REVIEW ARTICLE

Biocompatible microemulsion systems for drug encapsulation and delivery

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This review of biocompatible lipophilic drug encapsulating and delivery systems entails the basics of colloidal drug delivery with special reference to oil-in-water microemulsion. The behaviour of the biocompatible systems, their formulation and ingredients, physicochemical properties, applications through different routes, advantages and restrictions therein have been presented. *In vitro* and *in vivo* biological studies using typical formulations in different laboratories have been taken into account to show their current potential and future prospects.

Keywords: Biocompatible microemulsion, drug encapsulation and delivery, surfactant.

THE 20th century has witnessed a remarkable growth in drug discovery, development and use. But towards the end of the century, toxic side effects of the drugs became apparent in addition to their therapeutic effects, due to several reasons. Drugs which do not fulfil the 'magic bullet' concept of Paul Ehrlich (wherein the drugs are conceptualized as targeting the infected sites only, leaving aside healthy cells, tissues and organs) cause adverse side effects due to unwanted biodistribution. Incomplete absorption after *in vivo* administration leads to toxicity from lipophilic drugs. Some hydrophilic drugs and proteins are destroyed due to exposure to enzymes when administered orally. Probable remedies could be to target the drugs using site-specific ligands or to disperse them in suitable delivery systems so as to enhance their uptake/absorption after *in vivo* administration and also to protect them from enzymatic degradation.

Over the past few decades colloidal systems have been explored as potential delivery systems[1–4], because of their compartmentalized hydrophobic and hydrophilic domains (cavities), where both polar and non-polar molecules could be encapsulated and stabilized.

The dispersed systems[1–3] studied for the delivery of different kinds of drugs are liposome[4], noisome,[5] nanoparticle[6,7], microemulsion[8–11], hydrogel[12–14], organogel[15,16], lipid dispersion[17,18], microsphere[19], micelle[20,21], polymeric micelle[22,23] and cationic polymer[24–26].

Drug delivery research evaluates the potentials and benefits of synthetic gene carriers for gene therapy. Gene therapy provides a great opportunity for treating diseases from genetic disorders, infection, cancer, etc. The therapy involves the delivery of a particular gene to the targeted cells, thus fighting the disease at the point of its origin. There are two essential components in current gene therapy: (i) an effective therapeutic gene that can be expressed at a target site, and (ii) an effective and safe delivery system which delivers the therapeutic genes that can be expressed to a specific target tissue or organ. Gene delivery systems are designed to protect the genetic materials from premature degradation in the systemic blood stream and efficiently transfer the therapeutic genes to target cells.

Major attention is being paid to cationic polymers which are able to both condense large genes into smaller structures and mask the DNA charges. Polymeric nano (micro) particles (based on polycyanoacrylate, poly(D,L-lactic acid), gelatin, alginate, chitosan) which absorb or encapsulate oligonucleotides or genes are under study as sustained release matrices for generic drugs[24]. Recent developments in the design of cationic lipids and their applications in gene delivery are discussed in detail by Bhattacharya and Bajaj[25], where various structure–activity studies explaining the variations in gene transfection efficacies with respect to different molecular structures of the cationic lipids are discussed. The review presents gene transfer abilities in relation to aggregation properties of different aqueous formulations such as cationic liposomes and surfactant aggregates from various amphiphiles and cationic lipids as a function of their hydrophilic parts, linkers and head groups.

The clinical success of gene therapy is critically dependent on the development of efficient and safe delivery systems (transfection vectors, viral and non-viral). Although viral vectors have advantages over non-viral vectors in terms of gene transfer efficiency, non-viral vectors are the choice because of several advantages over the viral vectors. A review by Karmali and Choudhuri[27] highlights the major achievements of cationic liposomes as non-viral carriers of gene medicines. Gene therapy is considered as a suitable substitute for conventional protein therapy, since it can overcome inherent problems associated with protein drugs in terms of bioavailability, systemic toxicity, *in vivo* clearance rate and manufacturing cost.

The review by Park *et al.*[28] explores the recent developments of polymeric gene carriers and presents the future direction for the application of polymer-based gene

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delivery systems in gene therapy. Synthetic gene delivery vectors are gaining increasing importance as an alternative to recombinant viruses. Among the various types of non-viral vectors, cationic lipids are especially attractive as they can be prepared with relative ease and are extensively characterized[29]. Lipids with specifically tailored molecular architecture have been successfully employed as gene delivery vehicles for controlled drug release and the preparation of supramolecular gels. Designing of lipids and their characterization upon membrane formation helps in understanding the molecular basis of their properties[30].

Significant effort has been devoted to develop nanotechnology for drug delivery since it offers a suitable means of delivering small molecular weight drugs, and macromolecules such as proteins, peptides or genes by either localized or targeted delivery to the tissue of interest. Nanotechnology focuses on formulating therapeutic agents in biocompatible nanoconjugates. Nanoparticles are submicron-sized colloidal dispersions with the therapeutic agents encapsulated in their matrix or adsorbed or conjugated onto their surface[31]. Because of their small size, risk of capillary embolism is minimum. They can be used to provide targeted (cellular/tissue) delivery of drugs for intravascular delivery, and to improve the stability of therapeutic agents against enzymatic degradations[32]. Due to their sub-cellular and sub-micron size, nanoparticles can penetrate deep into tissues through fine capillaries and are generally taken up efficiently by the cells[33]. Biodegradable nanoparticles formulated from poly(D,L-lactide-*co*-glycolide; PLGA) have been extensively studied for sustained and targeted localized delivery of different agents, including plasmid DNA, proteins and peptides, and low molecular weight compounds. Panyam and Labhasetwar[34] have studied various potential applications of PLGA nanoparticles for delivery of therapeutic agents to the cells and tissues.

Polymeric micelles formed through the multimolecular assembly of block co-polymers are studied for drug and gene targeting. These carrier systems have been receiving much attention in the field of drug targeting because of their high loading capacity for poorly soluble pharmaceuticals[22]. Polymer–lipid amphiphilic compounds such as diacyl-lipid-polyethyelene glycol are attractive systems because of their tendency to accumulate in the areas of tumours and infarct zones[23], in addition to their high loading capacity.

Liposomes have been widely studied since 1970 as drug carriers[4] for improving the delivery of therapeutic agents to specific sites in the body and almost immediately were explored for cancer treatment[35]. One of the serious obstacles limiting therapeutic applications of liposomes is the short circulation residence time as a result of their elimination from the blood stream by the reticuloendothelial system after systemic administration. This problem was partially solved by covering the liposome surface with phosphoethanolamine derivatives of poly(ethyleneglycol) (PEG) or other polymers. These neutral and sterically stabilized liposomes (stealth liposomes)[36] used as carriers for hydrophilic anticancer drugs exhibited really long circulation time in the vascular system and showed enhanced accumulation of drugs in tumour tissues[4]. 'Neutral' and cationic liposomes were used for delivery of doxorubicin (DOX), antisense oligonucleotide (ASO) and small interfering RNA (siRNA). These liposomes provided an efficient intracellular delivery of DOX, ASO, siRNA *in vitro*. Intratracheal delivery of both types of liposomes *in vivo* led to higher peak concentrations and much longer retention of the drugs when compared with systemic administration[37].

