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RESEARCH PAPER

Understanding Biorelevant Drug Release from a Novel Thermoplastic Capsule by Considering Microstructural Formulation Changes During Hydration

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ABSTRACT

Purpose To study the biorelevant drug release from novel starch-based polyvinyl alcohol capsules (S-PVA-C). The effect of the shell material is studied by considering microstructural formulation changes during hydration.

Methods Two different self-emulsifying systems containing either fenofibrate or probucol were filled in S-PVA-C, as well as capsules of gelatin (SGC) and starch (VegaGels®). Release analysis employed a BioDis® apparatus, while disintegration was studied by texture analysis. For microstructural analysis we used small angle x-ray scattering (SAXS).

Results S-PVA-C opened only partially in biorelevant media compared to completely opened SGC and VegaGels®. In case of the fenofibrate formulation, this opening mechanism caused only a short lag time, while the probucol formulation in S-PVA-C resulted in a sustained release. The latter formulation demonstrated much higher viscosity upon hydration compared to the fenofibrate system. Such a rheological effect on drug release was barely noted for SGC or VegaGels® and SAXS revealed differences in the hydrated microstructure.

Conclusions Even though S-PVA-C are highly attractive for encapsulation of rather hydrophilic formulations, some care is needed regarding an immediate release form. The type of formulation hydration must be considered for adequate selection of the capsule material.

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KEY WORDS biorelevant release · controlled hydration · disintegration · lipid-based formulation · thermoplastic capsule

INTRODUCTION

There has been a great interest in finding gelatin substitutes for soft capsules in the last decade. This is due to known gelatin drawbacks, for example animal sources, potential issues of cross-linking, drug migration, or high water exchange during drying between a hydrophilic fill mass and the gelatin shell (1). Recently, we introduced (thermoplastic) starch-based polyvinyl alcohol capsules (S-PVA-C) in pharmaceutics (2). This soft capsule technology appeared to be especially promising for encapsulation of rather hydrophilic lipid-based systems. A typical problem of many self-microemulsifying or nano-emulsifying drug delivery systems is that the presence of hydrophilic formulation components can lead to issues of capsule compatibility when gelatin is used as shell forming material. The problem might be alleviated by addition of excipients to the soft gelatin shell, but a more versatile alternative is to employ the novel S-PVA-C technology. This thermoplastic capsule material was found to be suitable for rotary die encapsulation and the previous study demonstrated adequate drug release in a quality-control (QC) drug release test (2).

Compendial drug release methods are used either in QC testing or to predict *in vivo* drug release with regards to the absorption process. Depending on the focus, the appropriate dissolution media and conditions need to be selected. For example, simplified aqueous media are typically used for routine QC purposes and more complex media might be required for predicting *in vivo* absorption (3). Such more complex media are especially needed for lipid-based formulations. The presence of bile salts and phospholipids, as natural surfactants, has a special importance in these drug release media. Due to their amphiphilic nature, there can be an effect on drug solubilization as well as on reduction of interfacial oil tension, which

would affect formulation dispersion. To better simulate the environment of gastro-intestinal (GI) fluids, many biorelevant dissolution media have been developed and tested over the last decade (4). Moreover, standard dissolution methodology (paddles or baskets) may not adequately mimic the hydrodynamics occurring in the GI tract, which was one of the reasons to develop the reciprocal cylinder, i.e. USP apparatus 3 (BioDis®). Although it is the most attractive method for testing extended-release products, some immediate release products were also successfully analyzed (5). It seems to be attractive for *in vivo* absorption prediction to combine the use of a USP apparatus 3 with selecting biorelevant media. This is especially the case for poorly soluble, lipophilic drugs as demonstrated by Jantratid *et al.* (6,7). One could argue that the most physiological for lipid-based systems is to test *in vitro* lipolysis (8,9). Such tests are advantageous with formulations that are extensively digested. However, there is currently no standardized vessel defined for lipolysis testing so that hydrodynamics may differ from one equipment to another. Another drawback compared to compendial testing is that the final dosage form cannot be assessed with current lipolysis assays. Since we focused rather on the effect of the shell material on drug release, we selected biorelevant media in a USP apparatus 3.

In the last decade self-emulsifying lipid-based systems have attracted increasing interest among scientists. These systems are dosed as pre-concentrates in a capsule and upon administration they disperse to microemulsions with a large surface area. The process of self-emulsification is a complex and still debated topic. Excellent reviews on the mechanisms of spontaneous emulsification were written by Lopez-Montilla *et al.* and others (10–13). Three principal mechanisms have been proposed for spontaneous emulsification. A first mechanism is based on interfacial turbulence, while another is called "diffusion and stranding" and finally, the transient occurrence of a negative interfacial tension has been discussed. It is also possible that combinations of these principal mechanisms dominate the aqueous dispersion of a self-emulsifying formulation.

The aqueous dilution of a pre-concentrate can possibly exhibit a phase separation along the dilution line. Interesting is the work of Regev *et al.* (14) who demonstrated that even in case of a resulting one-phase system, the aqueous dilution of the pre-concentrate typically passes through liquid crystalline structures and/or a bicontinuous microemulsion area. Only recently, small angle x-ray scattering (SAXS) was used in pharmaceutics to study the intermediate hydration phases of self-emulsifying drug delivery systems (15–18).

In the present work, we addressed the need to study the biorelevant drug release from S-PVA-C. Self-emulsifying systems were used as model formulations and results were compared to the release from soft capsules made of gelatin and of a thermoplastic starch (VegaGels®). A particular aim was to evaluate the interaction of the shell material and the type of formulation hydration by studying microstructural changes.

