Development of Fixed Dose Combination Products Workshop Report: Considerations of Gastrointestinal Physiology and Overall Development Strategy

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Abstract

The gastrointestinal (GI) tract is one of the most popular and used routes of drug product administration due to the convenience for better patient compliance and reduced costs to the patient compared to other routes. However, its complex nature poses a great challenge for formulation scientists when developing more complex dosage forms such as those combining two or more drugs. Fix dosed combination (FDC) products are two or more single active ingredients combined in a single dosage form. This formulation strategy represents a novel formulation which is as safe and effective compared to every mono-product separately. A complex drug product, to be dosed through a complex route, requires judicious considerations for formulation development. Additionally, it represents a challenge from a regulatory perspective at the time of demonstrating bioequivalence (BE) for generic versions of such drug products. This report gives the reader a summary of a two-day short course that took place on the third and fourth of November at the annual association of pharmaceutical scientists (AAPS) meeting in 2018 at Washington, D.C. This manuscript will offer a comprehensive view of the most influential aspects of the GI physiology on the absorption of drugs and current techniques to help understand the fate of orally ingested drug products in the complex environment represented by the GI tract. Through case studies on FDC product development and regulatory issues, this manuscript will provide a great opportunity for readers to explore avenues for successfully developing FDC products and their generic versions.

From Stomach to Large Intestine: A Thorough Review of Gastrointestinal Physiology – Maura Corsetti, MD, PhD and Bart Hens, PharmD, PhD

From an anatomical point of view, the stomach is divided into a fundus, corpus (*i.e.*,body) and antrum region, but when it comes to motor function two parts can be distinguished: the proximal stomach, consisting of the fundus and the proximal part of the corpus, and the distal stomach consisting of the distal part of the corpus and the antrum. The motility of the proximal stomach is characterized by a maintained status of contractions of the smooth muscle (tone), whereas the distal stomach generates phasic contractions. During the interdigestive phase, the proximal stomach muscle tone is high, whereas the distal

stomach is engaged in a recurrent motor pattern known as the migrating motor complex (MMC) [1]. This complex involves the stomach and the majority of the small bowel (but not the distal small bowel) with three phases: phase I, a quiescent phase with no contractions; phase II with until recently considered random contractions; phase III with a sudden onset of repetitive contractions that also ends abruptly. The phase III can start in the stomach or in the proximal small intestine and then migrate towards the distal ileum. Gastric pH fluctuates during the MMC, with the antral pH being lowest (more acidic) just prior to the start of phase III contractions, and higher at the start of phase I. This change in pH is due to an increase in acid and pepsin secretion that accompanies phase III of the MMC, and bile-free, bicarbonate reflux from the duodenum. Intestinal and pancreatic secretions (*e.g.*, water, bicarbonate and pancreatic enzymes) increase during phase III contractions of the small intestine [2,3]. As soon as the food is ingested, the proximal stomach will relax to accommodate the food, followed by a tonic contraction of the proximal stomach which will push the food more distally. The distal stomach will mix and grind the food by powerful and regular contractions. The duodenum is exposed to nutrients almost directly after the ingestion of food and this will activate a multitude of duodeno-gastric negative-feedback mechanisms, as for instance mediated through vagal reflexes and hormonal signals. This will delay the arrival of acidic, hyperosmotic, or calorie-rich gastric contents into the duodenum by inhibiting proximal gastric tone, and phasic contractions, stimulating the closure of the pylorus [4]. The physical consistency, fat content and caloric load of the meal play a relevant role in regulating the motor response of the stomach. Liquids of low caloric density empty under the pressure gradient created by the fundus tone and the little motor action of the distal stomach in an exponential fashion. Digestible food of more solid consistency requires antral trituration until the particle size is reduced [5]. The time that the stomach takes to reduce the particles may explain the lag phase observed before emptying can start. Thus, gastric emptying occurs in two periods: the lag period (responsible for digestion of solid material) and the post-lag, linear emptying period when digested solid particles or liquids can easily be emptied from the stomach. Non-digestible solids are usually emptied from the stomach with the inter-digestive phase III of MMC [5]. A recent study demonstrated the impact of these phase III contractions to clear ibuprofen from the stomach into the small intestine [6]. These contractions

in combination with the pH played a pivotal role in the onset of intestinal absorption, determining the plasma C_{max} and T_{max}. A clinical aspiration study was recently performed to investigate the gastric emptying rate of a glass of water in fasted and fed state conditions [7]. A standardized dose of phenol red was added to the glass of water and ingested by healthy subjects. After drinking the glass of water, gastrointestinal (GI) fluids were aspirated from the stomach, duodenum and jejunum. Based on computational modeling, authors identified that gastric emptying of a glass of water is tremendously rapid, especially in fasted state, and will be triggered by the present motility at the time of water administration [7–9]. Scintigraphy is considered the gold standard to study gastric emptying in humans, and this is normally defined by the percentage of gastric retention at 1 h, 2 h and at 4 h. However, the use of a single summary outcome measurement does not allow to capture the above-reported complex mechanisms activated by a meal [10]. Nottingham has published the normal values of a gastric emptying test based on a liquid meal, as described by Parker and co-workers, to obtain a comprehensive assessment of gastric motor and sensory function. This test allows differentiating an early and a late phase of gastric emptying for a liquid meal that may reflect the gastric accommodation and the antral component of the gastric emptying [10,11]. Recently, two techniques have been developed to study the gastric function. The SmartPill® is an ingestible device (26 mm by 13 mm) measures intraluminal pH, pressure and temperature. It wirelessly transmits data to a wearable external recorder, allowing ambulatory studies at home [12,13]. The variations in luminal pH, as well as the drop in temperature after defecation, allows accurate measurement of regional as well as whole gut transit times. However, it should be noted that in consideration of the dimension of the device does not reflect the gastric emptying of normal digestible food and indeed the gastric emptying has been found to be longer than that measured by scintigraphy [13]. In any case, this technique has the advantage of being non-invasive and of combining the measurement of pH and of the whole gut transit time. Besides telemetric capsules, magnetic resonance imaging (MRI) has been recently applied to the study the GI function and this technique offers some major advantages compared to other techniques: it is non-invasive, does not expose subjects to ionizing radiation, and does not require any contrast medium. It is a unique technique

that offers the possibility of simultaneously measuring gastric, small intestinal and colonic volumes, the physicochemical characteristics of the luminal environment, transit rate and to quantify motility [14]. This technique has not yet been standardized across research centers and for the moment does not allow the evaluation of gastric function in an upright position [15]. In summary, the human stomach is more complex as it seems and can play a major role in further intraluminal drug behavior along the intestinal tract where absorption takes place.

