.3-6.6 mg. accordingly this quantity is coherent with results that were obtained in this thesis. Next step is to do use drug preparations that are given orally and complete the study.

4.1.3 The co-crystal melting point

The melting point is a physical property which can be determined by the temperature at which the solid phase at equilibrium with the liquid phase.

Ketoconazole is a pharmaceutical API, its melting point is 146-148˚C, the expected new co-crystal formed would have melting point to be different from the API and the diacid, which forms the co-crystals, the following **Table #10**, shows the melting points for API, diacids and the produced co-crystals:

Material name	**Melting point (°C)**
ketoconazole (API)	146.0 -148.0
Suberic acid (diacid)	143.0 -146.0
Sebacic acid (diacid)	135.0 -138.0
Suberic acid- ketoconazole co-crystal	141.0 - 143.0
Sebacic acid- ketoconazole co-crystal	115.0-119.0

Table #10: Melting point results

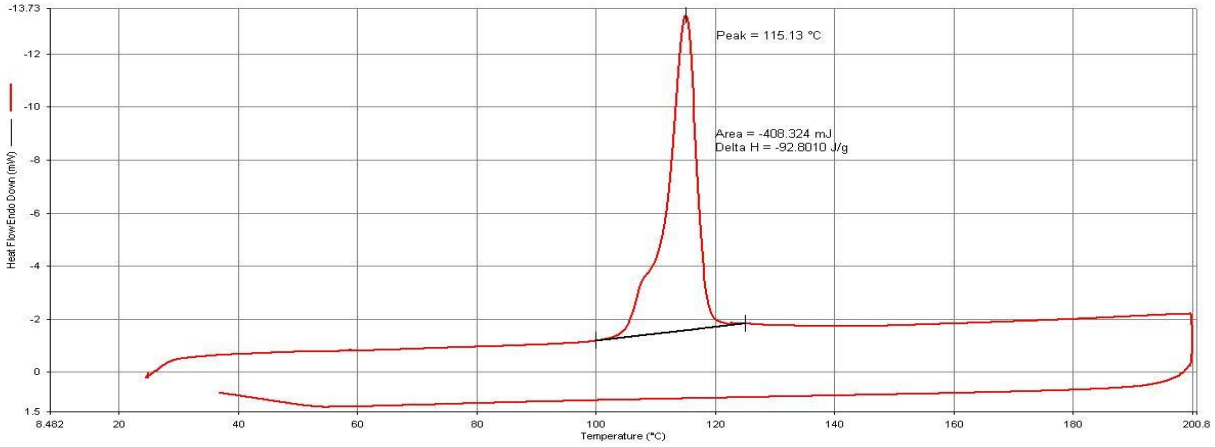
The melting points of cocrystal are different from those of ketoconazole and coformer , We noted that

An Indicative prove of the formation of the cocrystal is a change of the melting point, the results that were obtained is consistent with the success to obtain co-crystal.

4.1.4 DSC study of the pure compounds and the co-crystal

DSC study was done on the following compounds, pure sebacic acid, suberic acid, ketoconazole, and the co-crystal sebacicacid-ketoconazole, suberic acid-ketoconazole. A comprehensive study would be done in case it is done side by side with TGA study. In the case of the sebacic acid, ketoconazole, co-crystal sebacic acid-ketoconazole, we can see clearly a sharp exothermic peak attributed to melting point and the value ΔH_{fuss} can be easily calculated, the sharpness of the peak is indicative of the formation of co-crystal and the

melting occurred at that point (purity). If we look at the values where the melting occurred of the co-crystal we can see it occurred at a lower value than both pure compounds, with sharpness, this is coherent with formation of co-crystal. This evidence supports our previous results, FTIR, melting point and solubility. In the case of suberic acid, ketoconazole, co-crystal suberic acid-ketoconazole, it can be also seen that the DSC of the co-crystal gives a rise to sharp peak at 143` indicating that the melting is pure material, and it is clear that is no range in melting temperature is seen. Also comparing the melting point with that of pure material it can be seen that the melting point is lower and it is what is expected in cases of co-crystal.



Fig#7sebacic acid-KTZ co-crystal DSC

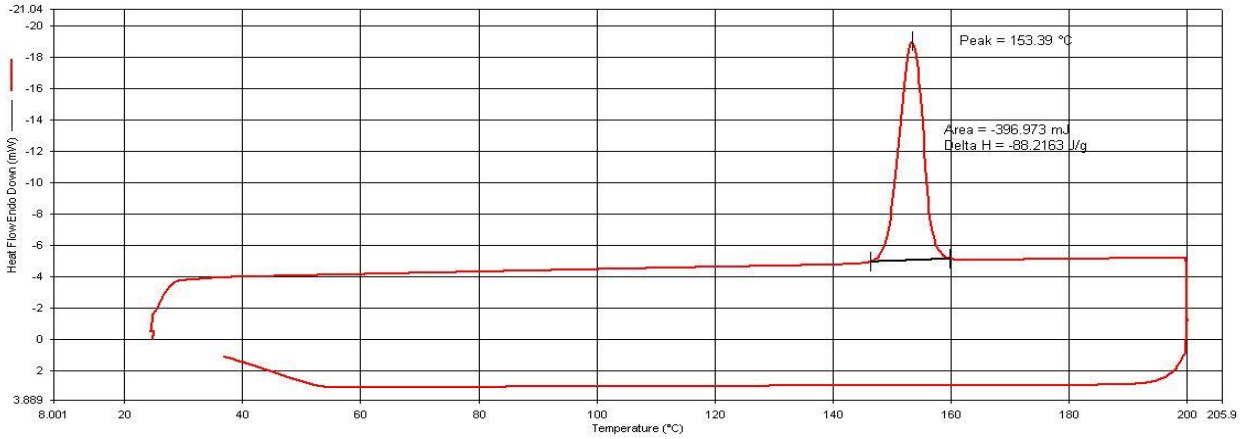


Fig.#8: DSC for KTZ

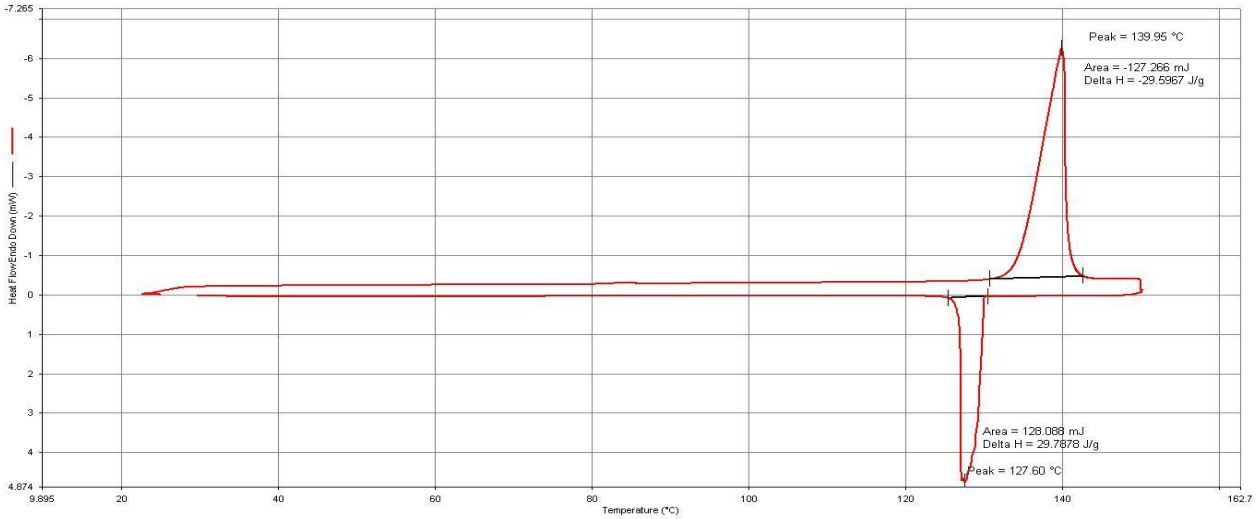
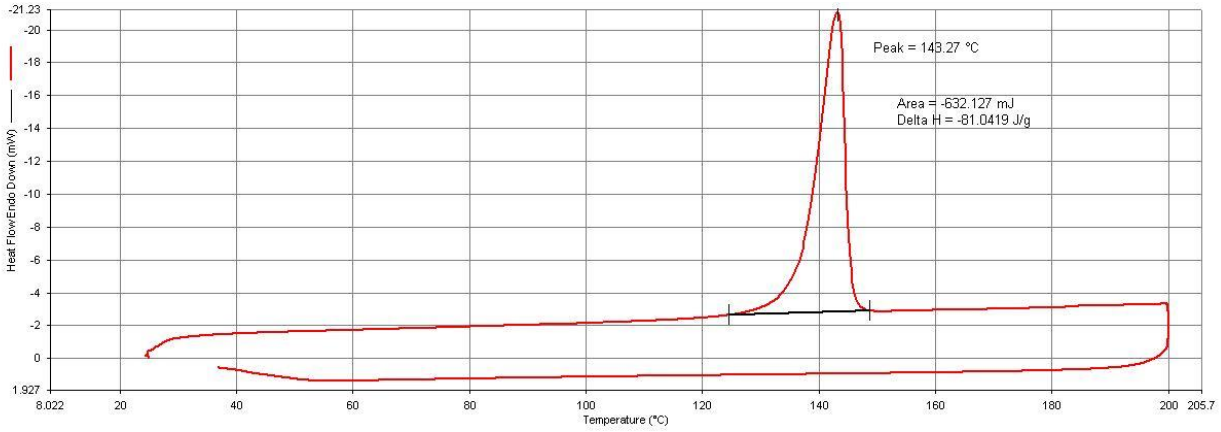