The self-assembly of non-ionic surfactants into vesicles was first reported also in the seventies by researchers in the cosmetic industry[5]. Since then a number of groups worldwide have studied non-ionic surfactant vesicles (niosomes) with a view to evaluate their potential as a drug carrier. Niosomes may be formed from a diverse array of amphiphiles bearing sugar, polyoxyethylene, polyglycerol, crown ether and amino acid hydrophilic head groups. These amphiphiles typically possess one or two hydrophobic alkyl, perfluoroalkyl or steroidal groups. These systems have been evaluated as immunological adjuvants, carriers for anticancer drugs and anti-inflammatory drugs as well[38]. They are more stable than the liposomes[5].

Hydrogel technologies have spurred developments in many biomedical applications, including controlled drug delivery. Many novel hydrogel-based delivery matrices have been designed and fabricated to fulfil the needs of the pharmaceutical field. Mathematical modelling plays a key role in facilitating hydrogel network design by identifying key parameters and molecule-release mechanisms. Recent interest has grown in hydrogels that degrade in recognition of a cellular response[12–14].

Organogels are semisolid systems in which an organic liquid phase is immobilized by a three-dimensional network composed of self-assembled, inter-wound gelator fibres[15]. In the last decade interest in organogels has grown rapidly with the discovery and synthesis of a large number of diverse molecules, which can gel organic solvents at low concentrations[16]. The gelator molecules immobilize large volumes of liquid following their self-assembly into a variety of aggregates such as rods, tubules, fibres and platelets. Due to their thermoreversibility, they find wide application in the industry. However, only a few organogels are currently being studied as drug/vaccine delivery vehicles, as most of them are composed of pharmaceutically unacceptable compositions[15]. The organogels commonly used for drug-delivery studies are biocompatible microemulsion gels and lecithin gels for transdermal delivery, sorbitan monostearate organogels for vaccine adjuvants and gels composed of alanine derivatives for oral and transdermal delivery[16].

Microemulsion systems are ternary or pseudoternary dispersed systems comprising mixtures of oil, water and

surfactant or surfactant + cosurfactant. They possess some unique characteristics such as thermodynamic stability (imparting long shelf-life), compartmentalized polar and non-polar dispersed nano-domains, ease of preparation, low viscosity, ultralow surface tension and optical transparency (infrequently faint translucency)[2]. Due to the existence of both polar and non-polar micro-domains, both hydrophilic and lipophilic drug molecules could be solubilized[8–11], encapsulated and stabilized in these microscopically heterogeneous and macroscopically homogeneous systems. Over the last two decades, water-in-oil (W/O) and oil-in-water (O/W) systems are being studied as prospective drug-delivery vehicles. The water droplets in W/O microemulsions are good microreactors for model biochemical reactions. DNA condensation in such microcompartments is comparable to biomimetic systems. Swami *et al.*[39] have shown that DNA can be confined in W/O microemulsion using various techniques such as light, X-ray and neutron-scattering. They have studied in detail, DNA distribution and structural modification of these microemulsion drops by varying the concentration and molecular weight of DNA. The DNA induces formation of large drops into which it is internalized. The size of the drops depends on the amount of DNA dissolved in water, as well on its molecular weight. By choosing proper ω (=[water]/[surfactant]) for W/O microemulsion systems of iso-octane/sodium-bis ethyl hexylsulpho succinate (AOT)/water, a high local concentration of DNA (comparable to natural organelles) could be obtained. The large drops co-exist with small, empty drops (not containing DNA), similar to those found in DNA-free microemulsion. According to Hwang *et al.*[40], Cisplatin loaded in W/O microemulsion is active against bladder cancer cells. Microemulsions with Brij 98/terpeneol and water exhibited complete inhibition of proliferation. Gupta *et al.*[41] developed a series of novel plant oil-based microemulsion systems with clove oil, corn oil, cottonseed oil, eucalyptus oil, orange oil and peppermint oil and non-ionic surfactants. These systems have shown good biocompatibility and efficient and non-toxic delivery systems for bioactive natural products, Quercetine[42], Diospyrin[43] and Basic acid[44].

Microstructure and encapsulated drug

Microemulsion comprises different structures, O/W, W/O and bicontinuous, and these structural changes help in preferential release of the drug[45]. Because of the presence of hydrophobic and hydrophilic components as part of the structure, these systems may serve as vehicles for drugs of different solubilities. To study the drug-delivery potential of microemulsion vehicles, it is necessary to characterize the microstructure of pure and drug-loaded microemulsion[46]. The incorporated drug may or may not influence the microstructure. O/W or O/W microemulsions may show different behaviours for release of both hydrophilic and lipophilic drugs. Release would be fast for a hydrophilic drug in case of an O/W microemulsion, since it is in the continuum region. The diffusion is difficult when incorporated in W/O, since it gets trapped in water droplets. Reverse is true for hydrophobic drugs. Since the microemulsion is subjected to infinite dilution by body fluids (*in vivo* conditions), it is important to know drug release at infinite dilution. It is found from the literature that the release of drugs from the micelle and microemulsion systems is a diffusion-controlled process[47]. Physico-chemical analysis of microemulsion systems has showed the occurrence of structural changes from W/O to O/W[9].

Criteria for drug-delivery systems

Pharmaceutical applications necessitate certain criteria for drug-delivery systems[1]. They are: tolerance towards additives and stability over a wide temperature range, up to 4°C in the lower range (many drugs are required to be stored at low temperature), and 37–40°C in the upper range (comparable to the human body temperature). Small size, low viscosity, biocompatibility as well as biodegradability and easy elimination from the body are also essential. It is preferable that the delivery system is not recognized and sequestered by the immune system of the body or phagocytosized by liver. In order to avoid capillary blockage (embolism), the size of the encapsulated particles needs to be controlled. As the diameter of the smallest blood capillary is 400 nm, it is preferable that the diameter of the drug-delivery system is smaller than this dimension. Also, small entities (vehicles) can remain in circulation for a prolonged period of time as they can traverse through the minutest of blood capillaries to maintain the required level of pharmaceuticals in the blood[1].

Choice of excipients

From biocompatibility consideration, the choice of excipients is restricted[48]. Pharmaceutically accepted oils tend to be more polar and of much higher molecular weight than the more commonly used aliphatic or aromatic oils for non-pharmaceutical microemulsion. Usually, the oil which has maximum solubilizing potential for the drug is selected for the formulation of the microemulsion in order to achieve maximal drug loading[49,50]. Oils with long hydrocarbon chains (or high molecular volume) such as olive, peanut, soybean, canola and sunflower are difficult to microemulsify, whereas oils with shorter (or low molecular volume) such as medium chain triglycerides (MCT), medium chain mono- and diglycerides are easier to microemulsify. The choice of oil is often a compromise between solubility of the drug in the oil and microemulsification. But, the capacity of solubilizing lipophilic moieties usually increases with the

chain length of the oil[51]. Amongst the various oils, mono- and diglycerides are preferred for oral delivery due to their ability to enhance permeation across the biological membranes[52], whereas for parenteral administration MCTs and fatty acid esters are preferred[53]. Triglycerides[54,55] and esters of fatty acids, isopropyl myristate (IPM)[56,57], ethyl oleate[58] and oils of plant origin (corn, cottonseed, orange, clove, peppermint, eucalyptol and coconut) are also used as the oil phase[41–44,59] to prepare pharmaceutical microemulsions for use through different routes of *in vivo* administration.