Beyond the stomach, the intraluminal processes in the small intestine will play a pivotal role with respect to drug absorption. In this second talk, there was a specific focus on (i) the residual intestinal fluid volumes, (ii) the characterization and composition of the intestinal fluids and (iii) the permeability of the intestinal wall for drug compounds. The residual fluid volumes in the intestinal tract are rather scarce and not homogenously distributed as a pool of water from the proximal towards the distal part. Distribution of these fluids is organized in different fluid pockets [9,16]. The variability in the number of pockets and the actual volume for each pocket is tremendously high between healthy subjects, as highlighted by Mudie and co-workers [9]. This finding was an important investigation for formulation scientists to be aware of the fact that the intestinal tract is not like a 'swimming pool', completely filled with water. The prediction of the *in vivo* performance of orally administered drug products has shown to be more accurate when applying the fluid dynamics as observed by Mudie *et al.* instead of using static and high volumes. This was observed for posaconazole, a weakly basic compound, for which the *in vivo* performance was predicted by using a dynamic fluid and pH model in simulation software [17]. Although this model shows to have an impact on predicting the *in vivo* performance for compounds suffering from a poorly aqueous solubility, authors concluded that this model may not have an immense impact on the predicted systemic exposure for compounds characterized by a high solubility. Moreover, as mentioned before, there is huge intersubject variability in the number and volume of pockets. For instance, one subject showed to have only 2 pockets with a total volume of 1.4 mL whereas another subject demonstrated to have 23 pockets with a total volume of 160 mL. A follow-up study aims to unravel a potential link between the appearance of fluid pockets and

the present motility [14]. In the seventies, Vantrappen *et al.* observed a higher secretion rate of bicarbonate shortly after an upper GI phase III contraction [3]. In doing so, the gastric acid of the stomach entering the small intestine could directly be neutralized by the bicarbonate buffer. This so-called '*secretomotor complex'* is highly likely to be a responsible factor in the formation of water pockets inside the intestinal tract. Besides gaining knowledge with respect to the present volumes in the GI tract, the composition of these fluids is another important aspect. In a recent study, human duodenal fluids were aspirated from 20 healthy subjects in the fasted and fed state [18]. The fed state was simulated by ingestion of a liquid meal (*i.e.*, 400 mL of Ensure Plus®, equal to 700 calories). After aspiration of these fluids as a function of time, fluids were analyzed for pH and endogenous constituents (bile salts, phospholipids, cholesterol, enzyme activity and lipid digestion products). The results of this study demonstrated wide variability in the presence of these constituents from person to person, although the study protocol was the same for each and every individual [18]. Especially for ionized compounds, the present pH in the intestinal tract is from paramount importance in order to dissolve and, subsequently, absorb. The research group of Prof. Amidon (University of Michigan) aspirated GI fluids from 37 healthy subjects after oral intake of an immediate-release ibuprofen tablet (800 mg) in fasted and fed state conditions [6,19]. Fluids were aspirated from different segments of the GI tract: stomach, duodenum and jejunum. This study demonstrated the highly fluctuating pH, especially in the duodenum, which was an important intrinsic factor besides motility explaining differences in systemic exposure of ibuprofen between and within subjects (Figure 1).