MATERIALS AND METHODS

Materials

Fenofibrate (Sigma-Aldrich Chemie GmbH, Buchs, Switzerland) and probucol (Euroasian Chemicals Pvt Ltd, Mumbai, India) were selected as poorly water-soluble model drugs. Medium-chain triglycerides (Miglyol® 812) were supplied by Hänseler AG (Herisau, Switzerland), soybean oil refined from Georges Walther AG (Pfäffikon, Switzerland) and Transcutol HP (2-(2-ethoxyethoxy)ethanol) by Gattefosse (Lyon, France). Imwitor® 742 (medium chain partial glyceride) was obtained from SASOL (Witten, Germany) and macrogol-glycerolhydroxystearat (Cremophor® RH 40) from BASF (Ludwigshafen, Germany). Polyoxyethylene (80) sorbitan monooleate (Tween® 80) and acetonitrile of high-pressure liquid chromatography (HPLC) grade were purchased from Sigma-Aldrich Chemie GmbH (Buchs, Switzerland).

For the preparation of the fasted state simulated gastric fluid (FaSSGF) and the fasted state simulated intestinal fluid (FaSSIF), we purchased sodium chloride and sodium hydroxide from Sigma-Aldrich Chemie GmbH (Buchs, Switzerland), pepsin and maleic acid from Hänseler AG (Herisau, Switzerland), sodium taurocholate from Prodotti chimici e alimentari S.p.A. (Basaluzzo, Italy), phosphatidylcholine from Lipoid GmbH (Ludwigshafen, Germany), and hydrochloric acid (1 N) and sodium hydroxide (1 N) from Scharlab (Barcelona, Spain).

The air-filled and oil-filled capsules (Table I), used for the release testing, and the disintegration testing, were produced at the technical facility at Swiss Caps AG, (Kirchberg, Switzerland).

Methods

Preparation and Hydration of the Formulations

Two different self-emulsifying systems consisting of Tween® 80, Transcutol HP, Miglyol® 812, Imwitor® 742 (45%: 15%: 20%: 20% w/w; TTMI) and Cremophor® RH 40, Transcutol HP, Miglyol® 812 (60%: 25%: 15%w/w; CrMTrans) were used as model formulations. Since lipid-based systems are generally tailor-made for each drug substance, we also employed two different formulations for the active substances, i.e. TTMI for fenofibrate and CrMTrans for probucol. Both mixtures (TTMI and CrMTrans) were compounded in glass vials (by weight) using an analytical balance (Mettler Toledo AB204-S). A magnetic stirring was employed at 40°C until a clear solution was obtained. We carefully checked for the absence of residual particles following the cooling at room temperature. The prepared formulations were hydrated with deionized water at water levels in the range of 5–90% w/w and stored for 24 h at room temperature (RT) before analysis.

Table I Characteristics of Different Capsule Types

Type of capsule	Capsule shell material	Method of production	Fill mass[a]
Soft gelatin capsules	Bovine gelatin, glycerol, and water 47.4%: 17.1%: 35.5% (w/w)	Casting and rotary die method[b]	Soybean oil
VegaGels®	Potato starch, glycerol, sorbitol, carrageenan, and water 66%: 18%: 10% : 1% : 5% (w/w)	Extrusion and rotary die method[c]	Soybean oil
S-PVA-C[d]	Potato starch, PVA, plasticizers (sorbitol solution and glycerol) 40% : 30% : 30% (w/w)	Extrusion and rotary die method[b]	Miglyol® 812

[a] Fill mass for those capsules used for the texture analysis of dosage form disintegration

[b] Methods of production are described in Misic *et al.* (2)

[c] Method of production of VegaGels® is the same as for S-PVA-C

[d] S-PVA-C: starch-based polyvinyl alcohol thermoplastic capsules

Preparation of Drug-Containing Formulations

Solubility Studies. For the system of fenofibrate in TTMI as well as probucol in CrMTrans, an excess amount of drug was added. The mixtures were constantly stirred (750 rpm on a magnetic stirrer) in glass vials at RT. To ensure that equilibrium was reached, the solubility was determined after 18 h, 24 h, and 72 h. The samples were then centrifuged at 13,362 g for 20 min using a Centrifuge 5415 R (Eppendorf AG, Hamburg, Germany). Finally, the concentrations of the active compounds in the supernatant were determined after diluting the supernatant with acetonitrile (1:500 v/v) by HPLC analyses.

Drug-containing formulations were prepared by adding fenofibrate to a clear solution of TTMI (FF-TTMI), and probucol to CrMTrans (Pro-CrMTrans), both in drug concentration of 60mg/mL.

Characterization

Particle Size Measurements. Particle size of the diluted self-emulsifying systems was analyzed by means of dynamic laser light scattering using a Zetasizer Nano ZS (Malvern, Worcestershire, United Kingdom). For individual measurements, an aliquot (100 μL) of each preconcentrate was obtained from a calibrated automatic pipette Eppendorf Research® plus (20–200 μL) and dispersed in 20 mL of water using a glass vial. Both formulations were assessed for their ease of self-emulsification and qualitatively described as "good", "moderate" or "poor" (15). The diluted samples were measured in dynamic light backscattering (173° angle) and the mean particle size (Z-average diameter) was calculated from the volume size distribution. All experiments were repeated in triplicates from fresh samples at RT.

Rheological Studies. The rheological properties of low-viscous lipid-containing colloids can be difficult to measure. For adequate measurements using a cone-plate rheometer, there must be a uniform force transmission in the sample. A loss of grip between the cone and the sample shear plane is a problem that especially occurs at comparatively high shear forces. For the measurements of the hydrated formulations, we therefore employed a new mechanical chip-based (MEMS) capillary rheometer (mVROC™ RheoSence, San Ramon, CA, USA). m-VROC™ (Viscometer-Rheometer-on-Chip) is a microfluidic slit rheometer used for fast and accurate measurements of the viscosity of microliter sample volume solutions. It measures the viscosity from the pressure drop of a sample as it flows through a rectangular slit. The glass syringe (Hamilton 1010 C SYR 10 mL or Hamilton 81260 SYR 500 μL, depending on the viscosity of the sample) was loaded with sample and placed inside of the thermal jacket (25 ± 0.5°C). When the measurement temperature was stable, the sample was pumped to flow (at shear rate 200 s^{-1}) through the flow channel of the chip. The pressure drop was detected by a sensor (cell m-VROC A-10 or D-10) and the viscosity was calculated using m-VROC Control Software™. The viscosities of TTMI and CrMTrans were measured for hydration levels of 5–90% w/w. Results were based on experiments in triplicates.