Choice of surfactant is crucial for the formulation of microemulsions, because preparation of a microemulsion generally requires the use of moderate to high concentration of surfactant. Even pharmaceutically accepted surfactants have adverse side effects above the recommended concentration[11]. Naturally occurring surfactants, lecithin and related phospholipids are preferred over synthetic surfactants, but they always need a co-surfactant because of the strongly lipophilic nature and its tendency to form rigid lamellar phase[60]. But, microemulsions containing this class of surfactants show a potential increase in the permeability of the drug through biological membranes, which generally results in an enhanced intracellular drug concentration. The ionic surfactants that are commonly used for microemulsification cannot be used for pharmaceutical formulations because of toxicity, such as membrane perturbation and skin irritation[61]. Zwitterionic and nonionic surfactants are commonly used to formulate microemulsion systems because of their lower toxicity than ionic surfactants, and their greater stability towards change in ionic strength and pH, which is likely to be encountered after *in vivo* administration[9]. The commonly used synthetic, non-ionic surfactants are polysorbates[41] (Tweens), polyoxyethylene alkyl ethers[57] (Brij), polyoxyethelene stearate[62] (Solutol-15), polyoxyethylene hydrogenated castor oil[63] (Cremophor RH) and sorbitan esters[64] (Span). Low hydrophilic hypophilic balance (HLB) surfactants (such as sorbitan monoesters) are preferred for W/O microemulsions, whereas high HLB surfactants such as polysorbates 80 or 20 are preferred for O/W microemulsion[65]. A mixture of lipophilic (low HLB) and hydrophilic (high HLB) surfactants is sometimes useful[66]. Temperature exerts an effect on the formation and region of existence of microemulsions, as the HLB of the surfactants can change with temperature and destabilize the surfactant interface. Although polysorbates (polyoxyethylene sorbitan monoesters of fatty acids, such as Tween-20) have been widely reported as useful amphiphiles for the preparation of pharmaceutical microemulsions, only Tween-20 and Tween-80 have approval for oral ingestion, and lecithin offers a possible non-toxic alternative for parenteral use[63,64]. The use of non-ionic surfactants has been widely accepted, since along with being biocompatible, they retain their utility over a broad range of pH values[67].

Use of short-chain alcohols as co-surfactants is generally considered undesirable[65]. The pharmaceutically acceptable cosurfactants are ethanol[68], 1,2-alkanediols and alkyl monoglucosides[69,70], sucrose–ethanol mixtures[71], alkyl monoglucosides and geraniol[72]. Co-surfactants, e.g. plurol isostearique, have recently been introduced into topical microemulsion formulations[73,74]. A serious disadvantage of microemulsions containing co-surfactants is their instability on dilution with aqueous-based biological fluids, which generally occurs after *in vivo* administration through most routes of administration. Use of co-surfactants also causes irritancy. Microemulsion systems devoid of co-surfactants could be prepared by using double alkyl chain surfactants[75] and non-ionic surfactants[76]. For fruitful use of microemulsion systems in drug delivery, proper understanding of their physico-chemical properties (phase behaviour, tolerance towards temperature and additives, droplet-size distribution and biological tolerance) is a prerequisite.

Different biocompatible systems

Different biocompatible microemulsion systems for drug delivery have been developed and characterized. A brief account presented below.

A linear increment of droplet size with an increase in oil content at several fixed surfactant/co-surfactant concentrations was observed for O/W microemulsions of isopropyl myristate (IPM) as oil, polysorbate (P) 40, 60, and 80 as amphiphile and sorbitol (S) as co-surfactant. The results were interpreted using a hard sphere model[77,78]. A series of modified phospholipids (m-PCs) possessing different acyl chains in position 2 (butanoyl to hexadecanoyl) influenced the microemulsion and liquid crystalline domains of the O/W region of the phase diagrams, when used as a mixture with soy lecithin (SbPC) for a system with a medium of MCT or IPM as oil, ethanol as co-surfactant, and water. Decreasing the acyl chain length (increasing the hydrophilicity of the surfactant), showed a corresponding increase in the microemulsion area in the phase diagram[79]. For a pseudoternary system of IPM/SbPC/and six straight or branched alcohols (1-butanol, 2-butanol, isobutanol,1-pentanol, 2-pentanol, and 3-pentanol) as co-surfactants and aqueous solution of monoalkyl phosphate as the polar phase, the microemulsion domain was a function of monoalkylphosphate(hexyl or oleyl) concentration[80]. For IPM/soy lecithin + polysorbate 80/water systems at different polysorbate/lecithin weight ratio, most of the compositions showed a non-Newtonian rheological behaviour. Both droplet size and viscosity of the dispersed systems were much more influenced by the total oil content than the weight ratios of the amphiphile mixtures[81–84].

The influence of addition of a drug on thermodynamic stability of the two microemulsion systems of IPM/AOT/water and IPM/lecithin/water was studied by Fubini

et al.[85]. A linear relationship between enthalpy change and concentration of added co-surfactant butanol was observed at mole fractions below microemulsion formation. The addition of butanolic solutions of two drugs of different lipophilicity (Menadione and Prednisone) to the same mixture did not cause any alteration in enthalpy change[86]. For microemulsion systems of isopropyl palmitate(IPP)/ polyoxyethylene(10) oleyl ether (Brij-97) +1-butanol (2 : 1)/water, compositions with < 15% water consisted of reverse micelles, with 15–30% water were W/O, and with > 35% were O/W type microemulsion[87]. Non-toxic microemulsions prepared and studied by Kahlweit *et al.*[69,70] with ethyl oleate as oil, soyabean lecithin (Epikuron 200) as amphiphile and 1,2-alkane diols as co-surfactant showed good stability up to a temperature of 40°C.

The effect of four aliphatic alcohols (1-propanol, 1-butanol, 1-hexanol, and 1-octanol) and four 1,2-alkane diols (1,2-propane diol, 1,2-pentane diol, 1,2-hexane diol and 1,2-octane diol) on the pseudoternary systems of ethyl oleate/surfactant blend (sorbitan monolaurate + polyoxyethylene 20 sorbitan mono-oleate)/water indicated that the requirements of a co-surfactant to produce a balanced microemulsion were HLB value of 7.0–8.0, carbon backbone of 4–6 atoms, carbon percentage of 60–65 and oxygen percentage of 20–30 (refs 88, 89). Neubert and co-workers[90–92] prepared microemulsion systems (devoid of co-surfactant) using a blend of low and high HLB surfactants (Tagat-20, HLB = 15 and Ploxamar 331, HLB = 1), propylene glycol, IPP, oleic acid and Eutanol G as oil phase, and (propylene glycol + water) as aqueous phase[90–92]. Microemulsion systems for parenteral use were developed using MCT as the oil phase, SbPC and polyethylene glycol 660 (PEG660)-12 hydroxystearate (12HSA-EO_{15}) as amphiphile, and polyethylene glycol 400 (PEG400) and ethanol as co-surfactants[52]. The isotropic regions of the microemulsion comprising IPM/egg lecithin and soy lecithin (SbPC)/water and a series of short-chain alcohols (*n*-propanol, isopropanol, *n*-butanol, sec butanol, isobutanol, *tert*-butanol and *n*-pentanol) depended on the nature of the surfactant/ co-surfactant mixing ratio (w/w), but was independent of the lecithin source[93–95].