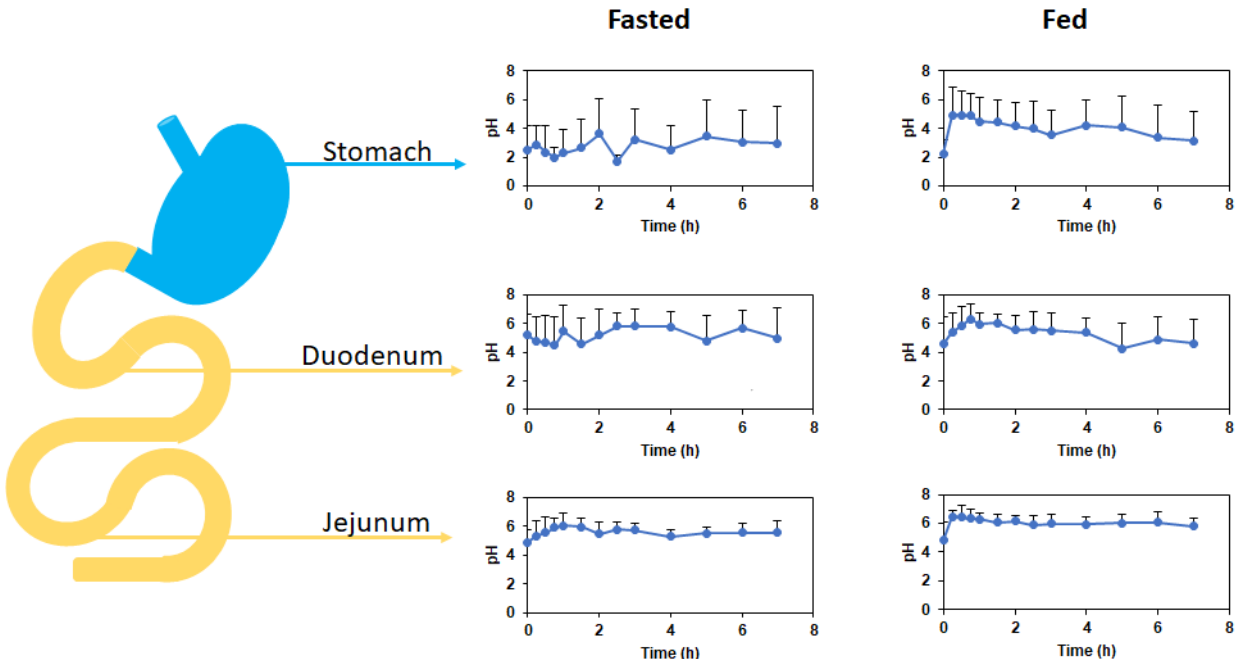

Figure 1: Mean pH versus time profiles in fasting (n = 20) and fed state (n = 17) conditions as measured in the stomach, the duodenum, and the jejunum (mean + SD). Figure depicted from Hens et al. 2017 [6]. Copyright ACS 2017.

Besides solubility, absorption has always been a key parameter in estimation of drug performance. Multiple techniques are described in the literature to assess the intestinal permeability of drug compounds. The Loc-I-Gut® method, *i.e.*, a double-balloon perfusion system, is an interesting study technique to explore the permeability for drug compounds in the different regions of the GI tract [20,21]. A specific region of the GI tract will be inflated by two balloons and thus separating a specific region of interest. Subsequently, a drug solution will be perfused and the amount of drug that will disappear is a measure for the amount of drug absorbed. The application of this technique has unraveled the intestinal permeability for hydrocortisone in the duodenum, jejunum and ileum. A recent review by Dahlgren *et al.* compiles historical P_{eff} data from 273 individual measurements of 80 substances from 61 studies performed in all parts of the human intestinal tract [22]. This impressive data set has served as a reference for researchers in order to optimize the protocols of *in vitro* setups in order to improve the predictive performance of their in-house absorption tools [23].

With respect to the colonic physiology, recent findings, applying high-resolution manometry (HRM), have demonstrated that colonic motility is mainly represented by non-propagating and retrograde activity and both these activities increased soon after intake of a meal. These colonic motor patterns have the role of delaying the arrival of colonic content to the rectum and of favoring the retrograde filling of the transverse and ascending colon, where the propagating contractions normally start. Propagating contractions, including the high-amplitude propagating contractions associated with movements of solid colon content, represent a minority of the colonic activity and are normally more frequent about 1-2 hours

after the meal and upon awakening [24]. The reason of this is likely related to the fact that, in these moments of the day, the arrival of the content accumulated in the distal small bowel during the night and during the inter-digestive periods determine the distension of the ascending and transverse colon that trigger the propagating activity. The prevalence of non-propagating and retrograde activity explains the fact that the normal colonic transit time is slower (about 35 hours) as compared to the small bowel. This allows the colon to perform its functions of absorption, fermentation and to be an adequate reservoir organ. HRM is a useful technique to study colonic motor function but is invasive and normally requires a preparation of the bowel. This makes the technique less attractive when the colonic function needs to be studied under physiological conditions. Recently, other techniques have been applied to study the colonic function. The electromagnetic capsule is an ingestible silicone coated cylindrical magnet (21 mm by 8 mm) used to map the real-time movements of colonic contents. A plate containing a detection matrix of 4 x 4 magnetic field sensors is worn by an ambulatory patient around the abdomen to detect the movements of the pill. This matrix allows mapping of the pill movements in the x-, y-, and z-axis as well as the inclination angles applied by the colon. The pill allows evaluation of the direction (anterograde and retrograde), velocity and length of movement of intraluminal content allowing the calculation of the colonic transit time. Recent studies have also demonstrated the first identification of colonic motor patterns consistent with those seen with HRM [25]. Moreover, MRI has also been introduced as it is able to measure both the colon free water content and the "fluidity" of the colonic content [26]. Recent animal studies have demonstrated that the colon is able to adapt to the physical characteristics of the intraluminal content and develops different motor response according to the presence of more or less fluid content [27]. It is highly likely that these physiological variables play a pivotal role in the dissolution and/or absorption of drugs that are triggered to be released at the colonic site in the human GI tract.

Integration of GI Physiology Into a Predictive Dissolution Device: Where to Start? – Raimar Löbenberg, PhD

The GI tract is a complex and not well-understood sequence of organs with changing environments as a function of time. However, an in-depth mechanistic understanding of the obstacles and opportunities in each segment is necessary to achieve optimal drug absorption and bioavailability (BA) (Figure 2).