Fig.#9: sebacic acid coformer



Fig,#10: suberic acid-KTZ co-crystal

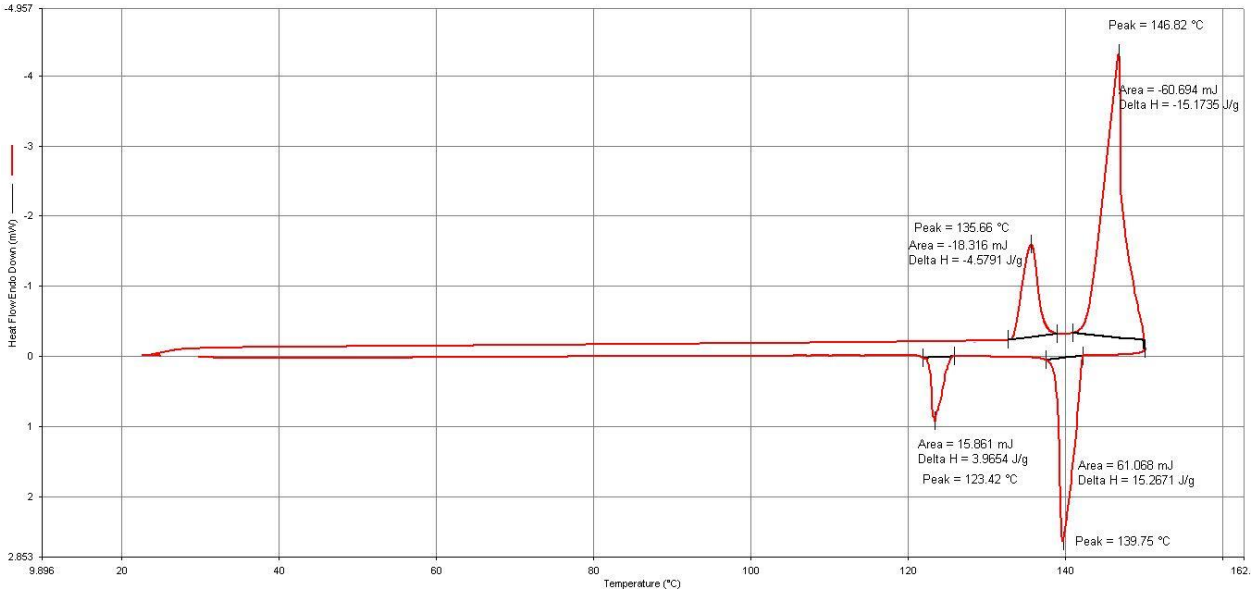


Fig.#11: suberic acid coformer

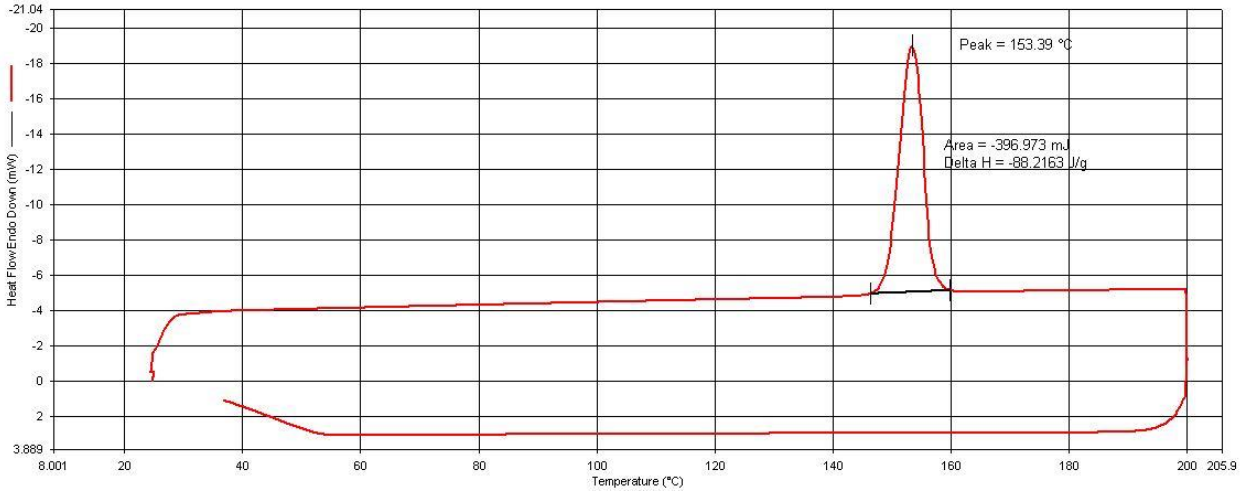


Fig.#12: DSC for KTZ

4.1.5 FTIR of The Co-crystal result

FTIR spectroscopy is used to identify the formation of the new crystalline phase. The FTIR of solid material is unique and can identify functional groups that exist in that particular product. The Ketoconazole has its own FTIR identity, and the diacids have their own identity. Upon the formation of a co-crystal the Characteristic bands of the FTIR will be affected from the new arrangement of molecules as result of the formation of the non-covalent interactions, in our case the hydrogen bonding. A Look at the FTIR spectrum of both products and comparing it with the FTIR spectrum of the co-crystal clearly indicate a shift in the position of the FTIR bands of the groups forming the hydrogen bonding. In addition, a change in the FTIR spectrum, in either the shape of the peaks or in the intensity of some of these peaks.

This change is indicative of the formation of Co-crystal, which means a new arrangement of the molecules of the ketoconazole. This change is attributed to the formation of co-crystal forming heteromeric arrangement of the array. This new arrangement can be identified and described and confirmed by X-Ray crystallography of single crystal. In our case we can support our results on the formation of this co-crystal, by FTIR spectrum, melting point, solubility, HNMR and 13CNMR.