Rheological studies, physico-chemical studies, temperature effect and thermodynamics of microemulsification of (xylene + cholesteryl benzoate)/(sodium deoxycholate (NaDC) + butanol/water) and (heptane + cholesteryl benzoate)/TX-100 + butanol/water) were conducted by Moulik and co-workers[96,97]. The systems showed good stability with time and temperature. Microemusion zone of heptane-containing system depended appreciably on the surfactant/co-surfactant ratio, but that of the xylene-containing system did not. The product of equivalent conductance and viscosity (Walden product) increased sharply with increased water content, suggesting a special mechanism of conduction via 'channel' formation and percolation[98]. The dissolution process of water in AOT/ cinnamic alcohol (CA) and in Tween-20/CA forming W/O microemulsion and that of CA in Tween-20/water forming O/W microemulsion (studied calorimetrically) was exothermic accompanied by negative entropy changes indicating ordering effect, which was supported by the measured specific heats of the resulting preparations[99].

Vegetable and plant oils could be mixed with water in different proportions in the presence of non-toxic amphiphile yielding mono-, bi- and triphasic zones. Variation of temperature and addition of cholesterol, crown ether, urea and brine brought about striking changes in the phase behaviour of saffola (73% linoleic acid (v/v)/(sodium-bis-ethyl-hexyl sulfosuccinate (AOT) + hexylamine)/water system[100] with the different zones interchanging positions among themselves were observed. The conductance of the microemulsion at different water/amphiphile ratio (ω) showed unusual dependence on temperature with a significant degree of 'percolation', whereas dependency of viscosity on tempearure showed a normal declining trend[98]. Microemulsions of different plant oils (ricebran, saffola, soyabean sesame, palm and linseed) with water was possible[101] with AOT and CA as mixed amphiphiles, with ~ 27% clear zone. The extent of multiphasic (two- and three) zones depended on oil type and temperature. Compositions with high water content were fairly viscous and the viscosity of sesame and saffola-containing systems decreased with the rate of shear[97]. The mixing behaviour of ricebran, saffola and clove oil with water in the presence of surfactants TX-100, polyoxethylene sorbitan monolaurate (Tween-20), AOT, Igepal and sodium oleate and co-surfactants (ethanol and CA) was also studied. The system containing sodium oleate and ethanol showed maximum single phase zone[59]. Gupta *et al.*[41] studied microemulsification of various combinations of different vegetable oils, viz. corn oil, cottonseed oil, clove oil, orange oil and peppermint oil using several nonionic surfactants (Tween-20, Brij-30, and Brij-92) and co-surfactants, ethanol and isopropanol (Figure 1). Balanced microemulsion systems with significant amount of O/W as well as O/W regions were observed. Interestingly, surfactant-less microemulsification of peppermint oil and IPM in pure form and in mixed condition was possible with the addition of isopropanol only. All the systems showed excellent stability with time (1 yr) and temperature variation of 4–40°C. The size of the dispersed clove-oil droplets in water was in the range 10–20 nm. The phase boundaries of the ternary systems for clove oil were not altered with the addition of phosphate buffer saline (PBS, pH 7.2), Tris-HCl buffer (pH 7.4), Ringer lactate solution, urea solution (30 mg/ml^{-1}), glucose solution (100 mg ml^{-1}) and 0.9% saline, indicating good tolerance of the systems to these additives[42]. The anti-inflammatory drug Indomethacin encapsulated in clove oil/Tween-20/water O/W microemulsion showed better transdermal penetration compared to conventional gel formulation[102]; clove oil being a penetration enhancer augmented the process.

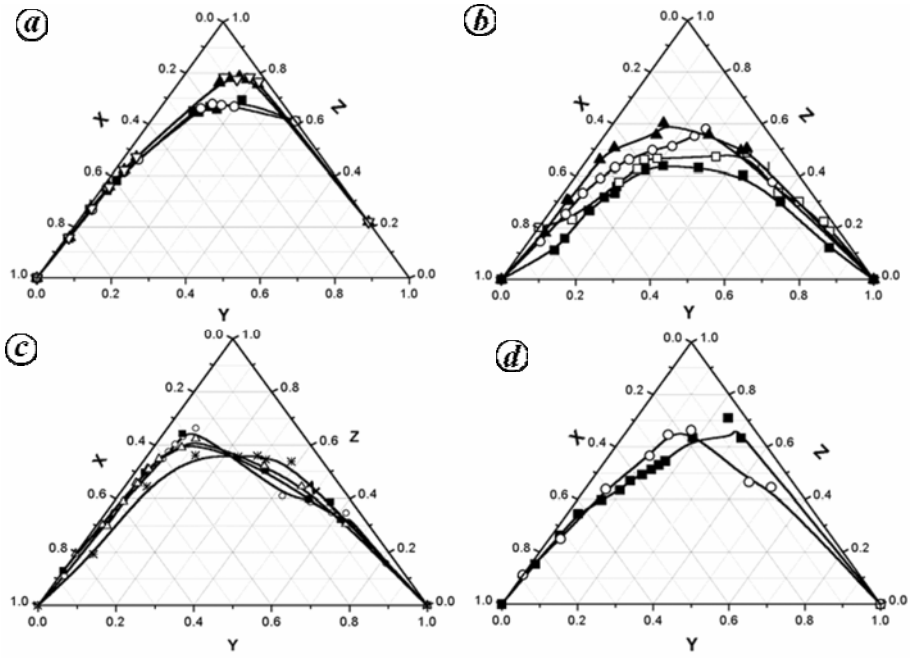

Figure 1. ***a***, Pseudo-ternary for different systems at 303 K. (■) Corn oil/Brij-92 + iPrOH (1 : 1 v/v)/water; (○) Cottonseed oil/Brij-92 + iPrOH (1 : 1 v/v)/water; (▲) Corn oil/Brij-30 + iPrOH (1:1v/v)/water; (Δ) Cottonseed oil/Brij-30 + iPrOH (1 : 1 v/v)/water. The scale magnitude is 1/100th of the actual. The axis representations are *X* = oil; *Y* = water and *Z* = surfactant + co-surfactant. ***b***, Ternary and pseudo-ternary phase diagrams for different systems at 303 K. (■) Clove oil/Tween-20/water; (▲) Orange oil/Tween-20/water; (■) Clove oil/Tween-20/EtOH (1 : 1 v/v)/water; (□) Orange oil/Tween-20 + EtOH (2 : 1 v/v)/water. Scale magnitude and axis representation as in (***a***). ***c***, Ternary and pseudo-ternary phase diagrams for different systems at 303 K. (✳) Peppermint oil + IPM (1 : 1 v/v)/iPrOH/water; (○) Peppermint oil/Tween-20/water; (■) Peppermint oil/Tween-20 + iPrOH (1 : 1v/v)/ water; (Δ) Peppermint oil/Tween-20+EtOH (2 : 1 v/v)/water. Scale magnitude and axis representation as in (***a***). ***d***, Pseudo-ternary phase diagrams for ternary systems at 303 K. (●) IPM/iPrOH/water; (○) Peppermint oil/iPrOH/ water. Scale magnitude and axis representation as in (***a***).