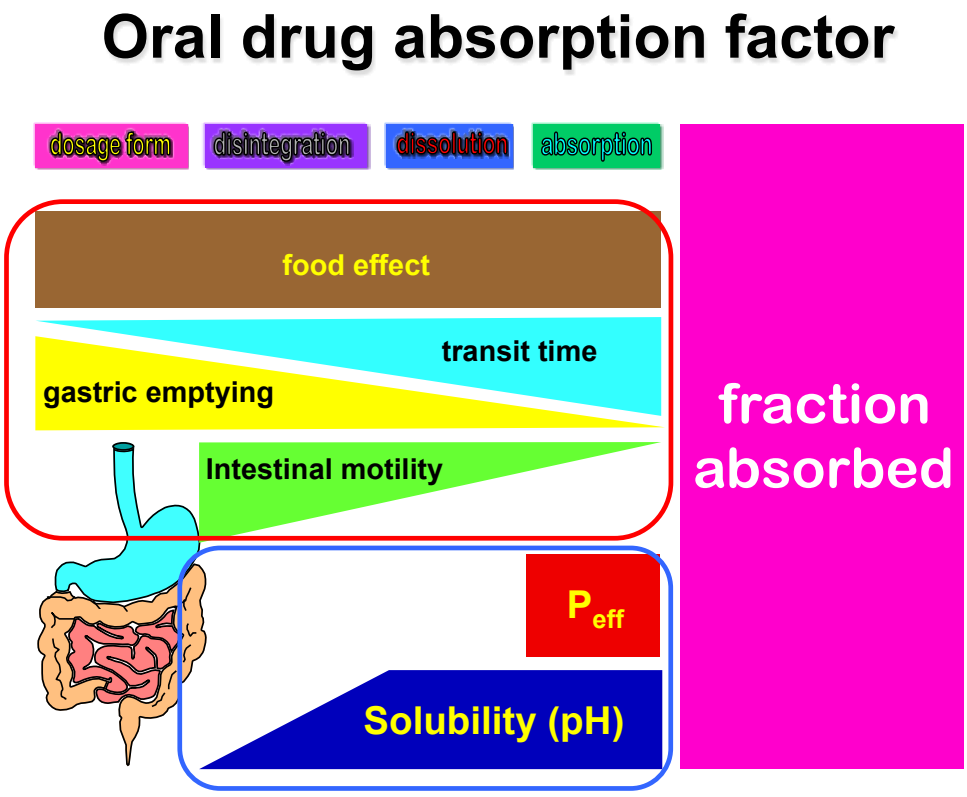

Figure 2: An overview of the different GI physiological variables that can have a major impact on oral drug behavior in the GI tract.

Figure 2 shows multiple factors impacting the fraction dose absorbed without considering metabolic or drug stability compromising degradation processes. The Biopharmaceutics Classification System (BCS), represented by the blue box, focuses on permeability and solubility [28]. However, drug dissolution and solubility depend on additional physiological factors that are summarized in the red box. Motility effects and gastric emptying are know to have an impact on the performance of a drug product but they are seldom considered in drug development. In contrast, food effects, pH effects and solubilization effects by bile salts were studied intensively in the past decades. However, today, there is still no consensus

on a universal dissolution media, which can be used in drug development and for *in vitro* performance testing to capture these effects. Early studies evaluated the solubility of glyburide, a BCS class II drug, in biorelevant media [29]. It was shown that the increased solubility in bile salt media (containing sodium taurocholate and egg-lecithin) was suitable to establish an *in vivo-in vitro* correlation (IVIVC) when computer simulations were applied. A linear regression was established in GastroPlus™. Applying a biorelevant solubility value resulted in a regression coefficient of 0.94 for the reference formulation. The prediction error (%) regarding simulated plasma C_{max} and AUC were7 and 14% , respectively, when using these biorelevant solubility value as an input in GastroPlus™. Solubility values obtained in aqueous media (pH 6.5) resulted in a 38 and 63% prediction error with respect to plasma C_{max} and AUC.Later on, a dynamic dissolution protocol was developed in biorelevant media (*i.e.,* FaSSIF) which again showed predictive power for establishing an IVIVC [30]. The dynamic dissolution protocol was then applied to a flow-through apparatus for montelukast sodium. Again, the biorelevant media gave the best fit to clinically observed data [31]. These early studies were successful to establish IVIVC without considering other GI factors. In a study by Almukainzi *et al.*, the impact of gastric motility on the pharmacokinetics (PK) of meloxicam was studied [32]. It was observed that two formulations (conventional versus fast dissolving) had a similar PK pattern when administered in a rodent model. However, when the gastric motility was impaired the stomach controlled the drug release and therefore the drug absorption for the conventional dosage form. The PK of the fast dissolving formulation was close to the pattern observed in the healthy state. This study indicated that formulation differences, which are not relevant under healthy conditions, might result in significant differences under disease state. This study showed that the stomach in disease conditions is able to negatively impact PK parameters such as plasma C_{max} and T_{max}. Furthermore, it is well accepted that gastric emptying impacts the PK in fasted versus fed state for many drugs. However, less attention is given to the fact that GI motility impacts C_{max} and T_{max} depending on the dosing time and the MMC phase. This might be due to the fact that the PK models used to quantify and describe the PK behavior of drugs sooth-out individually observed variability in the mean PK profiles. However, if motility and PK are both monitored a relationship between observed plasma levels and intestinal motility are getting more obvious. Another

243 factor for alternations in drug absorption is the composition of the intestinal juices. The buffer system in
244 the GI tract is carbonate-based. In routine pharmaceutical quality control (QC) and development, phosphate
245 buffers play a major role while carbonate buffers are seldom used. The choice of phosphate over bicarbonate
246 seems to impact the *in vivo* performance of enteric-coated dosage forms. Early reports show the failure of
247 enteric-coated products *in vivo* (1964) and are confirmed over several decades until today by *in vivo* studies
248 [33–35]. Also, there is evidence that phosphate and carbonate buffers seem to interact differently with the
249 enteric-coated polymers. It is obvious that a re-evaluation of established *in vitro* testing is important to
250 capture *in vivo* relevant performances to avoid product failure. The next important differences, besides
251 buffer nature, are buffer strengths used in *in vitro* dissolution protocols versus the present buffer strength
252 in the GI tract and the impact of the intestinal absorption on drug dissolution. Biphasic dissolution is known
253 for many years as a surrogate to assess the the in vivo performance of a drug formulation [36]. Based on
254 the permeated amount of drug appearing in the organic layer, estimations related to the fraction absorbed
255 can be performed [37]. However, its impact on IVIVC has not yet been fully appreciated. In a recent study,
256 we investigated the dissolution behavior of ibuprofen in pharmacopeial and GI equivalent phosphate buffer
257 strength. The results showed that ibuprofen dissolved fully under the pharmacopeial conditions in less than
258 15 min. However, at low buffer strengths, this process took much longer, and the pH of the media changed
259 significantly due to the acetic nature of ibuprofen. However, if a biphasic dissolution test was performed,
260 the pH recovered over time close to the original value. This again demonstrates how important
261 physiologically adapted *in vitro* testing can be to capture what happens *in vivo*. Only this can ensure that *in*
262 *vitro* methods are predictive of *in vivo* performance. The translation of such methods into QC methods
263 needs to be investigated in the future in more detail. The last aspect deals with the irrelevance of *in vitro*
264 behavior on the drug product performance *in vivo*. An example of such rare case is dextromethorphan [38].
265 This drug is absorbed to over 80% in 2 hours but it takes about 15-20 hours to observe the maximum
266 fraction dose absorbed. A classical IVIVC would correlate fraction dose absorbed versus the dose dissolved.
267 However, in this specific case, the IVIVC would be misleading. The drug dissolves fast in the gut and is
268 completely dissolved within 15 min. As mentioned before, >80% will be absorbed into the enterocytes