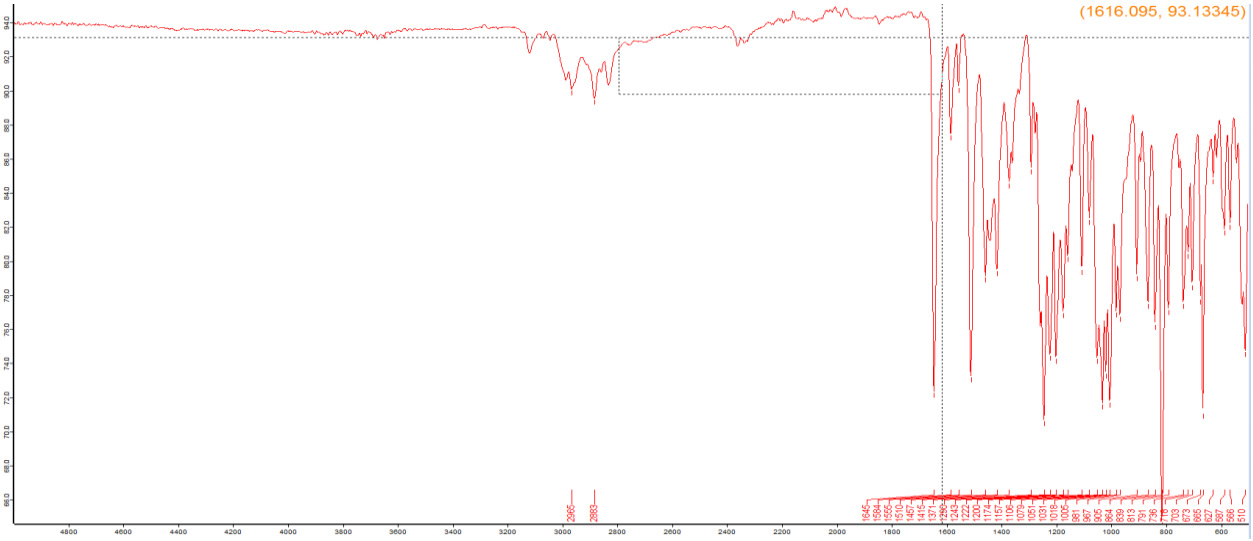


Fig.#13:FTIR of KTZ :

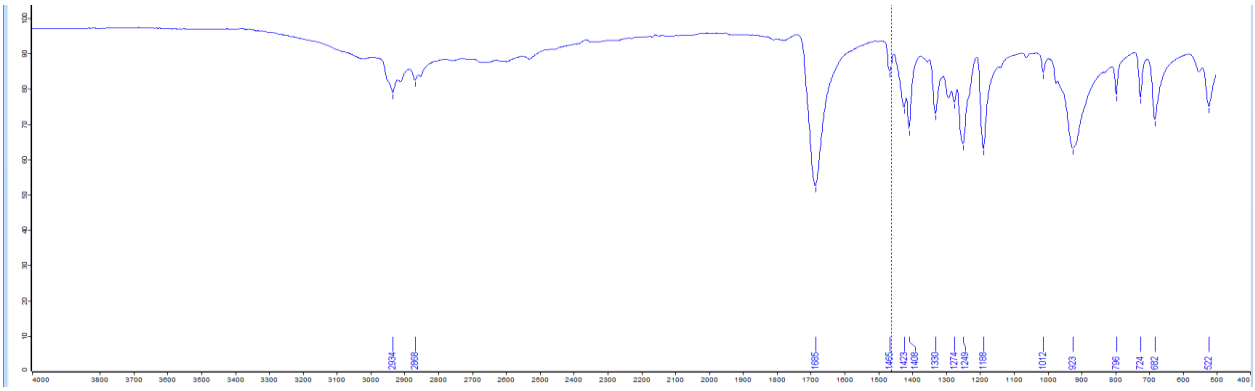


Fig.#14 FTIR of Suberic acid

Fig.#15FTIR of Suberic acid-KTZ cocrystal

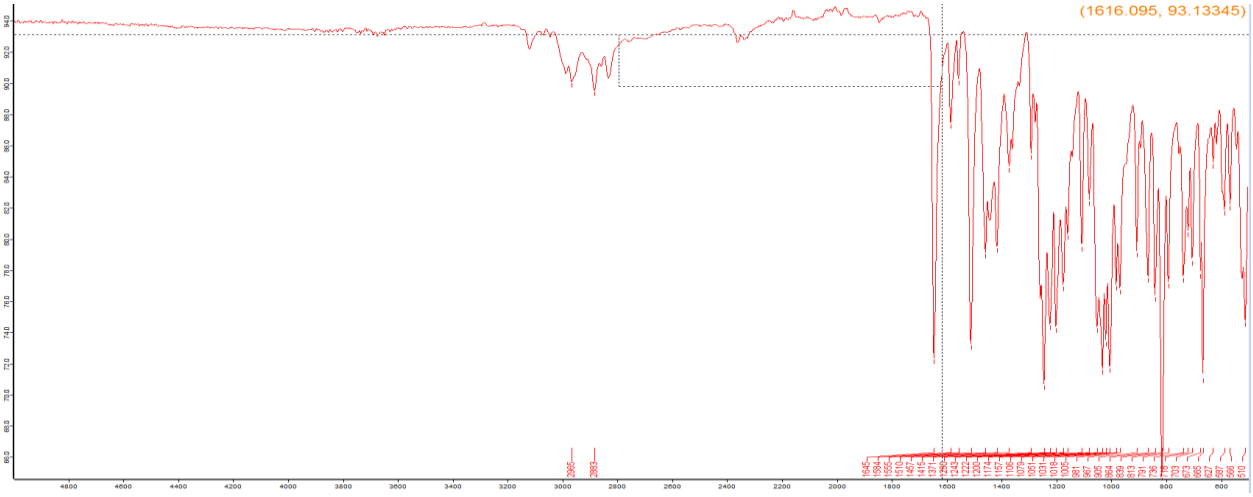


Fig.#16:FTIR of KTZ :

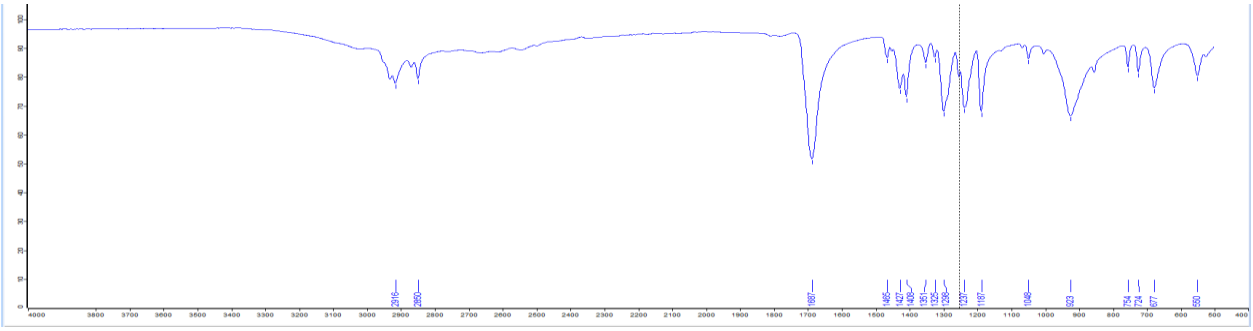


Fig.#17 FTIR Sebacic acid

Fig #18: FTIR of Sebacic acid-KTZcocrystal :

Table #11 Wave length shift in cm^{-1} for the major peaksof suberic acid-KTZ cocrystal :

KTZ	**suberic acid – KTZ cocrystal**	**suberic acid**
2965	3853	2934
2883	3735	2868
1645	3675	1685
1584	3124	1465
1555	2925	1423
1510	2360	1408
1457	1707	1330
1415	1606	1274
1371	1508	1249
1290	1466	1188
1243	1442	1012
1222	1375	923
1200	1326	796
1174	1282	724
1157	1270	682
1106	1243	522
1079	1183	
1051	1100	
1031	1082	
1005	1053	
981	1039	
967	1017	
905	987	
864	972	
839	930	
813	909	
791	870	
736	817	
718	788	
703	753	
673	728	
663	708	
665	675	
627	659	
587	595	
566	566	
510	546	

Table #12 Wave length shift in cm^{-1} for the major peaksof sebacic acid-KTZ cocrystal :

KTZ	**sebacic acid -KTZ cocrystal**	**sebacic acid**
2965	3675	2916
2883	2922	2850
1645	2360	1687
1584	1713	1465
1555	1646	1427
1510	1608	1408
1457	1509	1351
1415	1456	1325
1371	1435	1298
1290	1376	1237
1243	1342	1187
1222	1283	1048
1200	1239	923
1174	1223	754
1157	1176	724
1106	1103	677
1079	1080	550
1051	1052	
1031	1005	
1005	977	
981	942	
967	927	
905	894	
864	858	
839	836	
813	823	
791	787	
736	755	
718	726	
703	707	
673	660	
663	630	
665	595	
627	563	
587	542	

4.2 Microemulsion phase diagram results

4.2.1 Pseudo ternary phase diagram A

This phase diagram consists of Tween 80 and oleic acid oil with KTZ and double distilled water. Itdemonstrates thelocation of microemulsion at ambient temperature. This system produced a phase diagram with two regions.The first region obtained was a single clear, isotropic and not shiny solution ,while the second region was liquid crystalline structure , observed using polarizer light microscopy which was started as a single , clear ,viscous and shiny .