The topological behaviour of eucalyptus oil/Tween-20/water and CA/Tween-20/water systems revealed that the macro and microphase volume fractions were sensitive to temperature and the viscous phase underwent shear thinning. The enthalpy of dissolution of both water in (amphiphile + oil) and oil in (amphiphile + water) showed both endo and exothermic features for the eucalyptus oil-containing system and only endothermic change for the CA-containing system[103]. Microemulsification of eucalyptol and water was possible in the presence of polyoxyethylene (4) lauryl ether(Brij-30) and ethanol[104]. Mixing of IPM and water was possible using Brij-30 as the surfactant and isopropanol as the co-surfactant[57]. The same authors[105] also reported mixing of coconut oil and water with polyoxyethylene-2-cetyl ether (Brij-52) and ethanol or isopropanol. A fair extent of single-phase microemulsion was observed for all the systems. The preparations were thermodynamically stable but viscous and, showed shear thinning and temperature-dependent viscosity decrease. The dependence of enthalpy and entropy of activation for viscous flow on temperature was linear, but that of free energy of activation was nonlinear. The dispersions (2–64 nm in size) were fairly polydisperse. The dimensions of both O/W and W/O dispersions increased with increase in temperature, with associated increase in polydispersity. Under comparable conditions, mixed oils (rice bran and IPM) formed a larger microemulsion domain than their pure counterparts[56]. The effects of addition of isomers of butanol as co-surfactant in the microemulsification of eucalyptus oil with AOT or with polyoxylene (23) lauryl ether (Brij-35) individually or in mixed condition (AOT and Brij-35) were temperature-insensitive, and the free energy of dissolution of oil or water was a function of surfactant composition[106]. The energetics of microemulsification of different plant oils in pure (eucalyptus and IPM) and mixed conditions (rice bran oil and IPM) using Brij-92 as surfactant and EtOH and I-PrOH as co-surfactants is presented in Table 1. The diameter and diffusion coefficient of these systems at different temperatures are presented in Table 2. Biocompatible microemulsion systems for pharmaceutical application with more and more new compositions are in the process of continuous development and characterization.

Encapsulation of drugs and *in vitro* studies

Solubilization of testosterone propionate (2% w/w) in O/W microemulsion composed of large molecular volume oil caprylic/capric acid triglycerides (Miglyol 812)/Brij-96/

Table 1. Energetics of microemulsification of oil and water by calorimetry at 303 K

Composition (wt%)	Water added (10^{-3}mol)	Oil (eucalyptus) added (10^{-3} mol)	ΔH_s^0 (kJ mol^{-1})	ΔG_s^0 (kJ mol^{-1})	ΔS_s^0 (J K^{-1} mol^{-1})
(62/34/4)[a]	6.11	–	–1.09	3.89	–16.44
(43/45/12)[a]	20.0	–	–1.06	2.04	–10.23
(32/49/19)[a]	34.0	–	–0.89	1.43	–7.66
(25/50/25)[a]	48.62	–	–0.67	1.12	–5.91
(25/50/25)[a]	–	5.81	0.84	6.53	–18.78
(17/49/34)[a]	–	3.59	1.31	7.96	–21.95
(12.6/47.1/40.3)[a]	–	2.58	1.71	8.99	–24.03
(6.5/42.8/50.6)[a]	–	1.26	2.33	11.04	–28.75
Composition (wt%)	Water added (10^{-3}mol)	Oil (IPM) added (10^{-3} mol)	ΔH_s^0 (kJ mol^{-1})	ΔG_s^0 (kJ mol^{-1})	ΔS_s^0 (J K^{-1} mol^{-1})
(64/29/7)[b]	10.68	–	–0.69	2.13	–9.31
(47/35/18)[b]	31.14	–	–1.02	1.04	–6.80
(31/38/31)[b]	63.68	–	–0.70	0.62	–4.36
(20/39/41)[b]	–	2.55	7.19	9.09	–6.27
(11/37/52)[b]	–	1.28	10.1	11.10	–3.17
(4/30/66)[b]	–	0.44	8.36	14.10	–19.00
(51/43/6)[c]	8.98	–	–0.48	2.9	–11.0
(40/49/11)[c]	17.31	–	–0.46	2.0	–8.1
(30/53/17)[c]	28.58	–	–0.43	1.5	–6.3
(18/57/25)[c]	–	2.10	2.0	8.8	–22.4
(10/57/33)[c]	–	1.08	3.4	10.7	–24.0
(5/54/41)[c]	–	0.52	3.5	12.8	–30.7

[a]Eucalyptol/(Brij-30+EtOH); (1 : 1)/water[60]. [b](Rice bran oil + isopropyl myristate); (1 : 2)/Brij-92 + iPrOH (1 : 2)/water[6]. [c]Isopropyl myristate/(Brij-30+i-PrOH) (1 : 1)/water[7].

water was almost thrice that composed of small molecular volume oil, ethyl butyrate/Brij-96/water, though the solubility of the drug in Miglyol 812 was much less than in ethyl butyrate. According to phase inversion temperature studies, the structure of the microemulsion is sensitive to the type of oil. The smaller molecular volume oils (these oils penetrate the interfacial surfactant monolayer in much the same way as a co-surfactant), cause an alteration (presumably a dilution) of the relatively concentrated polyoxyethylene region close to the hydrophobic core, thereby destroying one of the main loci of drug solubilization and counteracting the advantage of high solubility of the drug in bulk oil[49,50]. Dissolution of a mixture of anesthetic drugs, Lidocaine (Lingocaine) and Prilocaine, was done using self-microemulsifying drug delivery systems (SMEDDS), where the oil phase itself was a mixture of the drugs (eutectic mixture), the other components being a blend of a high HLB and low HLB surfactant, i.e. Tween-80 (HLB = 15.0) and Poloxamer 331 (HLB = 1), and propylene glycol as the hydrophiic phase[107]. The solubility of the drugs, Progesterone and Indomethacin, was enhanced to 3300 and 500-fold respectively, in IPM-containing microemulsion[108]. Incorporation of Ibuprofen, Ketoprofane,Tamoxifen,Testosterone and Tolbutamide in O/W microemulsion of medium chain fatty acid triglyceride (MCT)/diglyceryl monooleate (DGMO-C) + polyethylene hydrogenated castor oil 40 (HCO-40)/ethanol/PBS (pH 6.8) increased its solubility 60–20,000 times[109]. The partcle size was 20 nm or less.