within 2 hours. The drug undergoes lysosomal trapping after entering the enterocyte. As a weak base, it is highly lipophilic at physiological pH in the cytoplasm. As the drug will migrate through the enterocytes from the apical to the basolateral side, it can pass through the membranes of the lysosomes and it can enter into an aqueous environment with a slightly acidic pH. In this organel, the weak base becomes more hydrophilic and, therefore, will be entrapped in the lysosomes. That isis why it takes more time to appear in the blood than it takes time to be absorbed. Such drugs dissolution tests are not useful surrogates for *in vivo* performance since the dissolution of the drug product cannot be directly correlated to the plasma levels. It is the biological system and its specific environments and drug partition between the cell compartments, that determine the appearance of the drug in the central compartment and not the drug dissolution. In summary, GI drug absorption is highly impacted by different physiological factors. *In vitro* performance testing should consider and include physiologically-adapted test protocols to identify potential clinical relevant dosage form factors. A BCS sub-classification system, which includes acids, bases and neutral molecules can help to identify potential obstacles for oral drug absorption for these different groups [39]. To meet all these standards, a potential *in vitro* apparatus, which can simulate the different GI conditions, is shown in Figure 3.

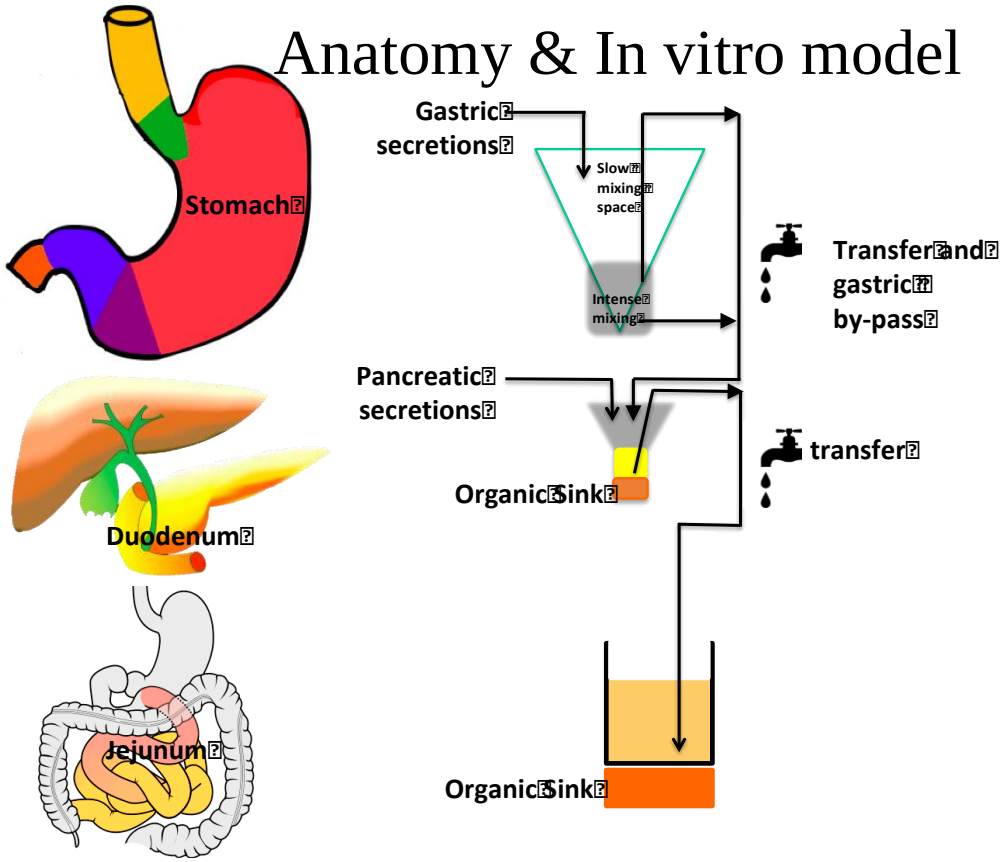

Figure 3: Illustrative presentation of an *in vitro* dissolution model taking into account the different physiological barriers of the GI tract that may have a major impact on drug's dissolution and absorption.

***In Vitro* Dissolution for a Marketed and Generic FDC Drug Product: Bioequivalent or not? – Marival Bermejo, PhD**

Development of Fixed-Dose Drug Combination (FDC) products could be challenging when both drugs do not belong to the same BCS class *i.e.*, when the limiting factors for their absorption are different. In the first part of the presentation, the relevance of exploring the biopharmaceutical properties of each drug in the combination product were discussed in the framework of different classification systems. The BCS system has evolved from a regulatory conservative classification framework in which the main concern is to ascertain the non-bioequivalence (non-BE) risk to a development tool which can help on the formulation strategy selection [40,41]. In order to understand the biopharmaceutical limiting factors for a given drug the cut-offs and methods for permeability and solubility estimation of BCS are modified in the

developability classification system (DCS). The DCS considers a higher available fluid volume (500 mL) in the small intestine and the solubility in human intestinal fluids for solubility classification. The volume of 500 mL is calculated based on the co-administered fluid and present residual fluid along the GI tract [40].Another relevant addition is the differentiation between solubility-limited and dissolution-limited drugs as the formulation approaches may differ. The selection of the dissolution test to explore the risk of the non-equivalence outcome *in vivo* can be made based on the drug physicochemical characteristics. For that purpose, a sub-classification system from BCS was proposed by Tsume *et al.* [39]. BCS class II drugs were sub-classified in neutral (BCS IIc), weak acids (BCS IIa) and weak bases (BCS IIb). Following these sub-divisions, the suggested dissolution tests to forecast *in vivo* behavior differ from class I and III for which simple dissolution apparatus (as USP II) could suffice and from class II and IV for which a gastric compartment and an absorptive sink should be included in order to increase the *in vivo* predictability. To accommodate that need, several dissolution system have been proposed in the literature and as example several transfer systems and two-phase or biphasic dissolution systems were described [37,42–47]. In the second part of the lecture, the potential effects of formulation excipients were discussed in as well as experimental preclinical models to study those effects. Excipients can affect membrane permeability and metabolism and GI motility either at gastric emptying level or at intestinal level. In Table 1, some experimental methods with useful references are summarized.