Small Angle X-ray Scattering (SAXS) Studies. SAXS was measured using a Bruker NANOSTAR setup with an Incoatec IμS-microfocus Cu-K_α anode (wavelength 1.54 Å) and a Bruker VANTEC-2000 detector. The samples were measured in flame-sealed glass capillaries with a diameter of 1.5 mm and a wall thickness of 1 μm. The 2D detector images were taken at ambient temperatures and were azimuthally averaged to produce 1D intensity profiles.

Fitting. The SAXS data were fitted using a Teubner-Strey model for microemulsions (17,19). The Teubner-Strey model is based on the Landau theory for microemulsions, where the order parameter is associated with the "water-to-oil ratio" (19). This model predicts a single broad maximum and a q^{-4}-dependance towards higher q-values (higher scattering angles).

Within this model the scattering intensity has the following form:

$$I(q) \propto \frac{1}{a + c_1 q^2 + c_2 q^4} \tag{1}$$

The obtained parameters a, c_1 and c_2 are used to calculate the two characteristic length scales d and ξ.

$$d = 2\pi \left[\frac{1}{2} \left(\frac{a}{c_2} \right)^{\frac{1}{2}} - \frac{1}{4} \frac{c_1}{c_2} \right]^{-\frac{1}{2}} \quad (2)$$

$$\xi = \left[\frac{1}{2} \left(\frac{a}{c_2} \right)^{\frac{1}{2}} + \frac{1}{4} \frac{c_1}{c_2} \right]^{-\frac{1}{2}} \quad (3)$$

Here, d is the domain size (or periodicity) whereas ξ is the correlation length, both together providing an indication of the order in the system. The ratio ξ/d is a measure of the correlation length in units of the periodicity.

Biorelevant Drug Release Studies. Drug release was studied using the BioDis® apparatus (RRT 8, CALEVA Ltd, Dorset, England) at 37±0.5°C using 220 mL of test medium in each vessel, mesh sizes of 420 μm for both the top and bottom mesh of the glass cylinders and a dip rate of 10 dpm or 30 dpm, respectively (20,21). Prior to each experiment, the air-filled capsules were weighed before and after manual filling with FF-TTMI or Pro-CrMTrans, to obtain an exact fill mass in each capsule. The experimental set-up in this study was supposed to mimic the fasted gastrointestinal tract (GIT). Therefore we used FaSSGF (30 min) followed by FaSSIF (60 min), prepared as described by Jantratid *et al.* (22). At predetermined time points, a 2 mL sample was taken by a syringe, immediately filtered through a 0.45 μm Nylon filter (Titan 2 Syringe Filter, SUN SRi, Rockwood, TN, USA) and analyzed by HPLC. The removed volume was replaced each time with 2 mL of fresh medium. All experiments were performed in triplicates.

Texture Analysis of Dosage Form Disintegration. A texture analyzer (TAXT2i; Stable Micro Systems, Surrey, United Kingdom) was assembled with a disintegration rig (originally developed for fast-melting tablets) to study the disintegration of capsules. This mechanical test mimics the gastric disintegration conditions, while constantly maintaining the force and measuring the distance as the sample disintegrates. The apparatus was equipped with a 5 kg load cell and fitted with a 20 mm diameter cylindrical probe. The capsule was attached with a strip of 3 mm wide double-sided tape to the underside flat region of the probe end. Each capsule type was filled with oil at the technical facility of Swiss Caps AG, (Kirchberg, Switzerland) (Table I) and was analyzed in triplicates. A double-jacketed glass vessel was connected with tubes to a thermostat system to keep the disintegration medium (FaSSGF, 100 mL) at 37±0.5°C. On the bottom of the double-jacketed glass vessel was a (30 mm diameter) platform, which was perforated to allow ingress of water beneath the capsule. The speed of the probe with an attached capsule was initially 2.0 mm/s until the surface of perforated platform was detected at the force of 0.029 N (threshold value for triggering the onset of texture analysis). Subsequently, the force of the probe was 0.098 N with a speed of 3.0 mm/s and the distance was measured to obtain a capsule disintegration profile. The time of analysis was 30 min (consistent with *2.2.3.4. Biorelevant drug release studies*). All capsules were produced either on pilot- (S-PVA-C) or on a production-scale (SGC, VegaGels®) using rotary die filling machines at the Swiss Caps facility in Kirchberg, Switzerland (Table I).

HPLC Method. The drug content of each sample was determined by HPLC (Agilent Technologies 1200 Series) using a degasser (G1379 B), an isocratic pump (G1310A), an autosampler (G1329A), a variable wavelength detector (G1314B), and a LiChrospher 60, RP select B 125–4 (5 μm) column (Merck, Darmstadt, Germany) at a flow rate of 1 mL/min. The HPLC conditions are described in Table II.

RESULTS

Characterization

Particle Size Measurements

The self-emulsifying systems (CrMTrans and TTMI) used in our study showed different spontaneous dispersion behavior in water (ratio 1:200 v/v) at room temperature. We checked each of the evolving dispersions visually and qualitatively classified the ease of their self-emulsification as "good", "moderate" or "poor". Since TTMI spontaneously formed a transparent dispersion, it was categorized as "good" emulsifying system. This ease of spontaneous self-emulsification behavior was already described by Groves and Pouton (23,24). In contrast, the formulation CrMTrans was rated as rather "poor" emulsifier, since it formed flakes upon gentle agitation, while following short vigorous stirring, the mixture became completely transparent. This result was obviously caused by the formation of viscous structures, which required higher shear forces to disperse the system. Despite the differences in their ease of self-emulsification, both systems resulted in nanodroplets. The droplet sizes of CrMTrans and TTMI

Table II Overview of HPLC Methods Used for Active Compounds Quantification

Active compound	Mobile phase	Injection volume	UV detection
Fenofibrate	Acetonitrile : ammoniumacetate buffer (pH 3.5; 25 mM) (65:35, v/v)	20 μL	287 nm
Probucol	Acetonitrile : water (90:10, v/v)	10 μL	241 nm

were found to be 23.0±1.6 nm (PDI=0.15) and 17.9±1.7 nm (PDI=0.07), respectively. A statistically significant difference between the droplet sizes of diluted formulations was confirmed with $p<0.01$ (*t*-test following an F-test that demonstrated the homogeneity of sample variances).