Artificial Neural Network (ANN) methodology (a computer simulation program) was used to develop colloidal dosage forms of two antitumour drugs Rifampicin (RIF) and Isoniazid (INH), with aqueous solubility of 1.82 and 128.8 mg ml^{-1} respectively from pseudoternary systems of Miglycol 812, polyoxyl 40 hydrogenated castor oil (cremophore RH 40), glyceryl monostearate, glycerol mono- and dicaprylate/caprate, respectively having (Imwitor 308 and Imwitor 7429) as oil phase, Brij-97 and polyoxyethylene (20) sorbitan monostearate as surfactant, and sorbitol as co-surfactant, in various combinations taking into consideration the widely different solubilities of the drugs. The formulation was predicted to be capable of delivery of the drugs at the desired target concentrations (150 mg ml^{-1} for RIF and 100 mg, 15 ml^{-1} for INH) for treatment of children with tuberculosis. The formulation provided an answer to the problem of combining two drugs with different solubilities[110]. The release of Nortryptilene hydrochloride from O/W microemulsion (IPM/Tween-80/propylene glycol + phosphate buffer (pH 7.4)) was enhanced six times in the presence of increasing amounts of PEG 400 at concentrations of 50% (v/v). PEG 400 was used to facilitate the diffusion of the drug from the inner oil phase of the microemulsion to the outer aqueous phase[81]. The *in vitro* release rates of the drug Indomethacin from microemulsions containing IPM, lecithin, lysolecithin and alcohol depended on the size of the dispersed phase and the amount of co-surfactant used[111]. The *in vitro* release of a nonsteroid anti-inflammatory

Table 2. Dynamic light scattering results on the plant oil-based microemulsion systems

Composition (wt%)	Temperature (K)	Diameter (d) (nm)	Polydispersity index (PDI)	Diffusion coefficient (D) × 10^8 cm^2 s^{-1}
(45/45/10)[a]	293	8.0	0.134	5.42
	298	30.2	0.034	1.49
	308	34.6	0.980	1.58
(31/50/19)[a]	293	3.6	0.732	9.23
	298	13.4	0.823	3.70
	308	22.5	0.848	3.32
(18/50/32)[a]	298	203	0.350	1.64
	308	64.5	0.398	0.65
(9/46/45)[a]	293	13.1	1.052	178
	298	10.8	0.772	2.69
	308	7.2	0.575	5.30
(3/39/58)[a]	298	0.8	0.565	0.30
	308	1.2	0.429	0.26
(40/51/9)[b]	293	1.3	0.50	3.60
	303	0.9	0.53	4.18
	313	27.0	1.06	0.21
	323	105	3.51	0.06
(28.5/57.5/14)[b]	293	4.2	0.63	0.91
	303	3.7	0.54	1.81
	313	4.7	0.51	1.66
	323	6.9	0.50	1.29
(18/60/22)[b]	293	2.7	0.61	1.17
	303	2.8	0.52	1.63
	313	2.8	0.54	1.98
	323	2.9	0.48	2.27
(9/59/32)[b]	293	2.2	0.56	1.45
	303	3.0	0.50	1.48
	313	4.9	0.43	1.43
	323	7.6	0.39	1.42
(3/55/42)[b]	293	2.6	0.59	1.52
	303	3.7	0.45	1.57
	313	4.5	0.42	1.64
	323	7.6	0.21	1.26
(63/32/5)[c]	298	32.0	0.917	1.95
	308	34.4	0.904	1.98
(43/38/19)[c]	293	6.2	0.536	4.18
	298	5.5	0.527	5.48
	308	4.7	0.496	7.98
(28/41/31)[c]	298	1.7	0.6	17.85
	308	1.9	0.569	19.6
(12/41/47)[c]	293	17.9	0.499	1.27
	298	8.9	0.687	3.68
	308	2.0	0.641	38.87
(3/33/64)[c]	298	1.3	0.519	21.75
	308	3.1	0.318	13.8

[a]Eucalyptol/(Brij-30 + EtOH), (1 : 1)/water[60]. [b](Rice bran oil + isopropyl myristate), (1 : 2)/(Brij-92 + iPrOH) (1 : 2)/water[6]. [c]Isopropyl myristate/(Brij-30/i-PrOHO) (1 : 1)/water[7].

drug, Diclofenac diethylamine (DAA) from caprylo capryl macroglyceride-based microemulsion vehicles, IPM/PEG-8 caprylic/capric glycerides (labrasol) + polyglyceryl-6 dioleate/water was influenced by both formulation parameters and vehicle structure[112,113]. The skin permeation flux of Felodipine (a highly vascular, selective calcium antagonist) could be enhanced 10–50 times from O/W microemulsion containing benzyl alcohol or different

ratios of benzyl alcohol + IPM as oil phase, than from an aqueous suspension[114]. *In vitro* studies with Franz-type diffusion cells for model drugs, Lidocaine and Prilocaine hydrochloride showed improved transdermal flux of Lidocaine up to four times and that of Prilocaine hydrochloride almost ten times from microemulsion labrasol/isostearique/isostearyl stearate/water, than O/W emulsion and a hydrogel, respectively. Improved skin bioavailability of Lidocaine was observed from *in vitro* precutaneous penetration studies of the drug from quarternary microemulsion system of IPP(oil)/glyceryl oleate + polyoxyl-40 fatty acid derivative(surfactant)/tetraglycol(co-surfactant)/water. Water content and the co-surfactant/surfactant ratio goverened the transdermal penetration. Significant improvement in analgesic response was observed when compared with conventional cream formulation[115].

Use of penetration enhancers was reported by different groups. Release of accelofenac (calcium channel blocker) incorporated into the O/W microemulsion systems (10–100 nm) of labrafil M 1944/cremophore ELP + ethanol/water (determined by Franz-type diffusion cell mounted on rat skin) increased fivefold than from ethanol. Among the terpenes tested as permeation enhancers, limolene had the best effect (threefold increase over control)[116]. Dermal permeation efficacy of a highly hydrophilic model drug Diphenyl hydramine (DPH) from a microemulsion of IPP (74%)/Tween-80 and Span-20 mixture (2 : 3) (20%)/water, improved with addition of a glycolipid[117]. Addition of transdermal enhancers *n*-methyl pyrrolidone and oleyl alcohol to O/W microemulsion systems of IPM/(Tween-80)/water enhanced drug permeabilities;17-fold for Lidocaine free base, 30-fold for Lidocaine HCl, 58-fold for Estradiol, and 520-fold for Diltiazem HCl[118]. The microemulsion system was capable of simultaneous delivery of hydrophobic (Lidocaine free base + Estradiol) and hydrophilic drugs (Lidocaine HCl + Diltiazem). Skin irritation caused by an aqueous solution of Triptolide (having immunosuppressive, anti-fertility and anti cancer activities) in 20% propylene glycol (containing 0.025% triptolide) could be avoided through encapsulation of the drug in oleic acid/Tween-20/propylene glycol/water microemulsion. Use of methanol enhanced *in vitro* skin permeation[119]. The precutaneous absorption of DDA from microemulsions (DDA (16%)/lauryl alcohol (5%)/Labrasol + ethanol (2 : 1) (34.54%)/water (60%)) showed marked improvement with increase in the lauryl alcohol and water contents and decrease in labrasol : ethanol mixing ratio[120]. Addition of terpenes (5%) enhanced the skin permeability of the drug. Oleic acid showed both excellent solubility and skin permeation enhancing effect for piroxicam in an optimum formulation of the drug (0.5%) in oleic acid (10%)/labrasol/ethanol (1 : 5) (60%) and water[121]. The clinical use of Celecoxib, a specific Cox-2 inhibitor is restricted because of its failure to block the characteristic cutaneous inflammatory response and lower availability at the site of inflammation when administered orally. Topical application of Celecoxib has been effective compared to oral administration in certain clinical conditions. But, the *in vitro* permeation rate of Celecoxib through rat skin was up to 5 and 11 times more from IPM + medium chain mono-/diglyceride/Tween-80/water than from microemulsion gel and cream[122]. The drug solubility of Vinpocetin[123] was about 3160-fold higher in 1% Vinpocetin in oleic acid (4%)/labrasol (20.5%) + diethyl glycol monoethyl ether (TranscutolP) (20.5%)/water (55%), than in water and the apparent permeation rate was 36.4 ± 21 G cm^{-2} h^{-1}. Shelf-life of the microemulsion was one year at 25°C. Estradiol is widely used for the treatment of hormonal insufficiencies, but oral delivery limits its bioavailability due to its hepatic first-pass metabolism. Encapsulation of the drug in various microemulsions leads to 200–700-fold improvement in transdermal flux over the control (due to 1500-fold improvement in solubilization)[124]. *In vitro* studies showed that transdermal permeation of the antineoplastic agent 5-flurouracil from W/O microemulsion of IPM/AOT/water increased in the skin flux depending on the composition of the microemulsion[125]. The transdermal drug delivery of the immunosuppressive drug Cyclosporin A (CysA) increased up to ten times after encapsulation in O/W microemulsion (IPM/Tween-20/water) compared to the suspension in normal saline containing 20% ethanol, which was dependent on water concentration and not on the microstructure of the microemulsion[126]. The therapeutic advantage of dermal administration of Cys A in rat model was determined. Local (subcutaneous and skin) systemic concentration and organ distribution (liver and kidney) were evaluated serially following topical and oral applications of the drug. Deposition of the drug into the skin and subcutaneous fat respectively, was almost 30 and 15-fold higher compared with oral administration. Systemic distribution in blood, liver and kidney was much lower following topical administration than oral administration. An eight-fold improvement in analgesic response of Tetracaine hydrochloride was possible through its encapsulation in IPM/AOT/water, compared to the aqueous solution of the drug. The analgesic response depended considerably on AOT and water concentrations, and the microemulsion system proved to be a good transdermal carrier of the drug with a concentration of AOT in IPM up to 21.79 w/w (ref. 49).