Table 1: Overview of potential in *vitro/in situ* methods to apply in order to explore a physiological variable of interest.

Effect	Model	Reference (PMID)
Intestinal permeability	Caco-2; *In situ* perfusion (Rat –Mouse)	[48–52]
Intestinal metabolism	*In situ* perfusion in addition to mesenteric vein canulation	[48,53,54]
Gastric emptying	Charcoal suspension Rat; Phenol red + Loperamide; Barium suspension	[55–57]
Intestinal motility	Charcoal suspension Rat	[55]

For instance, the effect of sodium lauryl sulfate (SLS) on the intestinal permeability of fexofenadine was characterized with Doluisio's closed-loop perfusion method and further evidenced by *in vivo* BA studies in rats [58,59], while the relevance of gastric emptying changes due to excipients as the reason for a failed bioequivalence study was assessed with a barium sulfate gastric emptying test in rats [60]. Finally, the concept of using BCS as a risk assessment tool of bioequivalence(BE) issues was with the aid of a case study of an FDC development. A valsartan/hydrochlorothiazide generic product failed twice the BE test in each one failing for one of the drugs while succeeding for the other one. The application of a biopredictive dissolution test using the Gastrointestinal Simulator (GIS) was successful in reproducing the *in vivo* outcome as differences in disintegration in the stomach chamber and differences in dissolution rate on the intestinal compartments were the apparent reasons for the *in vivo* failure due to different levels of sorbitol and SLS on the generic formulations. To conclude, BCS and/or DCS classification of drugs in an FDC is a tool to define the absorption limiting factors and the relevant physiological variables affecting BA. For FDC with drugs belonging to different BCS classes a combination *in vitro* dissolution methods and preclinical models is necessary to assess formulation performance.

Challenges and Opportunities to Grant BCS and Dose Strength Based Biowaivers for FDC Products – Pablo M. González, PhD

FDC products combine two or more active pharmaceutical ingredients (API) in a finished pharmaceutical dosage form at a fixed ratio of doses [61]. FDC products are approved based on the combination rule that states that each component should contribute to product effectiveness and that the combination should also be safe in a particular patient population [62,63]. Safety and efficacy data can be totally (New Drug Application) or partially (505(b)(2)) original or based on previous reports (Abbreviated New Drug Application) [64]. FDC products offer several advantages over co-administration of the single entity product (SEP) such as greatest patient compliance, increased safety and efficacy, minimized abuse potential, and reduced cost for patients. They also offer opportunities for manufacturers to extend intellectual property and exclusivity along product life-cycle [65]. On the other hand, formulating FDC products impose several challenges related to incompatibility between APIs and incompatible interactions with certain excipients. Some drugs might degrade in presence of another (amiodaquine HCL-artesunate), others might be pharmaceutically incompatible (simvastatin-telmisartan) [66], some drugs could display very different viscoelastic properties (metformin-glibenclamide), and others might interact at the absorptive (*e.g.*., intestinal transporters) or post-absorptive (*e.g.*, metabolic enzymes, renal transporters) level.

WHO classifies FDC products into 4 different scenarios regarding regulatory requirements for product registration:

- Scenario I: The new FDC product has the same APIs and doses as an existing FDC product
- Scenario II: The new FDC product has same APIs and doses as an established regimen of single entity products (SEP)
- Scenario III:
    - a) The new FDC product combines APIs with established safety and efficacy data but that have not been used in combination for that particular indication

- b) The new FDC product comprises a combination of APIs with established safety and efficacy but will be used in a different dosage regimen

- Scenario IV: The new FDC product contains one or more new chemical entity (NCE)

BE studies are required in order to bridge pivotal clinical data of the reference listed drug (RLD) product(s) to safety and efficacy of FDC products belonging to Scenarios I and II. While the design of BE study for scenario I is standard, in scenario II the *in vivo* performance (*e.g.,* PK end-points) of the FDC product is compared to the co-administration of the SEPs. In both cases, successful BE indicates the absence of (or similar) PK interactions between APIs. However, BE studies for FDC products are challenging due to: i) potential changes in PK intra-subject variability in the combination product; ii) non-linear PK in a line of strengths; iii) drug-formulation interactions; and iv) differential impact of food on API PK when administered as a combination product [67]. These considerations make biowaivers a highly attractive opportunity for manufacturers to fulfill the BE requirement. Currently, WHO, the Food and Drug Administration (FDA), the European Medicines Agency (EMA), the International Conference on Harmonisation (ICH), and Health Canada allow BCS-based biowaivers for immediate release (IR) FDC products containing high-solubility APIs only [68–70]. Thus, FDC products containing BCS class I and/or III APIs could apply for a biowaiver. In general, dissolution, and compositional requirements are the same as those for SEP, with some differences among jurisdictions. For BCS class I API FDA requires the use of excipients present in currently FDA-approved IR products, while EMA encourages the use of similar amounts of the same excipients as the reference product. The 2018 ICH Guidance on BCS-based biowaivers states that critical excipients (*e.g.,* polysorbate 80, sorbitol) must be within ± 10% of reference product. On the other hand, there is a consensus among jurisdictions regarding the impact excipients might have on BCS class III drugs, such that agencies require excipients to be qualitatively (Q1) the same and quantitatively (Q2) very similar to the reference product. FDA and ICH guidances contain tables with allowable compositional differences of excipients (by function) relative to the reference product. The implementation of a BCS-based biowaiver for a scenario I-type FDC product is straight forward provided products are