Rheological Studies

Figure 1 shows differences in viscosities of the hydrated self-emulsifying systems (TTMI and CrMTrans). We observed that, in the case of CrMTrans, the viscosity at lower water contents (10–25% w/w) increased with dilution. It may be related to the swelling of the amphiphilic film as part of the generated microstructure. This increase in viscosity, caused by instantaneous formation of a transparent gel upon hydration, was already visually observed during sample preparation. With further increase in water volume fraction, continuous hydration of the structure was observed. There was a sharp increase in viscosity observed with the CrMTrans system demonstrating a maximum at 30% w/w water, followed by a decrease with further hydration. In contrast, TTMI exhibited only a slight increase in viscosity at the hydration levels of 40%–50% (w/w).

Small Angle X-ray Scattering (SAXS)

SAXS was used to study microstructural changes in hydrated formulations over the whole hydration range. We did not expect a marked effect of the compounds on hydration, because both drugs are neutral, highly lipophilic and low concentrated. Therefore, the controlled hydration was studied using placebo formulations. The peak positions of hydrated TTMI shifted to smaller *q*-values with increasing water content and became narrower up to 55% water content (Fig. 2). For the hydrated CrMTrans, with the increasing water content from 10 to 25%, a shift of the peak to lower *q*-values and a narrowing of the peak could be observed. The narrowest peak was obtained at a water content of 30%. Further water increase resulted in only slight shifts of the peak positions, while a broadening of the peak with increasing water content was noted. Both formulations exhibited at higher hydration levels (for TTMI from 60% up to 90% and for CrMTrans from 35% to 70%) an additional change in SAXS intensity for smaller *q*-values (0.01–0.02 $Å^{-1}$). The appearance of an additional SAXS signal can be attributed to a coexistence of an additional colloidal phase with the general microemulsion structure.

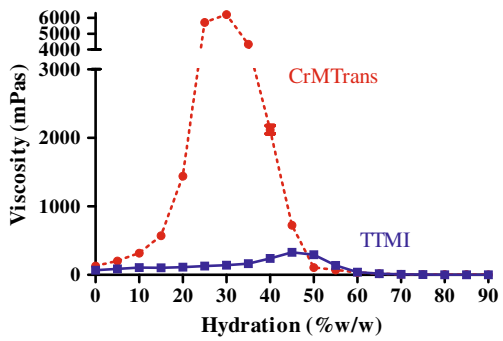

Fig. 1 Viscosity profiles of the self-emulsifying systems upon hydration measured at 200 s^{-1} and 25°C by m-VROC™ rheometer [CrMTrans (*red line*), TTMI (*blue line*)]. The lines are presented as guides to the eye.

The calculated domain sizes *d* and correlation lengths ξ are shown in Figs. 3 and 4. For CrMTrans hydrated from 15% to 25% w/w, and from 45% up to 60% w/w an increase in *d* was found, which was followed by a slight decrease for water contents up to 90%. In contrast, a strong (linear) increase of the correlation length ξ with increasing water content from 10% to 30% w/w could be seen. After reaching a maximum in ξ at 30% water content ($\xi \approx 190$ Å), ξ sharply decreased between 30% and 65% w/w with a subsequent slower decrease towards higher water contents. Both the calculated domain size *d* and the correlation length ξ of hydrated TTMI demonstrated a linear increase with rising water content up to 50% w/w, followed by slight decrease at higher water contents. The exception was at 65% w/w hydration level when *d* showed some increase compared to other hydration levels.

Both formulations exhibited a maximum value in *d* as well as with respect to the correlation length ξ. This scattering behavior was reflecting a hydration process that induced the formation of a more ordered phase. Following the maximum domain size and correlation length, a reverse trend in the swelling process started, which can be viewed as a phase transition. Thus, dominance of a bicontinuous microemulsion structure (with possible existence of local lamellar structures) was receding to evolving colloids of a droplet type.

Analysis of Capsule Disintegration and Drug Release

To better characterize the mechanical changes of the shell material, we used a capsule disintegration testing rig of texture analysis. Thus, the capsule filled with oil (Table I) was placed in biorelevant test medium (FaSSGF) at 37 ± 0.5°C and a plate was kept at constant force to monitor changes in distance. Such changes of displacement were caused by the specific hydration and capsule disintegration mechanisms (Fig. 5). Both S-PVA-C and VegaGels® were first swelling before rupture at the seam occurred. For S-PVA-C partial opening at the seam was noticed by visual observation, whereas for VegaGels® there was a fast and complete opening along the seam. This difference in opening can also be seen in their disintegration profiles. VegaGels® appeared to disintegrate in a two-steps process. A first stage corresponded to the opening at the capsule seam, while the second stage was given by the disintegration of the residual capsule halves. SGC