Encapsulation of drugs and *in vivo* studies through different routes

Subcutaneous route

Colloidal drug-delivery systems are taken up non-specifically by the reticuloendothelial systems[42]. So the drugs to be targeted to the macrophages of the RES are

expected to yield good results when they are encapsulated in a microemulsion. Experimental Leishmaniasis was used as a model disease where the amstegotes of the causative parasite, *Leishmania donovani* resides and proliferates within the macrophages of the RES. Thus, antileishmanial drugs after their incorporation into appropriate delivery systems, could be targeted to the RES and examined for their efficacy. *In vivo* studies by Gupta *et al.*[42] using subcutaneous routes of delivery of the natural product (quercetine with anti-leishmanial property) solubilized and encapsulated in clove oil/Tween-20/water (5/30/65 wt%) showed significant improvement in the efficacy of the drug (compared to maceration in DMSO) in hamstar models. Both *in vitro* and *in vivo* studies of another natural product, Bassic acid[44] (unsaturated triterpene acid with antileishmanial properties), using the same microemulsion system as the vehicle also showed encouraging results. The efficacy in free form and after encapsulation in two delivery systems (microemulsions and poly-lactide nanoparticles) was compared.

Oral route

The solubility of Ibuprofen–euginol ester (synthesized from an anti-inflammatory drug, Ibuprofen) in the optimized pharmaceutical microemulsion formulation of Ibuprofen euginol (6.4%)/Miglyol 812 (96%) + SbPC (6%) + polyethylene glycol (660)-hydroxystearate (Solutol-HS-15) (6%)/PEG400 (8.4%) + ethanol (36%)/water (60%) was 21,000 times more than in water. The area under the curve (AUC) of Ibuprofen from the prodrug showed a remarkable increase compared to the oral Ibuprofen suspension[127,128].

A premicroemulsion concentrate of oil, surfactant and co-surfactant was prepared for oral administration of Biphenyl dimethyl carboxylate (BDC), a drug for treating liver diseases. Among the non-ionic surfactants and the oil studied, Tween-80 (which led to the highest solubility of BDD, 109.7 g ml^{-1}) and Neobee M5-R were chosen for preparing a premicroemulsion concentrate. At the ratio of 2 : 1 of Tween-80 and Neobee M5-R, the solubility of BDD increased seven-fold compared to that at the ratio of 1 : 4. The solubility of BDC was further improved with the addition of 35% triacetin used as co-surfactant. BDC in the premicroemulsion concentrate rapidly dissolved in distilled water and artificial gastric fluid (without pepsin, pH 12) whereas a 92 : 1 mixture of BDC powder and calcium carboxymethyl cellulose (Ca–CMC) hardly dissolved during the 120 min incubation period. After oral administration of premicroemulsion concentrate, AUV (0–24 h) and mean maximum plasma level (C_{max}) of BDC were fivefold and tenfold higher respectively, than those of BDC with another formulation, Ca–CMC. Thus, the premicroemulsion concentrate of BDC greatly enhanced its bioavailability after administration due to the increase in solubility and immediate dispersion of the drug in the gastrointestinal tract[129].

A microemulsion system of Capryol 90/Cremophor EL/Transcutol (30 nm in mean diameter) was prepared with improved solubilization capacity (up to 30 mg ml^{-1}, even after 20 times dilution with normal saline) and improved oral bioavailability of the hydrophobic drug, Docetaxel (Dx). Three different formulations did not show significant differences in the *in vitro* lipid digestion study. Ultrafiltration and dialysis studies revealed the release of 80% of Dx from the microemulsions within 12 h. The oral bioavailability of the formulation (Capryol 90/Cremophor EL/Transcutol at 29.4 : 24.9 : 12.4 w/w) in rats rose dramatically (34.42%) compared to that of the orally administered Taxotere® (6.63%). This increase in bioavailability was probably due to the combined effect of the enhancement in solubility and the increase in permeability[130]. An oral microemulsion formulation of the anticancer herbal drug Berberine (Bb) was encapsulated in oleic acid, Tween-80 and PEG400 microemulsion, and its *in vivo* pharmacokinetic profile and oral bioavailability were studied in rats. The optimized formulation was 15 wt% oleic acid, 17 wt% Tween-80, 17 wt% PEG400, and 51 wt% water. The formulated microemulsion was found to be relatively uniform in size (24 nm). The bioavailability of the oral Bb-loaded microemulsion formulation was 6.47 times greater than that of the Bb tablet suspensions[131].

Intravenous route

The drug Propofol itself was chosen as the oil phase (1% w/w) in an O/W microemulsion for parenteral delivery using the surfactant : co-surfactant mixture of 12-hydroxystearate (Solutol HS-15) and ethylalcohol (5/1). The optimum composition chosen was 1% (w/w) of propofol solubilized with 8% (w/w) of Solutol Hs-15 + ethyl alcohol (5/1) water. The average droplet size was 150 nm and the microemulsion system was stable for 8 weeks at 40°C. The hemolysis test showed that the formulation was non-toxic to red blood cells[132]. Zhang *et al.*[133] encapsulated the hepatogenic drug Norcantheridine (NCTD) in W/O microemulsion of water (7%)/soy lecithin and ethanol (2 : 1) (45%)/ethyl oleate (48%) (droplet size, 44 ± 8.6 nm). The drug-loaded microemulsion had relatively longer circulation time in mice than the injection formulation after a single intravenous (iv) application at a dose of 5 mg kg^{-1}; the overall drug targeting efficiency to the liver enhanced from 3.66% to 610%.