pharmaceutically equivalents and dissolution and compositional requirements are fulfilled. Additionally, FDA might accept BCS-based biowaivers for pharmaceutical alternatives if appropriately justified. On the other hand, a BCS-based biowaiver for scenario II-type FDC products impose some challenges for both manufacturers and regulatory agencies. First, different single-entity RLD products might be registered in different regions, implying that a manufacturer would have to perform multiple biowaiver studies if pursuing approval in various jurisdictions. This can be further complicated by the fact that unlike FDA, EMA does not publish a list with RLD for different European countries. Second, FDC containing incompatible APIs need to incorporate a segregation technology (*e.g.*, bilayer tables, tablet-in-tablet, etc.) in order to obtain a stable product. In this case, it might be difficult to account for the compositional requirement between the FDC product and the respective SEP. Third, dissolution methods to study FDC products with large dose disparity between APIs (*i.e.*, dose ratio > 50) might be analytically challenging. This could be further complicated in cases where APIs display divergent pH-dependent stability in the physiological range. Furthermore, RLD SEPs might use different dissolution apparatus (*e.g.*, basket or paddle) such that manufacturer might have to develop and validate two dissolution methods for one FDC product. Fourth, there is a chance for pre-absorptive PK drug-drug or drug-formulation interactions (DFI) in FDC products that could be either different or absent when the SEPs are co-administered. Both FDA and EMA have published guidelines regarding studying drug-drug interactions (DDI) at the transporter level [71,72]. FDA has also published methodological recommendations to study *in vitro* transporter-mediated DDI [71]. While agencies require sponsors to study intestinal efflux transporter-mediated DDI (*i.e.*, P-glycoprotein, breast cancer resistance protein), there is currently no published recommendation on studying potential DDI mediated by intestinal uptake transporters. This seems surprising since it is well recognized that intestinally expressed uptake transporters interact with a vast number of drugs belonging to structurally diverse chemical and therapeutic classes [73]. Moreover, there is growing evidence that pharmaceutical excipients can inhibit both efflux and uptake intestinal transporters *in vitro* and *in situ*. Documented examples include PEG-ylated surfactants, sorbitan fatty acid esters, and polyethylene glycol [74–76]. While there is a consensus that DDI or DFI might be of minor clinical relevance for BCS class I drugs, there also

413 a concern that these interactions could greatly impact the oral absorption of low permeability APIs. FDC
414 products also offer opportunities for developing a line of strengths that can be used to optimize therapy by
415 dose titration. Intermediate and low strengths could apply for a dose strength (DS)-based biowaiver
416 provided there is at least one strength (typically the highest) that successfully demonstrated BE to the
417 reference product *in vivo*. Dose strength-based biowaivers are applicable to APIs that are not eligible for
418 BCS-based biowaivers and to pharmaceutical forms other than IR (*i.e.*, modified-release, delayed-release).
419 Common requirements for DS-based biowaivers among jurisdictions are linear PK in the therapeutic dose
420 range, with a chance for bracketing approach between the highest and the lowest strength, and same
421 manufacturing process for the strength line [77]. The dose range for a FDC will be dependent on the additive
422 or synergistic effect of the investigational drugs. The interaction between the drugs are assessed in drug-
423 drug interaction and PK-PD studies. Subsequently, exposure-response models can be used for Phase 2B
424 dose selection [78]. As in the case of BCS-based biowaivers, DS-based biowaiver requirements are an
425 extension of those for SEPs. Tables 2 and 3 summarize FDA and EMA compositional requirements and
426 dissolution method recommendations for DS-based biowaivers. Data presented in Tables 2 and 3 imply that
427 manufacturers pursuing a DS-based biowaivers in the US and European market might face challenges
428 fulfilling compositional requirements for FDC products based on segregation technologies (*e.g.*, bi-layer
429 tablets) since EMA treats each layer as a separate entity while FDA considers bi-layer tablets as a single
430 unit. Also, in the case of single unit FDC products with large dose disparity between APIs it might be very
431 difficult to fulfill proportionality requirements by both FDA and EMA. More specifically, EMA states that
432 in order to calculate API/excipients proportionality the other API must be considered an excipient.
433 However, it is not clear whether the other API must be considered as a filler for proportionality calculations.
434 Similarly, there is no specific FDA recommendation as to how to consider the other API in bi-layer tables.
435 These discrepancies can hinder simultaneous registration of an FDC product in both USA and Europe.
436 Additionally, while FDA requires bioequivalence studies for the highest dose in the strength line, EMA
437 requires studies at the lowest strength in addition to the highest strength. Finally, the existence of different

reference products among jurisdictions increases the number of studies a sponsor needs to execute if seeking approval in various regions.

Table 2: Comparative compositional requirements to grant dose strength-based biowaivers by FDA and EMA*.

Criteria	FDA	EMA
General composition	All ingredients and APIs are in the same proportion between diff. strengths	Q1 the same and Q2 proportional across different strengths
High-potency APIs	• Total weight nearly constant across strengths (±10% from bio-batch) • Q1the same across strengths • Only APIs vary across strengths, and one or more excps.	• Amount of API <5% core weight or capsule filling • Amount of excps. constant only API varies • Only filler changes to account for changes in APIs
ANDA	Proportion between API and excps. might vary across strengths if same BA is achieved	No special considerations
Bi-layer tablets	Bi-layer tablets are considered as a single unit	Each layer is considered independently
Prolonged Release	• Beaded capsules: only number of beads varies across strengths • Single unit products similar general requirements	• Multiple unit formulation: BEq for the highest strength • Single unit formulation: bracketing approach • Release-controlling (or coating) excps. must be the same for the line of strengths.
FDC	Not discussed	• Proportionality requirements must be fulfilled for all APIs • The other APIs must be considered an excp., except in bi-layer tablets

*Adapted from [77].