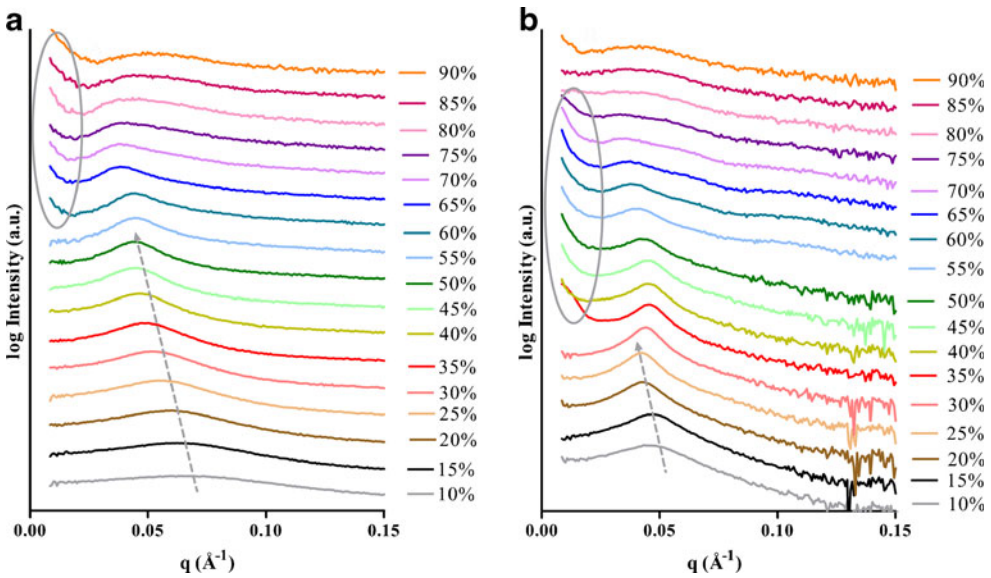

Fig. 2 SAXS intensity profiles (logarithmic scale) for microemulsions consisting of (**a**) TTMI and water and (**b**) CrMTrans and water. The *numbers* next to the each curve indicate different water concentrations in the mixtures. Each curve is shifted by one order of magnitude with respect to the previous one.

exhibited a very fast and complete opening prior to dissolution of the gelatin fragments.

The different disintegration behavior was also reflected by the drug release profiles. Figure 6 depicts the release behavior of fenofibrate formulation (FF-TTMI) in all three capsule types using the biorelevant media. SGC and VegaGels® showed an almost immediate drug release within the first 10 min, which was in agreement with the disintegration profiles confirming a complete capsule opening. A short lag time was noted for the S-PVA-C, which was due to the partial opening of these capsules.

Figure 7 presents the release behavior of the probucol formulation (Pro-CrMTrans) in all three capsule types using biorelevant media. The drug release of Pro-CrMTrans filled in S-PVA-C in FaSSGF was incomplete with high variations (10–30%). A subsequent change to FaSSIF exhibited sustained and comparatively slow drug release also with high variability. This observation may be attributed to the differences in viscosity of Pro-CrMTrans compared to FF-TTMI. The drug release of Pro-CrMTrans filled in SGC and VegaGels® was complete after 30 min with a short delay.

To evaluate the effect of mechanical force on release behavior of the probucol formulation in S-PVA-C, we increased the dipping rate from 10 dpm to 30 dpm. A rate of 10 dpm provided an agitation that is generally considered as physiological for dosage form release in the USP 3 (25). An increase of the dip rate was therefore primarily of interest to mechanistically study the probucol system. We targeted higher shear forces caused by an increased agitation of the reciprocal cylinder. However, the probucol formulation CrMTrans barely revealed a difference in drug release when comparing the two dip rates (Fig. 8).

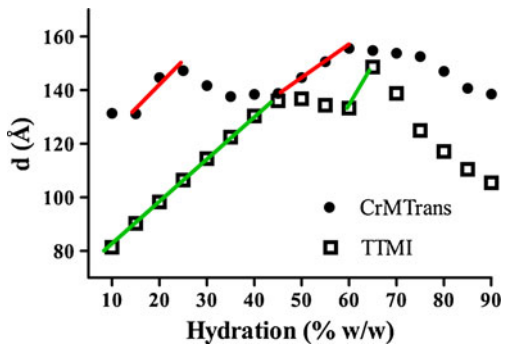

Fig. 3 Plot of domain size (periodicity) *d versus* hydration levels for both model formulations (TTMI and CrMTrans). [CrMTrans (*red line*), TTMI (*green line*)]. The lines are presented as guides to the eye to point at increase in periodicity.

DISCUSSION

For any new capsule technology it is important to study drug release not only in standard buffer solutions but also using

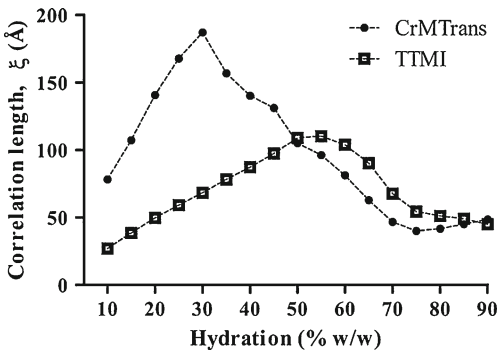

Fig. 4 Plot of correlation length ξ *versus* hydration levels for both model formulations (TTMI and CrMTrans). The connecting lines are for the ease of visualization.

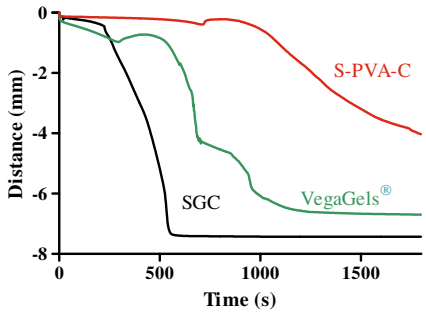

Fig. 5 Comparison of disintegration profiles of oil-filled SGC, VegaGels®, and S-PVA-C analyzed by texture analyzer in biorelevant medium (FaSSGF) at 37 ± 0.5°C. Each curve is a single representative example of $n = 3$ experiments.

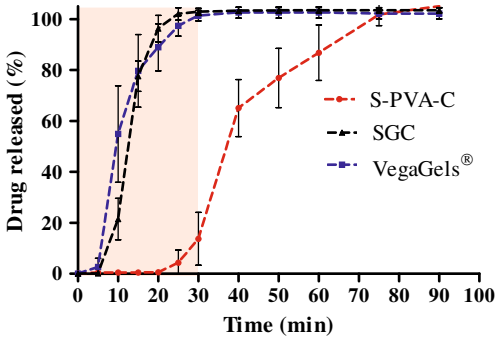

Fig. 7 Release behavior ($n = 3 \pm$ SD) of the probucol formulation (Pro-CrMTrans) from S-PVA-C, SGC, and VegaGels® using the BioDis® apparatus in biorelevant media at 37 ± 0.5°C, 10 dpm (*shaded part* represents drug release in FaSSGF, and *non-shaded* in FaSSIF).

biorelevant media. Although these media only approximate the composition of gastro-intestinal fluids, they still contain the most important components, namely bile salts and phospholipids, which facilitate solubilization of lipophilic drugs in micelles. So the release of poorly soluble, lipophilic drugs is usually enhanced compared to the release rate in simple aqueous solutions (6).