The solubilization capacity of the microemulsion system MCT/(SbPC + poly(ethylene glycol)(660)-12-hydroxystearate ($12HAS\text{-}EO_{15}$) + PEG-400 + EtOH)/water for Felodipine and H290/58 (an antioxidant) was ~ 10 times higher than that of PEG 400 vehicle and soyabean emulsion. This micoemulsion can be administered by iv infu-

sion to conscious rats up to 0.5 mg kg^{-1} without producing any significant effect on acid–base balance, blood gases, plasma electrolytes, heat rate (HR) and PQ times (the time between depolarization of atrium and chamber) and up to 1.5 mg kg^{-1} with only a minor and temporary increase in mean arterial blood pressure (MAP), bradycardia, and prolongation of the PQ time and no changes in the behaviour of the animals[53]. The low solubility of Amphotericin B (AmB, a membrane-active polyene antibiotic with strong antifungal activity and the drug of choice for AIDS, organ transplants, cancer chemotherapy) leads to poor bioavailability by the oral route and is thus parenterally administered as a solubilizate in sodium deoxycholate (Fungizone^R; Bristol Mayer Squibb). Results of *in vivo*, single dose acute toxicity studies by iv administration of the drug encapsulated in IPM/(SbPC + Tween-80)/water microemulsion in male Albino mice showed low toxicity and improved pharmacokinetic behaviour and enhanced solubilizing capacity than Fungizone^R. The LD_{50} for AmB microemulsion was of 4 mg kg^{-1} compared to 1 mg kg^{-1} for the commercial sodium deoxycholate suspension. For similar formulation with IPM/(SbPC + Brij-92)/water, LD_{50} for AmB formulation was 2.9 mg kg^{-1}. *In vitro* studies (partitioning and spectral studies) showed that the drug was intercalated at the oil–water interface as a complex, formed with the lecithin molecules[79,80]. Flurbiprofen was incorporated in ethyl oleate/Tween-20/water for parenteral administration[134]. The mean droplet diameter of the microemulsion containing less than 1% (w/w) of Flurbiprofen was below 100 nm. The pharmacokinetic parameters of Flurbiprofen after iv administration of drug-loaded microemulsion in rats were not significantly different from those of Flurbiprofen in PBS. Solubility of Flurbiprofen increased up to 10 mg ml^{-1} (eightfold higher compared with flurbiprofen solubility in PBS solution). Similar enhancement of the solubility of the drug was observed in ethyloleate/lecithin + distearolyl-phosphatidyl-ethanolamine-*N*-poly (ethylene glycol) 200 (DSPE-PEG) + ethanol/water microemulsion.

Nasal route

The nasal absorption of Diazepam from ethyl laurate (15%)/(Tween-80 : propylene glycol : ethanol at 1 : 1 : 1 weight ratio (70%)/water (15%) microemulsion was fairly rapid at 2 mg kg^{-1} dose. The maximum drug plasma concentration was reached within 2–3 min and the bioavailability (0–2 h) after nasal spray compared to iv injection was about 50%. The formulation might be useful for the rapid onset delivery of Diazepam during the emergency treatment of status epilepticus. The drug displayed a high solubility of 41 mg ml^{-1} in the microemulsion[135].

Microemulsion containing another epileptic drug clonazepam was found to be effective from animal studies for rapid delivery to the brain to treat acute status epileptic patients[136]. Efficient delivery of anti-migraine drugs, Sumatriptan and Sumatriptan succinate to the brain in acute attack of migraine through intranasal and iv injection was possible through encapsulation in mucoadhesive microemulsion. Pharmacokinetic parameters, drug targeting efficiency and a direct drug transport showed rapid, effective and a larger extent of drug availability through the intranasal mode[137]. Three microemulsion systems stabilized by non-ionic surfactants (Cremophor RH 40 or Labrasol), containing a variety of oils (IPM, Labrafil M 944C, etc.) developed by Zhang *et al.*[138], were suitable for nasal delivery. Enhancement of solubility and brain uptake of Nimodipine (NM; for neurodegenerative diseases) was observed by *in vivo* studies from microemulsions. Maximum solubility of NM up to 6.4 mg ml^{-1} was possible in a chosen composition of 30% Chremophore RH40/ethanol (3 : 10) and water, and the size of the dispersed droplets was 30 ± 5.3 nm. After a single intranasal administration, the plasma concentration peaked at 1 h and the absolute bioavailability was about 32%. The uptake of the drug in the olafactory bulb from the nasal route was threefold compared with iv injection. The ratio of AUC in brain tissues and cerebrospinal fluid to that in plasma obtained after intranasal administration was significantly higher than that after iv administration.

Ophthalmic route

The microemulsion-based Dexamethasone eye drop developed by Fialho *et al.*[139] is well-tolerated by the eye with higher degree of bioavailability. The developed system showed greater penetration in allowing the possibility of decreasing the frequency of application of the eye drop. O/W and O/W microemulsion systems with Retinil and its esters as oil component, Tween-80, Tween-60, SbPC as surfactants and *n*-butanol, triacetin, polypropylene glycol as co-surfactants were studied for ocular delivery and the drug containing optimum composition proved suitable with regard to their ophthalmic parameters (refractive index, viscosity, pH and osmotic tension). No physical change was observed for six months at 20°C and preparation was physiologically tolerated[140]. Chloramphenicol (used for the treatment of trachoma and keratitis) in the common eye drops hydrolyses easily into glycols. Encapsulation of the drug in O/W microemulsion of IPM/(Span-20/80 + Tween-20/80 + *n*-butanol)/water remarkably improved the stability of the drug. NMR experiments confirmed that the drug molecules remained trapped into the hydrophilic shells of the microemulsion drops. The drug molecules were thus screened and protected from the bulk water[141]. Similar observations were made for microemulsions free of alcohols[142]. An elaborate review on the related topics may be found in the literature[2,8,10].

General conditions and limitations of microemulsion systems

A large number of microemulsion systems with pharmaceutical uses for drug encapsulation and delivery have been developed and characterized by physico-chemical methods. But each system has not been substantiated by biological studies, a pre-requisite for pharmaceutical use. There are specific criteria and limitations for each route of delivery. For example, for oral route the delivery system needs to have stability over a wide pH range. For intravenous application, it should be less viscous so as to not cause pain on injection, and it should not be hemolytic. For topical applications, the formulations need to be viscous so as to spread evenly on the surface and should be nonirritant to the skin. For transdermal application the requirement is good penetration through the skin layers; for ocular delivery the formulation needs to be isotonic with tear fluid, etc. The delivery systems are meant to reduce the toxic side effects of the drugs. So each of the delivery systems should be examined for *in vivo* toxicity and biocompatibility.

It is observed that the concentration of surfactants in microemulsions is quite significant, and the high surfactant concentration poses the main obstacle for the fruitful use of the microemulsions for pharmaceutical purposes. Efforts are needed to develop microemulsions with lower surfactant content, and also to synthesize biocompatible surfactants.

The most studied pharmaceutical microemulsion systems for drug delivery have been through the topical route. Development of microemulsion systems useful for the other routes is required.

Conclusions

The biocompatible microemulsion system is a relatively new 'drug delivery system' for pharmaceutical application. A significant number of microemulsion systems is reported in the literature, but the systems are of widely divergent composition. This poses a hurdle to data correlation and meaningful clinical uses.

A concerted effort is required to categorize the compositions with reference to more generalized patterns based on chemical compositions of the excepients. In this respect, the scope of research in the field remains fairly open.

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