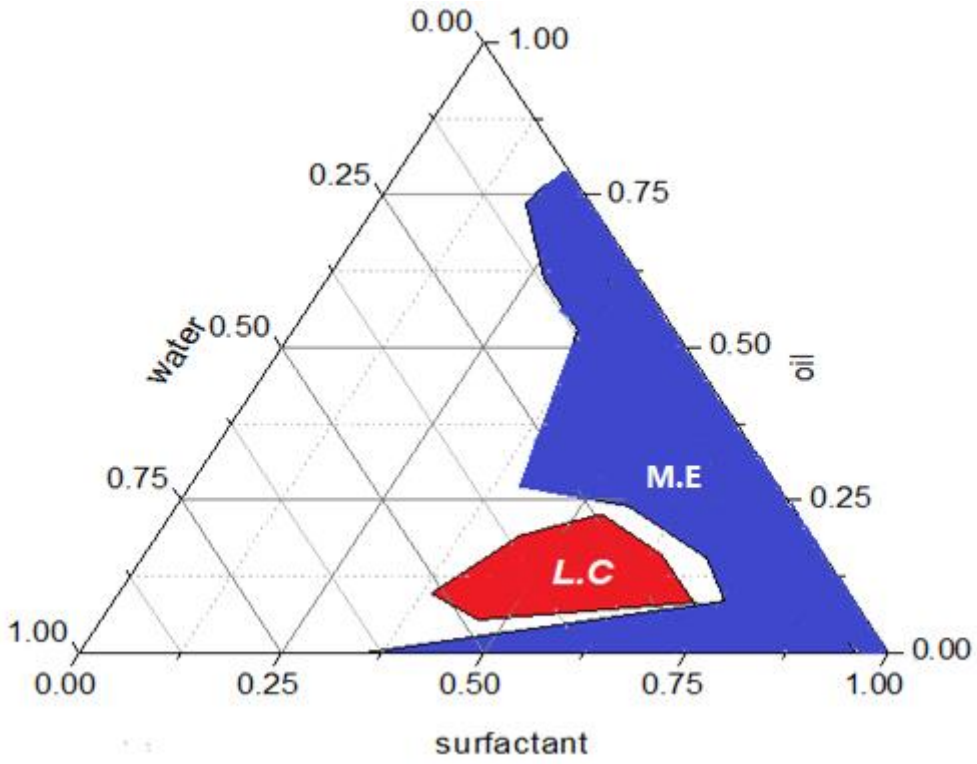


Figure #19: Oliec acid with KTZ , Tween 80 , doubled distilled Water

4.2.2 Pseudo ternary phase diagram A1

Consist of Tween 80:ethanol (1:1) in which is the ethanol was used as cosurfactant and oleic acid oil with KTZ and double distilled water.This system produced a phase diagram with single region,which demonstrates that the microemulsion at ambient temperature was obtained and started as a single clear, isotropic and not shiny solution .

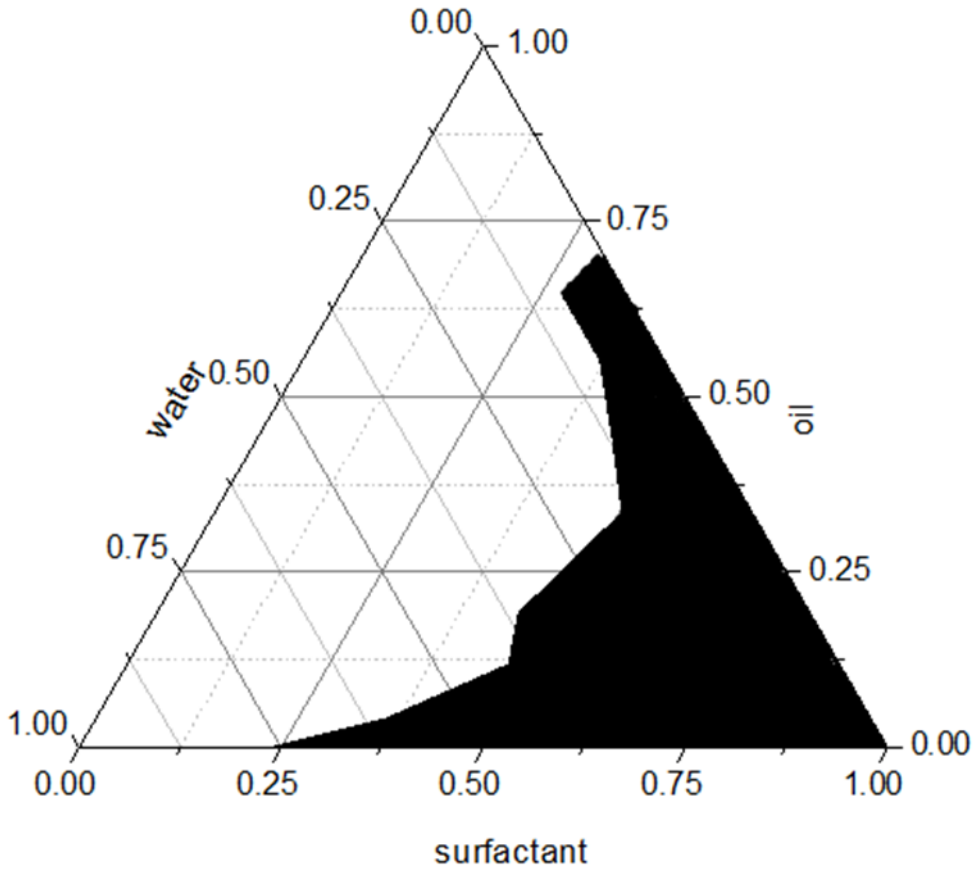


Figure #20 : oleic acid with KTZ , (tween 80 , ethanol (1:1)),double distilled water

4.2.3 Pseudo ternary phase diagram A2

Consist of Tween 80:ethanol (2:1) in which the ethanol was used as co-surfactant and oleic acid oil with KTZ and double distilled water demonstrates that the microemulsion at ambient temperature was obtained and started as a single clear, isotropic and not shiny solution .