In our previous work we focused on manufacturability and general characterization of the novel thermoplastic soft capsules (S-PVA-C). Benefits were shown for the encapsulation of rather hydrophilic formulations and drug release was studied in water only (2). Considering the importance of more physiological drug dissolution, we focused in the present study on biorelevant drug release using the USP apparatus 3. A special aim was to better understand how the shell material affects drug release by considering changes in the hydrated microstructure of selected self-emulsifying formulations.

Effect of the Shell Material

Disintegration in a compendial sense is defined as the state in which any residue of the unit, except fragments of the capsule shell, is a soft mass having no palpably firm core (26). The process of disintegration consists theoretically of two steps: opening of the capsules and the disintegration of the capsule shell, which may overlap in time. In case of the gelatin capsules, the opening process was usually fast (within the first 5 min) and seemed not to limit the drug release. In contrast to gelatin, there was a general lack of knowledge about disintegration of thermoplastic capsules or their drug release in biorelevant media. This was not only true for the novel material S-PVA-C but also for other thermoplastic capsules such as VegaGels®. Since the thermoplastic capsules may have a specific opening mechanism, depending on the nature of the polymer, their disintegration profiles can vary. Both VegaGels® and S-PVA-C are starch-based thermoplastic capsules that are similar in their qualitative composition (Table I). Their similarity was also reflected by the observed disintegration profiles in biorelevant media. Texture analysis of their disintegration process revealed a swelling (pre-disintegration) before a rupture occurred at the seam of the capsules (Fig. 5). Following an initial swelling, VegaGels® demonstrated a step-wise disintegration pattern. It was clearly possible to distinguish a complete opening of the capsule from the disintegration of the residual shell material. In contrast, S-PVA-C first partially opened and then started to disintegrate, while

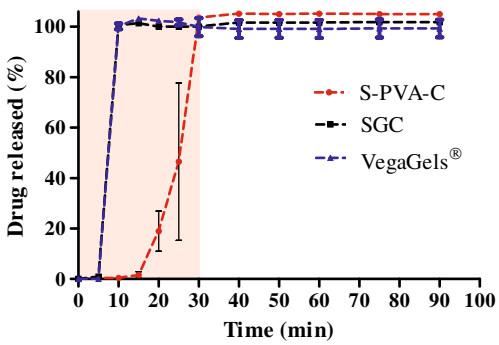

Fig. 6 Release behavior ($n = 3 \pm$ SD) of fenofibrate formulation (FF-TTMI) from S-PVA-C, SGC, and VegaGels® using the BioDis® apparatus in biorelevant media at 37 ± 0.5°C, 10 dpm (*shaded part* represents drug release in FaSSGF, and *non-shaded* in FaSSIF).

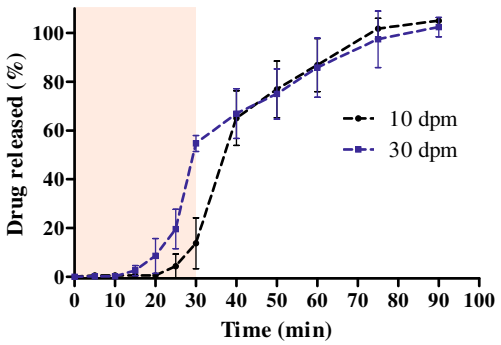

Fig. 8 Effect of the mechanical force on release behavior of probucol formulation (Pro-CrMTrans) in S-PVA-C in BioDis® apparatus in the biorelevant media at 37 ± 0.5°C (*shaded part* represents drug release in FaSSGF, and *non-shaded* FaSSIF).

they continued to open. This different opening pattern was visually observed and was supported by the texture analytical profiles. Moreover, this finding was in agreement with the observed release profiles of S-PVA-C. In the case of the fenofibrate formulation, a short lag time was noted, which was attributed to the partial capsule opening. For the probucol formulation, the interaction of the opening mechanism and poor formulation dispersibility resulted in sustained and comparatively slow drug release. Furthermore, there was no influence noted of a varied hydrodynamics on the release profile (Fig. 8). This was similar to the results of a previous study in which a difference in typical dip rates of the USP apparatus 3 barely affected drug release of tablets (27). A complete and fast opening of SGC and VegaGels® assured an immediate drug release, which was not depending on the nature of the formulation with its hydration process.

In summary, the fenofibrate system easily self-emulsified and drug release was therefore practically independent of the used capsule shell. However, the model formulation of probucol was critically depending on the type of capsule opening. The partial opening of the S-PVA-C in combination with the highly viscous hydrated formulation obviously sustained the drug release. Such hindered drug diffusion was not observed with the same formulation using the other tested capsules that opened rapidly along their entire seam.

Effect of Microstructural Formulation Change

The results of the capsule opening as well as the drug release testing showed, especially for S-PVA-C, that formulation changes during water hydration can be critical. To better understand such effects during self-emulsification, we studied the preconcentrates at different hydration levels. The drug-free formulations were studied while assuming that the compounds would have a neglectable effect on hydration at the rather low concentrations used in this study.

Both model formulations (CrMTrans and TTMI) exhibited along the different hydration levels some similarities. Until the viscosity maximum was reached, the system remained a clear single phase. The drop of viscosity in both systems resulted in slightly turbid mixtures that became practically transparent as the water hydration levels increased. The probucol system (CrMTrans) revealed a sharp increase in viscosity at an intermediate hydration (10–25%, w/w), which was quite different from the hydrated fenofibrate system (TTMI). Such a viscosity peak was previously observed by Fanun (28) in hydrated preconcentrates that were fully dilutable (as a single-phase). It was assumed that structural transitions occurred from water-in-oil to bicontinuous to oil-in-water microemulsions. This view of structural transitions has also been reported by other authors who studied similar self-emulsifying formulations (29). However, the microstructural changes may still be specific for a given system so that different rheological properties evolve during hydration.

For the swelling of an amphiphile, the polar head group is expected to play a key role. Both model systems comprised rather hydrophilic surfactants with a substantial amount of ethylene oxide units. However, the Cremophor-containing formulation (CrMTrans) had a much higher surfactant concentration than the Tween-system (TTMI). This probably contributed to the more viscous structures that were formed upon water addition. Unfortunately, we could not compare surfactant effects at the same concentration levels, because of the individual phase behavior of the formulations that were tailor-made for the different model drugs.

A better understanding of the microstructure was targeted by the analysis of SAXS data. Both systems demonstrated a typical peak for the hydrated systems as it has been observed before in hydrated microemulsion preconcentrates (16–18). Recently, Patil *et al.* discussed the possibility that at least a part of such structure was consisting of local lamellar structures (17). Such structures can be viewed as small stacks of randomly oriented lamellar structures. Local lamellar structures were initially reported in microemulsions by Cabos *et al.* (30). Since there is a lack of long-range and orientational order, no macroscopic birefringence would be expected. Kogan *et al.* described the existence of "ordered bicontinuous structures" as bicontinuous microemulsions based on lamellar phases that have lost the long-range order due to thermal distortions but kept the short-range order (31). In our case, the SAXS diffractograms of both systems developed comparatively ordered structures at an intermediate hydration regime. This increase in ordering has been seen in a shift of the peak maxima to the lower angles *(q)* and a sharpening of the peaks. The samples at intermediate hydration levels were also examined by a polarized light microscope and did not reveal any birefringence, which indicated that no marked liquid crystalline structures were present. The obtained structures were considered to be microemulsions with some short-range order. For such intermediate hydration levels, a bicontinuous microemulsions type is expected. Our data showed in case of TTMI some linearity of d as a function of different water amounts. More complex were changes in the domain size of the CrMTrans system upon addition of water. This formulation showed a maximum in $\xi \approx 190$ Å at 30% w/w, whereas TTMI exhibited a strong linear increase and reached its maximum only at $\xi \approx 110$ Å at 55% w/w. It was a notable finding that the maximum in the correlation length of the CrMTrans system corresponded to the observed sharp viscosity peak. In contrast, hydrated TTMI demonstrated neither a sharp peak in correlation length nor in viscosity.

Our systems also exhibited a typical bell-shaped curve of ξ as a function of the hydration level (Fig. 4). As earlier reported by Fanun (18,28) this behavior could be correlated with structural transitions. Another interesting property is the

ratio of the correlation length to the domain size ξ/d, which indicates a length of order that is relative to a typical domain size. It has been argued that the product 2π *times* ξ/d might be used to differentiate a bicontinuous microemulsion from another type of a Windsor type IV system (14). However, some care is needed with this theoretical argument because real Windsor type IV systems can be difficult to assign to an ideal structural type. Structures of different colloidal nature can also co-exist. As explained earlier, it is well possible that stacks of local lamellar structures were present in our model systems. However, they could have been part of a bicontinuous microemulsion at intermediate hydration levels.

A co-existence of different structures was also an interpretation of the additional SAXS features at very low q, which was observed at relatively higher water amounts in both systems. This signal was likely due to larger colloidal structures such as oil-swollen micelles. Hence, it may hold for the onset of a transition to an oil in water microemulsion.

In summary, the SAXS data demonstrated clear differences in the microstructures of the model systems in terms of how the domain size d and the correlation length ξ changed at different hydration levels. Interesting was the more complex hydration process of the CrMTrans system, which was inferred from the non-linearity of d as a function of increasing water amounts. Our data suggested that different colloidal structures can co-exist in hydrated self-emulsifying systems. The higher structural ordering might have caused an increased viscosity. Although, the microstructural analysis using SAXS effectively complemented the rheological studies of the hydrated formulations, it did not fully explain the huge differences in viscosity between the two hydrated systems. More research is needed to better understand how the microstructure of hydrated self-emulsifying systems is affecting rheological properties.

CONCLUSIONS

This study focused on the biorelevant drug release from S-PVA capsules. Texture analysis of capsule disintegration revealed interesting findings. A clear difference was noted in the mechanism of how the novel S-PVA capsules opened compared to soft capsules of gelatin or of another thermoplastic material (VegaGels®). The different opening mechanism was relevant for the drug release depending on the given formulation. We studied two model formulations which were both preconcentrates of microemulsions, but they significantly differed in their hydrated microstructure. A better understanding of these microstructures was achieved by SAXS analysis. An interaction of the hydrated structures and the shell material was identified for the drug release from the novel thermoplastic capsules.

Our results support the view that a capsule shell material may not be freely selectable for a given formulation. While initial formulation development is typically optimizing the biopharmaceutical performance, some care is needed with selecting an appropriate capsule technology. The choice of a suitable shell material has to be based on improved knowledge of formulation characteristics. Quality control failures in drug release can be avoided by a better understanding of the mechanisms of capsule opening and the microstructural formulation changes during hydration. Ultimately, such knowledge contributes to designing quality into a capsule dosage form. This is a step in the right direction towards the desired state of the quality by design initiative as it is targeted by regulatory authorities as well as by developers of drug products.