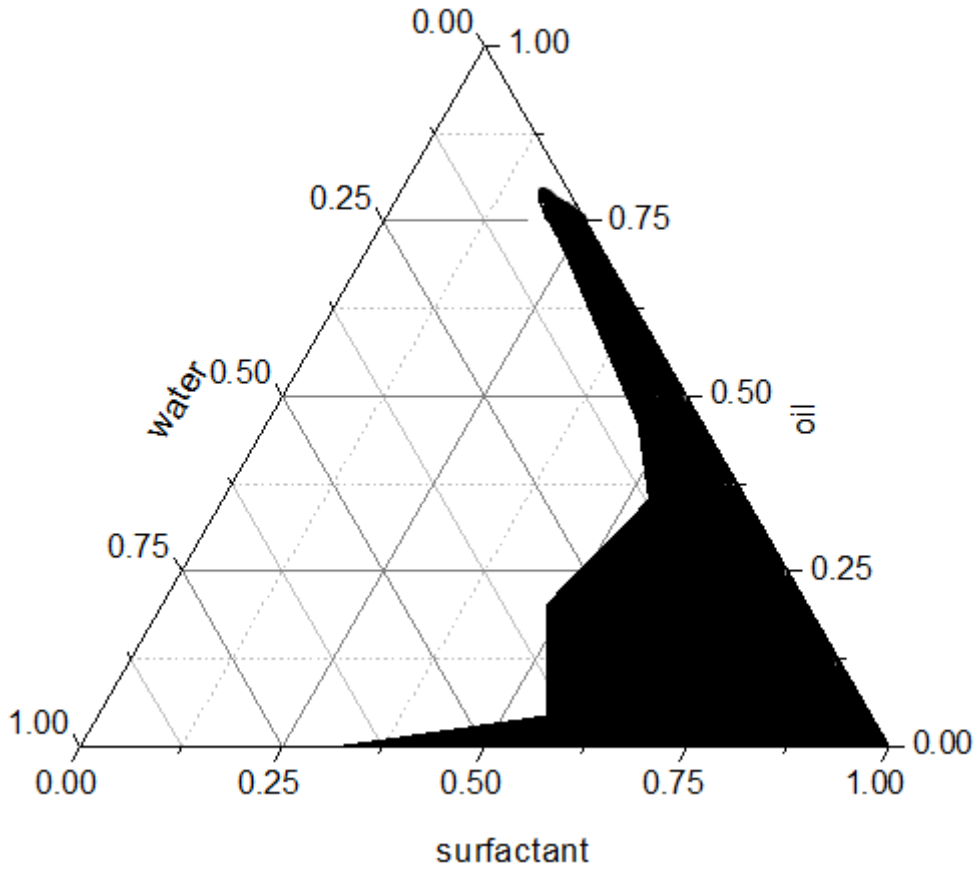


Figure #21: oleic acid with KTZ , (tween 80 , ethanol (2:1)),double distilled water

4.2.4 Pseudo ternary phase diagram A3

Consist of Tween 80:ethanol (3:1)in which the ethanol was used as cosurfactant and oleic acid oil with KTZ and double distilled water .This system produced a phase diagram with two regions which demonstrates that the microemulsion at ambient temperature was obtained and started as a single clear, isotropic and not shiny solution. Another region is liquid crystal region at ambient temperature which was observed by polarizer light microscopy and started as single phase,clear viscous and shiny.

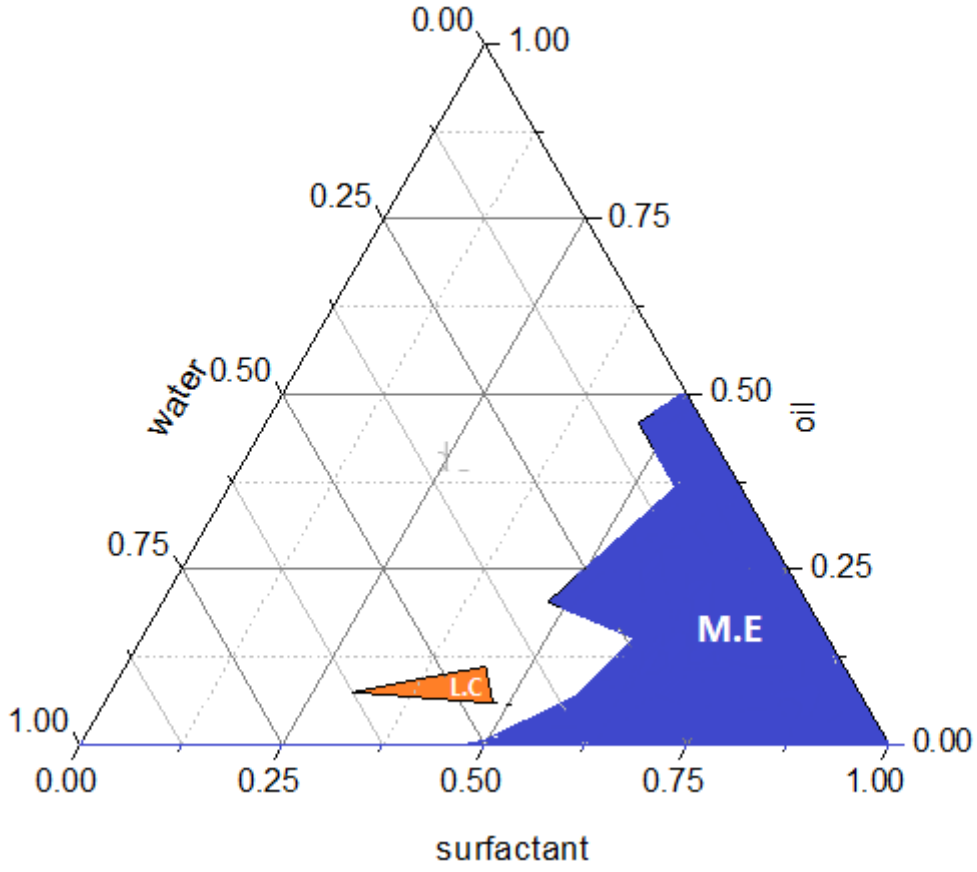


Figure #22 :Oliec Acid with KTZ , (Tween 80 : Ethanol (3:1)) , doubled distilled Water

4.2.5 Pseudo ternary phase diagram B

Consist of Tween 80 and oleic acid oil with KTZ and double distilled water with propylene glycol (1:1) which was used as co-solventwhich helps to dissolve relatively high concentrations of surfactants as well as lipid soluble drugs. Hence co-solvents are also considered as co-surfactants, demonstrates that the microemulsion at ambient temperature was obtained and started as a single phase, clear, isotropic and not shiny solution. While the liquid crystal region in this system was achieved and started as one phase ,clear ,viscous and shiny which was observed by polarizer light microscopy.

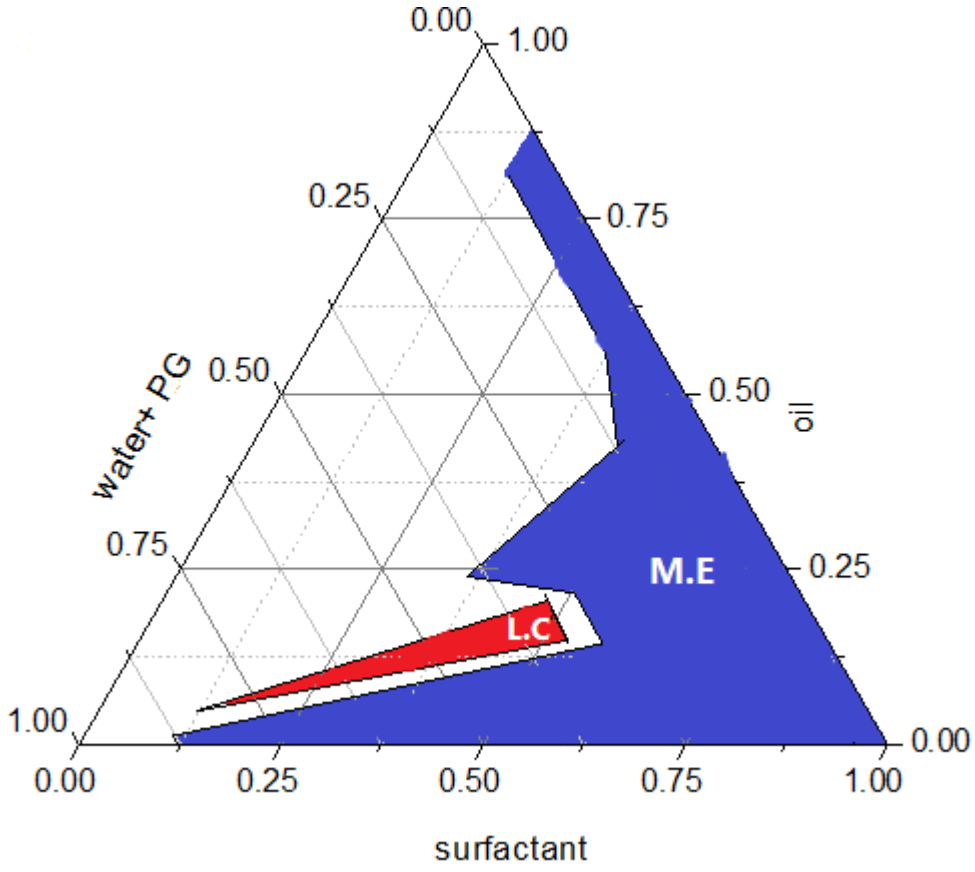


Figure #23: Oleic acid with KTZ , Tween 80 , (double distilled Water + PG)

After studying the phase behavior of the all systems of microemulsion we conclude that the area covered under the microemulsion was found to be larger in presence of higher amount of surfactant than cosurfactant. The surfactant\cosurfactant ratio (3:1) was found to be more efficient than the ratio (1:1) in formation of isotropic region and produced liquid crystal region. The efficiency of the surfactant\cosurfactant mixture decreased with decreasing amount of surfactant.

After studying the phase behavior of the system A and system B of microemulsionwhich their systems produced a phase diagram with two regions but the area covered under the microemulsion and liquid crystal regions were found to be larger in presence co-solvent as PG in system B than absence co-solvent in system A.

After studying the phase behavior of the systems A,A3,B we noted that their systems produced a phase diagram with two regions microemulsion and liquid crystal which we can produce good carrier for drug as liposomes .

4.3 Comparison between result in my study and results in previous studies

In my thesis i used ketoconazole as API and sebacicacid ,suberic acid as coformer to produce co-crystal of ketoconazole , suberic acid and sebacic acid using solvent crystallization method. The aim of my study was improvement the solubility for ketoconazole after cocrystal of ketoconazole and coformer was produced, we characterized them by melting point, DSC ,FTIR and solubility test .

After studying the melting point, DSC,FTIR and solubility for cocrystal, it was found that m.p was lower compared with ketoconazole and coformer alone , and DSC,FTIR confirm the formation of cocrystal whereas solubility test was appeared the improvement in solubility of ketoconazole with coformer compared with ketoconazole alone . In previous study the researcher used different coformer with ketoconazole such as ascorbic acid, oxalic acid,

furamic acid and nicotinamide, And different method to produce ketoconazole co-crystal slurry method, Solid dispersions method, which characterized the cocrystal with different coformer was found the melting point was less compared with the ketoconazole and coformer alone. The solubility for ketoconazole –ascorbic acid cocrystal was showed an increased by 50 times compared to ketoconazole alone. Whereas the solubility was shown to increase by 100times for ketoconazole with(fumaric and adipic acids) cocrystal compared to ketoconazole alone .

In the second part of my thesis the microemulsion result showed the ketoconazole solubility in oleic acid (oil phase) is good .

After studying the phase behavior of the all systems of microemulsion we conclude that the area covered under the microemulsion was found to be larger in presence of higher amount of surfactant than co-surfactant. The surfactant\cosurfactant ratio (3:1) was found to be more efficient than the ratio (1:1) in formation of isotropic region and produced liquid crystal region . The efficiency of the surfactant\cosurfactant mixture decreased with decreasing amount of surfactant .

In previous study ,the pseudo ternary phase diagrme with various weight ratios of tween80 and propylene glycol ,the transparent microemulsion region is presented in phase diagrame no distinct conversion from o/w to w/o , microemulsion was observed no liquid crystal was observed using cross polarizer ,the area of microemulsion isotropic region changed slightly in size with the increasing ratio of surfactant to co-surfactant.

5.Conclusion

This study aims to improve the solubility of KTZ using crystal engineering technique. Among these,co-crystal formation of KTZ was developed with coformers namely, subreic acid and sebacic acid .Solvent crystallization method allowed the formation of co-crystals of KTZ in 1:1 ratio. was tested by using FTIR,DSC, melting point and the solubility in water, Changes in FTIR values of bands, and DSC transforms in melting transitions, which corroborated the development of co-crystals and also equally supported each other among instrumental techniques. The co-crystals showed enhanced solubility. The prepared KTZ cocrystals can be formulated into various dosage forms. Solvent crystallization method can be a feasible technique and may be applied to various other therapeutically important poorly water-soluble molecules.

Also in My research used one of the most important sugar-based surfactants, sorbitan esters, which is used in pharmaceutical drugs such as Polyoxyethylenesorbitanmonooleate (Tween 80), and the structural feature provides unique physicochemical properties to these surfactants. On other hand, the co-surfactant with short chain of alcohol like ethanol was used as tuning parameter for all ingredients and contributes clearly in forming the microemulsion. Also, the addition of different amounts of short chain co-surfactants helps in the initiation of formation of drug product microemulsion in the formula of water, ethanol and Tween 80. The microemulsion in the five phase diagrams obtained gives clear isotropic, and not shiny solution upon the addition of 4% surfactant at ambient temperatures.The microemulsionchange at different ratio of co-surfactant.The decrease in the amount of co-surfactant will have an

effect on the nonionic surfactant micelle structure (W/O), and give two regions ofmicroemulsion and liquid crystal .

6. Recommendations

- To do the antifungal test to prove the efficiency of the KTZ suberic acid and sebacic acid cocrystals .
- To do the SEM and TEM tests, tocharacterize the cocrystal more perfectly.
- To use the uv-visible spectrophotometer in order to give more precise results of solubility.
- To make liposome from LC according to the results of microemulsion of KTZ , because the liposome is a good drug carrier which increase the efficiency of the drug.
- To make in-vivo tests of KTZ microemulsion to demonstrate the efficiency and safety of the KTZ new cocrystals.

7. References

1. Wiener, H. J.,Structural Determination of Paraffin Boiling Points, Am. Chem. Soc. 1947, 69, 17-20.
2. Wiener, H. J.,Correlation of Heats of Isomerization, and Differences in Heats of Vaporization of Isomers, Among the Paraffin Hydrocarbons, Am. Chem. Soc. 1947, 69, 2636-2638.
3. D'Amato, R.J.; Loughnan, M.S.; Flynn, E.; Folkman, J. Proc. Nati. Acad.Thalidomide is an inhibitor of angiogenesis Sci. USA 1994, 91, 4082-4085.
4. Bernstein, J. J. crystal growth polymorphism and structure –property relationship in organic crystal ,Phys. D: Appl. Phys. 1993, 26, B66
5. Desiraju, G.R.Polymorphism: The Same and Not Quite the Same,Cryst. Growth Des. 2008, 8, 3-5
6. Redinha, J.S.; Lopes Jesus, A.J.; Pais, A.A.C.C.; Almeida, J. A. S. Crystallization: From the Conformer to the Crystal, Advanced Topics on Crystal Growth, InTech, 2013.
7. A.D.; Boese, R.; Desiraju, G.R. Angew.On the polymorphism of aspirin: crystalline aspirin as intergrowths of two "polymorphic" domains , Chem. Int. Ed. Engl. 2007, 46, 618-622
8. Lehn, J.M.The Design of Molecular Hosts, Guests, and Their Complexes, Nobel Lecture, December 8, 1987.
9. Desiraju, G. R. Chemistry Beyond the Molecule,Nature 2001, 412, 397-400 .
10. Lehn, J.M. Supramolecular Chemistry: Concepts and Perspectives, VCH, Weinheim, 1995.