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REFERENCES

1. Reich G. Formulation and physical properties of soft capsules. In: Podczeck F, Jones BE, editors. Pharmaceutical capsules. London: Pharmaceutical Press; 2004. p. 201–12.
2. Misic Z, Muffler K, Sydow G, Kuentz M. Novel starch-based PVA thermoplastic capsules for hydrophilic lipid-based formulations. J Pharm Sci. 2012;101:4516–28.
3. Lue BM, Nielsen FS, Magnussen T, Schou HM, Kristensen K, Jacobsen LO, *et al.* Using biorelevant dissolution to obtain IVIVC of solid dosage forms containing a poorly-soluble model compound. Eur J Pharm Biopharm. 2008;69:648–57.
4. Dressman J, Schamp K, Beltz K, Alsenz J. Characterizing release from lipid-based formulations. In: Hauss DJ, editor. Oral lipid-based formulations: enhancing the bioavailability of poorly water-soluble drugs. New York: Informa Healthcare USA, Inc.; 2007. p. 241–56.
5. Borst I, Ugwu S, Beckett AH. New and extended applications for USP drug release apparatus 3. Dissolution Technol. 1997;4(1):11–18.
6. Jantratid E, Janssen N, Chokshi H, Tang K, Dressman JB. Designing biorelevant release tests for lipid formulations: case example - lipid suspension of RZ-50. Eur J Pharm Biopharm. 2008;69:776–85.
7. Jantratid E, De Maio V, Ronda E, Mattavelli V, Vertzoni M, Dressman JB. Application of biorelevant release tests to the prediction of in vivo performance of diclofenac sodium from an oral modified-release pellet dosage form. Eur J Pharm Sci. 2009;37:434–41.
8. Porter CJH, Trevaskis NL, Charman WN. Lipids and lipid-based formulations: optimizing the oral delivery of lipophilic drugs. Nature Rev Drug Discov. 2007;6:231–48.
9. Müllertz A, Ogbonna A, Ren S, Rades T. New perspectives on lipid and surfactant based drug delivery systems for oral delivery of poorly soluble drugs. J Pharm Pharmacol. 2010;62:1622–36.
10. Lopez-Montilla JC, Herrera-Morales PE, Pandey S, Shah DO. Spontaneous emulsification: mechanisms, physicochemical aspects, modeling and applications. J Dispersion Sci Techn. 2002;23(1–3):219–68.
11. Wakerly MG, Pouton CW, Meakin BJ. Evaluation of the self-emulsifying performance of a non-ionic surfactant-vegetable oil mixture. J Pharm Pharmacol. 1987;39:6P.
12. Wakerly MG, Pouton CW, Meakin BJ, Morton FS. The effect of surfactant HLB on the self-emulsifying efficiency of non-ionic surfactant vegetable oil mixtures. J Pharm Pharmacol. 1987;38(S12):2P.

13. Pouton CW. Formulation of self-emulsifying drug delivery systems. Adv Drug Delivery Rev. 1997;25:47–58.
14. Regev O, Ezrahi S, Aserin A, Garti N, Wachtel E, Kaler EW, *et al.* A study of the microstructure of a four-component nonionic microemulsion by cryo-TEM, NMR, SAXS, and SANS. Langmuir. 1996;12(3):668–74.
15. Biradar SV, Dhumal RS, Paradkar A. Rheological investigation of self-emulsification process: effect of co-surfactant. J Pharm Pharmaceut Sci. 2009;12(2):164–74.
16. Patil SS, Venugopal E, Bhat S, Mahadik R, Paradkar AR. Probing influence of mesophasic transformation on performance of self-emulsifying system: effect of ion. Mol Pharmaceutics. 2012;9:318–24.
17. Patil SS, Venugopal E, Bhat S, Mahadik R, Paradkar AR. Micro-structural elucidation of self-emulsifying system: effect of chemical structure. Pharm Res. 2012;29(8):2180–8.
18. Fanun M. Oil type effect on diclofenac solubilization in mixed nonionic surfactants microemulsions. Colloids Surf, A: Physicochem Eng Aspects. 2009;343:75–82.
19. Teubner M, Strey R. Origin of the scattering peak in microemulsions. J Chem Phys. 1987;87(5):3195–200.
20. Klein S. Dissolution test methods for modified release dosage forms, Doctoral thesis. Frankfurt am Main: Shaker-Verlag; 2005.
21. Yu LX, Wang JT, Hussain AS. Evaluation of USP apparatus 3 for dissolution testing of immediate-release products. AAPS Pharm Sci. 2002;4:1–5.
22. Jantratid E, Janssen N, Reppas C, Dressman JB. Dissolution media simulating conditions in the proximal human gastrointestinal tract: an update. Pharm Res. 2008;25(7):1663–76.
23. Groves MJ. Rheological characterization of self-emulsifying oil/surfactant systems. Acta Pharm Suecica. 1976;13:353–60.
24. Pouton CW. Self-emulsifying drug delivery systems: assessment of the efficiency of emulsification. Int J Pharm. 1985;27:335–48.
25. Rohrs BR, Burch-Clark DL, Witt MJ, Stelzer DJ. USP dissolution apparatus 3 (reciprocating cylinder): instrument parameter effects on drug release from sustained release formulations. J Pharm Sci. 1995;84:922–6.
26. The United States Pharmacopeia & The National Formulary. The Official Compendia of Standards, USP 35-NF30 2012. Pharmacopoeial Convention Inc., 2012.
27. Fotaki N, Aivaliotis A, Butler J, Dressman J, Fischbach M, Hempenstall J, *et al.* A comparative study of different release apparatus in generating in vitro-in vivo correlations for extended release formulations. Eur J Pharm Biopharm. 2009;73:115–20.
28. Fanun M. Properties of microemulsions based on mixed nonionic surfactants and mixed oils. J Mol Liq. 2009;150:25–32.
29. Mohsin K, Long MA, Pouton CW. Design of lipid-based formulations for oral administration of poorly water-soluble drugs: precipitation of drug after dispersion of formulations in aqueous solution. J Pharm Sci. 2009;98(10):3582–95.
30. Cabos C, Delord P, Marignan J. Local lamellar structure in dense microemulsions. Phys Rev B. 1988;37(16):9796–9.
31. Kogan A, Shalev DE, Raviv U, Aserin A, Garti N. Formation and characterization of ordered bicontinuous microemulsions. J Phys Chem B. 2009;113:10669–78.