11. Lehn, J.M.Dinuclearcryptates: dimetallicmacropolycyclic inclusion complexes: concepts - design – prospects, Pure & Appl. Chem. 1980, 52, 2441-2459
12. Sekhon BS, Pharmaceutical co-crystals - a review ,Ars Pharm, 2009, Vol.50 nº 3; 99-117.
13. Zhang, C.; Yang, Z.; Zhou, X.; Zhang, C.; Ma, Y.; Xu, J.; Zhang, Q.; Nie, F.; Li, H.Evident Hydrogen Bonded Chains Building CL-20-Based Cocrystals,Cryst. Growth Des. 2014, 14, 3923-3928 .
14. Dunitz, J.D.Crystal and co-crystal: a second opinion,CrystEngComm 2003, 5, 506-506.
15. Desiraju, G.R.Crystal and co-crystal,CrystEngComm 2003, 5, 466-467.
16. Bond, A.D.What is a co-crystal?,CrystEngComm 2007, 9, 833-834
17. Aakeröy, C. B.; Salmon, D. J.Building co-crystals with molecular sense and supramolecular sensibility,CrystEngComm 2005, 7, 439-448.
18. Chang, Y.C.; Chen, Y.D.; Chen, C.H.; Wen, Y.S.; Lin, J.T.; Chen, H.Y.; Kuo, M.Y.; Chao, I. J.Quantitative Insights into the Crystal Structures of Nitro Derivatives of Ethyl (2E)-2-cyano-3-phenylprop-2-enoate: Inputs from X-Ray Diffraction, DFT Calculations and Hirshfeld Surface Analysis, Org. Chem. 2008, 73, 4608-4614.
19. Madura, I. D.; Czerwińska, K.; Jakubczyk, M.; Pawełko, A.; Adamczyk-Woźniak, A.; Sporzyński, A.Structural and Energetic Landscape of Fluorinated 1,4-Phenylenediboronic Acids,Cryst. Growth Des. 2013, 13, 5344-5352.
20. Meazza, L.; Foster, J. A.; Fucke, K.; Metrangolo, P.; Resnati, G.; Steed, J. W.Halogen-bonding-triggered Supramolecular Gel Formation, Nature Chem. 2013, 5, 42-47;

21. Präsang, C.; Whitwood, A.C.; Bruce, D.W.Halogen-Bonded Cocrystals of 4-(N,N-Dimethylamino)pyridine with Fluorinated Iodobenzenes,Cryst. Growth Des. 2009, 9, 5319-5326.
22. Skovsgaard, S.; Bond, A. D.Structure-directing factors when introducing hydrogen bond functionality to metal–organic frameworks,CrystEngComm 2009, 11, 444-453.
23. Glidewell, C.; Ferguson, G.; Gregson, R. M.; Lough, A. J. ActaCrystallogr., Sect. C.Co-crystals: Preparation, Characterization and Applications ,Cryst. Struct. Commun. 1999, 55, 2133-2136.
24. Wegner, F.M.; Lerner, H.W.; Bolte, M. Acta,hydrogen- and halogen-bond driven co-crrystallization form fundemuntal supramolecular chemistry to practical material scince ,Cryst. 2010, C66, o182-o184.
25. Arunan, E.; Desiraju, G.R.; Klein, R.A.; Sadlej, J.; Scheiner, S.; Alkorta, I.; Clary, D.C.; Crabtree, R.H.; Dannenberg, J.J.; Hobza, P.; Kjaergaard, H.G.; Legon, A.C.; Mennucci, B.; Nesbitt, D.J.Definition of the hydrogen bond , Pure Appl. Chem. 2011, 83, 1619-1636.
26. Seto, C. T.; Whitesides, G. M. J. Am.Comprehensive Coordination Chemistry II: From Biology to Nanotechnology,Chem. Soc. 1991, 113, 712-713 .
27. Aakeröy, C. B.; Seddon, K. R.The hydrogen bond and crystal engineering, Chem. Soc. Rev. 1993, 22, 397-407
28. Tiekink, E. R. T.; Vittal, J.; Zaworotko, M. Organic Crystal Engineering: Frontiers in Crystal Engineering, Wiley, John & Sons Inc., 2010.
29. Etter, M. C. Acc. Current Challenges on Large Supramolecular Assemblies ,Chem. Res. 1990, 23, 120-126;
30. Etter, M. C. J.Current Challenges on Large Supramolecular Assemblies, Phys. Chem. 1991, 95, 4601-4610.

31. Donohue, J. J. The Hydrogen Bond in Organic Crystals,Phys. Chem. 1952, 56, 502-510.
32. Etter, M.C. J. Am. Electron delocalization in mixed-valence molybdenum polyanions ,Chem. Soc. 1982, 104, 1095-1096.
33. Taylor, R.; Kennard, O. Acc.Analysis of Hydrogen Bonds in Crystals, Chem. Res. 1984, 17, 320-326.
34. . Dale, S.H.; Elsegood, M.R.J.; Hemmings, M.; Wilkinson, A.L.The co-crystallisation of pyridine with benzenepolycarboxylic acids: The interplay of strong and weak hydrogen bonding motifs,CrystEngComm 2004, 6, 207-214.
35. N. Rodrı´guez-Hornedo, S. J. Nehm, and A. Jayasankar. Cocrystals:Design, Properties and Formation Mechanisms, Encyclopedia of Pharmaceutical Technology, 2006 .
36. Michael J. Zaworotko, Molecules to Crystals, Crystals to Molecules ... and Back Again, CRYSTAL GROWTH & DESIGN 2007 VOL.7,NO.1 4-9.
37. Jayasankar A, Somwangthanaroj A, Shao ZJ, Rodrıguez-Hornedo N. Co-crystal formation during cogrinding and storage is mediated by amorphous phase. Pharm Res 2006; 23: 2381-2392.
38. Rodríguez-Hornedo N, Nehm SJ, Jayasankar A, Cocrystals: design, properties and formation mechanisms, In Encyclopedia of Pharmaceutical Technology, 3rd ed, Taylor & Francis, London, 2007, 615-635.
39. Indra Indra,FitriMiftahulJanah, RatihAryani Enhancing the Solubility of Ketoconazole via Pharmaceutical Cocrystal, J. Phys.Conf ,2019. Ser. 1179 012134
40. Remenar JF, Morissette SL, Peterson ML, Moulton B, MacPhee JM, Guzmán H, Almarsson O, Crystal engineering of novel co-crystals of a triazole drug with 1,4-dicarboxylic acids. J Am ChemSoc 2003; 125:8456-8457.

41. R. N. Kamble,C. Bothiraja1 · P. P. Mehta · V. Varghese, Synthesis, solid state characterization and antifungal activity of ketoconazole cocrystals Journal of Pharmaceutical Investigation,2017.
42. E.I. Korotkova, B. Kratochvíl, Pharmaceutical cocrystals, Procedia Chem. 2014, 10, 473–476, doi:10.1016/j.proche.2014.10.079.
43. S.R. Fukte, M.P. Wagh, S. Rawat, Coformer selection: an important tool in cocrystal formation, Int. J. Pharm. Pharm. Sci. 2014,6, 1–6.
44. FUKTE SULBHA et al, COFORMER SELECTION: AN IMPORTANT TOOL IN COCRYSTAL FORMATION. Int J Pharm PharmSci, 2014, Vol 6, Issue 7, 9-14
45. Choi, J. Y.; Park, C. H.; Lee, J. Bioavailability Enhancement of Poorly Water-Soluble Drugs via Nanocomposites: Formulation–Processing Aspects and Challenges,Drug Dev. Ind. Pharm. 2008, 34(11), 1209−1218.
46. Aggarwal AK, Jain S. Physicochemical characterization and dissolution study of solid dispersions of ketoconazole with nicotinamide. Chem Pharm Bull 2011;59:629-38.
47. Martin Flavia , Mihaela M. Pop, Gheorghe Borodi, Xenia Filip, and Irina Kacso Ketoconazole Salt and Co-crystals with Enhanced Aqueous Solubility, ACS pub, Des. 2013, 13, 4295−4304.
48. Pradip Kumar Ghosh, Rita J. Majithiya, Manish L. Umrethia, and Rayasa S. R. Murthy,Design and Development of Microemulsion Drug Delivery System of Acyclovirfor Improvement of Oral Bioavailability, AAPS PharmSciTech. 2006 Sep; 7(3): E172–E177.

49. Ibrahim Kayali, KhawlaQamhieh, Ulf Olsson, Lada Bemert, andReinhardStrey, Water-Diesel Micro emulsion s Stabilized by an Anionic Extended Surfactant and a Cationic Hydrotrope, Journal of Dispersion Science and Technology, 2012,33(4):516–520 .
50. 49Hire n. N,etal, microparticulate drug delivery system for topical administration of itraconazole, asian journal of Pharmaceutic , 2007,Volume 1, Issue 1, April - June,
51. Chen, J., Ma, X. H., Yao, G. L., Zhang, W. T., & Zhao, Y.,Microemulsion-based anthocyanin systems: effect of surfactants, cosurfactants, and its stability. International journal of food properties,2018, 21(1), 1152-1165.
52. Callender, S. P., Mathews, J. A., Kobernyk, K., &Wettig, S. D. Microemulsion utility in pharmaceuticals: Implications for multi-drug delivery. International journal of pharmaceutics,2017, 526(1-2), 425-442.
53. Maqsood Ahmad Malik, Mohammad YounusWani, Mohd Ali Hashim,Microemulsion method: A novel route to synthesize organic and inorganic nanomaterials, Arabian Journal of Chemistry,2012, Volume 5, Issue 4, Pages 397–417
54. Park ES, Yun BL, Ko In-Ja and Chi SC, Transdermal Delivery of Piroxicam Using Microemulsions, Arch.Pharm. Res., 2005, 28 (2), 243-248.
55. Mishra, A., Panola, R., & Rana, A. C. Microemulsions: As drug delivery system. J. SciInnov Res,2014, 3(4), 467-474.
56. Chaudhari V.M, Patel M.S, Patel M.R, Microemulsion Based Gel: A Review, International Journal Of Universal Pharmacy And Bio Sciences, 2014 (4).
57. Gadhave A.D, Waghmare J.T, A ,Short Review On Microemulsion And Its Application In Extraction Of Vegetable Oil, International Journal Of Research In Engineering And Technology, 2014, 2321-7308

58. Kumar P, Mittal KL. Handbook of Micro emulsion Science and Technology; 1st Edition; CRC Press, New York,1999

59. Tang JL, Sun J, He ZG. Self-Emulsifying Drug Delivery Systems: Strategy For Improving Oral Delivery of Poorly Soluble Drugs. Current Drug Therapy.2007;2(1):85-93.http://dx.doi.org/10.2174/157488507779422400.

60. Richard J. Farn, "Chemistry and Technology of Surfactants" by Blackwell Publishing Ltd,2006

61. Gautam S, Singharun K, Self Nanoemulsifying Drug Delivery System-A Novel Approach For Approach For Improving Bioavailability, Journal Of Drug Delivery And Therapeutics, 2013 (6), 33-38.

62. Drew Myers, , "Surfactant Science and Technology".3 Th Ed., John Wiley & Sons, Inc., Hoboken, New Jersey.2006

63. Jha S. K. Dey S, KarkiR.Microemulsion-Poential Carrier For Improved Drug Delivery ,Asian Journal Of Biomedical And Pharmaceutical Sciences, 2011 (1),5-9.

64. Panwar A.S, Upadhyay N, Bairagi M, Gujar S, Darwhekar G.N, Jain D.K, Emulgel: A Review. Asian Journal Of Pharmacy And Life Science, 2011

65. Tang JL, Sun J, He ZG. Self-Emulsifying Drug Delivery Systems: Strategy For Improving Oral Delivery of Poorly Soluble Drugs. Current Drug Therapy. 2007;2(1):85 93. http://dx.doi.org/10.2174/157488507779422400.

66. Talegaonkar S, Azeem A, Ahmad FJ, Khar RK, Pathan SA, Khan ZI. Micro-emulsions: A Novel Approach to Enhanced Drug Delivery. Recent Patents on Drug Delivery&Formulation. 2008;2(3):238-257.

.http://dx.doi. org/10.2174/187221108786241679.

67. Bipin SarveshKatiyar,etal, A Novel Drug Carrier System,Int. J. Pharm. Sci. Rev. Res., 20(2), May – Jun 2013; n° 24, 138-148.
68. Ghosh PK, Murthy RS, "Microemulsions: A potential drug deliverysystem",Current Drug Delivery,2006, 3(2):167-80.
69. Hiendrawan.s, Hartanti.a , Vriansyah.b, widjojokusumo.e, raymond r..solubility enhancement of ketoconazole via salt and cocrystal formation,int j pharm pharm Sci,2015,7(7),pp.0975-1491.
70. Shayanfar.A ,Jouyban.A. Physicochemical characterization of a new cocrystal of ketoconazole,ScienceDirect,2014,10,pp. 242–248.
71. Fatemeh Keramatnia,et al, Ketoconazole ionic liquids with citric and tartaric acid: Synthesis, characterization and solubility study.Fluid Phase Equilibria,2014,425, pp108-113
72. Patel,Mrunali et al , investigating the effect of vehicle on in vitro skin permeation of Ketoconazole applied on O\W microemulsions, actapharmaceuticasciencia ,2010,52: 65-77
73. Tiwari, N., Sivakumar, A., Mukherjee, A. and Chandrasekaran, N.. Enhanced antifungal activity of Ketoconazole using rose oil based novel microemulsion formulation. Journal of Drug Delivery Science and Technology,2018, 47, pp.434-444

تحسين الذائبية للدواء (ketoconazole) عن طريق استخدام البنية البلورية في دمج الأحماض المستخدمة والاستحلاب

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الملخص

تهدف هذه الدراسة إلى تصنيع وتوصيف الكريستال الشريك مع الكيتوكونازول لتحسين ذائبية الكيتوكونازول، هذا الكريستال الشريك للكيتوكونازول تشكل عن طريق التبلور من خلال استخدام مذيب معين مع أحماض تساعد على التبلور، ويمكن الكشف عن هذا الكريستال الشريك باستخدام مطياف الأشعة تحت الحمراء (FTIR)،مسعر المسح التفاضلي (DSC)، وفحص مدى درجة الانصهار ، وفحص الذائبية، ويتميز الكريستال المشترك الجديد ببلورات رائعة، ويُظهر طيف (FTIR) الترابط الهيدروجيني في الكريستال المشترك ، مما يمنحه خصائص فيزيائية أفضل (مثل الذوبان ودرجة الانصهار) مقارنة بالكيتوكونازول لوحده.

الكريستال المشترك للكيتوكونازول برهن على ذائبية محسنة بنسبة عالية مقارنة مع الكيتوكونازول لوحده. ويظهر هذا البحث أن الكريستال المشترك بين الكيتوكونازول وحمض السوبريك Suberic) (acidوحمض السيباسيك(Sebacic acid) ومذيب قد تم تشكيلهما بشكل فعال وقد تم تعزيز قابلية ذوبان الدواء.

بالإضافة إلى ذلك، يهدف هذا البحث إلى تحضير المستحلب الصغير للكيتوكونازول بتركيبات مختلفة مطبقة باستخدام الحد الأدنى من مركب يقلل التوتر السّطحي للسائل المذاب به في درجة الحرارة المحيطة عن طريق استخدام المعايرة المائية.

وايضاً تم دراسة تأثير النسب المئوية المختلفة من المواد الخافضة للتوتر السطحي والمذيب المشترك على سلوك الطور للأنظمة المقترحة على درجة الحرارة المحيطة للنظام. وهذا المركب الذي تم استخدامه في هذا البحث كمادة خافضة للتوتر السطحي هو توين 80 (بولي أوكسي إيثيلين أحادي السوربتان). ويستخدم الكحول قصير السلسلة كعامل مساعد للذائبية للسطوح المنشطة وهو الإيثانول في هذا البحث. طور الزيت المستخدم هو حمض الأوليك. وطور الماء هي ماء مزدوج التقطير مع بروبيلين غلايكول حيث يعمل كمذيب مشترك. حيث بينت الدراسة الصيغة التي أظهرت أن المنطقة المغطاة تحت المستحلب الدقيق أكبر في وجود كمية أعلى من الفاعل بالسطح من الفاعل بالسطح المشترك.(surfactant:cosurfactant) تم العثور على نسبة الفاعل بالسطح/ الفاعل بالسطح المشترك (3: 1) لتكون أكثر كفاءة من النسبة (1: 1) في تكوين منطقة مستحلب دقيق متناحي ومنطقة بلورية سائلة منتجة. وأيضاً أعطت التركيبة التي تحتوي على البروبيلين جلايكول كمذيب مشترك مساحة أكبر من البلورة السائلة والمستحلب الدقيق مقارنة بغياب المذيب المشترك.