449 **Table 3:** Dissolution method recommendations by FDA and EMA for dose strength-based biowaivers*.

Criteria	FDA	EMA
IR products	i) Compendial method ii) FDA recommended /USP general chapter iii) Develop new method using diff. agitation speeds, pH (1.2, 4.5, 6.8). Water can be used. Add surfactants if API is poorly soluble	i) pH (1.2, 4.5, 6.8) and QC method ii) If sink condition cannot be achieved at a particular pH for all strengths, compare to dissolution profile of RLD at same dose or using multiple units of lower strengths
MR products	• If no compendial method submit ii) + pH (1.2, 4.5, 6.8) for comparisons • Select the most discriminating conditions (agitation, media) based on in vitro and in vivo data	

450 *Adapted from reference [77].

451

452 **<u>Considering the Biopharmaceutics and Physicochemical Aspects of FDC – Amitava Mitra, PhD</u>**

453 Amitava Mitra, Ph.D. (Sandoz, Inc, A Novartis Division) discussed the key challenges and strategies to
454 overcome such challenges, in achieving BE for FDC products containing two or more of active ingredients
455 [67]. The active ingredients of these products may work through different pharmacological pathways and
456 offer advantages of additive/synergistic effect, reduced dose of each active, and improved patient
457 compliance. Novel FDCs of Parkinson's drug, Levodopa, are an example of efforts to improve the clinical
458 outcome of an old drug using new technologies and mechanisms to improve patient function [79]. However,
459 combining multiple active ingredients may complicate their individual biopharmaceutic and PK behavior.
460 The development of controlled or modified release FDC products does add additional challenges due to
461 changes to the drug release profiles. Such changes in the release profile can change the biopharmaceutic
462 and pharmacokinetic profiles of the API. Interested readers should review the following published
463 references [67,80–82]. The importance of critically reviewing the physicochemical and biopharmaceutics
464 properties and their impact on PK of the individual drugs being considered for the FDC was also discussed.
465 Gaining a thorough understanding of the PK properties of the individual drugs along with the formulation
466 variables being considered for the FDC is an equally important consideration. Pilot BA studies designed

to answer the most pertinent questions relating to the FDC strategy are important and encouraged. However, underpowered studies with too many variables can further confound an already complex issue and should be avoided. Pivotal BE studies should be designed with due consideration of all the physicochemical, biopharmaceutic, and PK data for the compound from all sources. BE study designs specific to highly variable drugs such as scaled BE or cross-over replicate designs may be considered. Leveraging the knowledge gained from varying but synergistic techniques such as *in vitro* solubility/dissolution studies, in silico absorption models and IVIVC's, *in vivo* preclinical animal models, and the available *in vivo* clinical data is paramount to the success of the FDC strategy for a given combination. Two case studies were discussed where the use of oral absorption modeling, dissolution data and clinical PK data were used to successfully develop FDC products. In the first case study, the development of a triple combination product was discussed, where one of the active ingredients had a highly variable C_{max} and another active had a long T_{max} due to bile secretion and slow absorption. In this case oral absorption modeling was key to understanding the impact of formulation changes on PK of the three actives and ultimately in development of the FDC product. In the second case study, the development of a double combination product was discussed, where one of the active ingredients was a weak base with high intra-subject CV and steep pH-solubility profile. In this case, data from several relative BA studies and a thorough understanding of the PK and biopharmaceutic properties helped with the successful development of the FDC.

Current Regulatory Requirements to Assess Bioequivalence of FDC Products Worldwide (EU/USA/ Latin America/Japan) – Alexis Aceituno, PhD

Although one of the purposes is to combine drugs at fixed dose ratios to simplify treatment of chronic diseases and improve patient adherence, there is a general consensus that this rationale cannot be the only goal behind any development or formulation design [83]. An overview regarding regulations for filing FDC products throughout various jurisdictions around the world shows that progress on this matter has been rather slow. Overall, the development of FDC products by combining previously approved mono-

products or starting from the co-formulation of NCEs can follow limited regulatory pathways. Under US regulations the FDCs regulatory fundamentals are described in the Code of Federal Regulations and guidelines that outline the requirements for FDC product approval. The introduction of co-development guidance in 2013 reflects the importance of these pharmaceutical products from a regulatory perspective [62]. The guidances describe that drug product efficacy can rely on BE testing if there is no change in dosing or proposed therapeutic indication for a novel FDC or clinical data are required otherwise. FDC products could follow one of the following regulatory pathways: 505 b(1), 505 b(2) or 505 j covering all the possibilities from new development to generic development. On the other hand, EMA launched several guidelines with respect to the clinical development of FDC products reflecting the proposed therapeutic used and indications of any FDC development [63]. The guidance describes three possible situations with specific requirements for demonstrations of efficacy: 1) the use of an FDC product as add-on treatment if there is a deficient response to one or more drugs to be included in the proposed combination. Drug-Drug (DDI) or PK interaction study may be required if the combination poses a threat with potential clinical consequences; 2) substitution by an FDC product when a reduction of pill burden is sought after. Bioequivalence testing is required and special attention should be paid if the FDC product is dosed at different time intervals, and 3) FDC therapy initiation if the FDC product has not been used previously for any particular indication. Both clinical and pk trial, as well as DDI study, should be performed and submitted prior to approval. In Latin America, there is only one specific guidance for registration of FDC products since 2010 [84]. It describes the definition of FDC products, general consideration for filing and regulatory requirements that depend on the proposed dose scheme or the drugs to be combined. FDC approval can be granted under the following conditions: 1) An FDC product contains the same actives, dose and dose regimes as mono products used concomitantly, therefore the safety and efficacy profiles are well known; to demonstrate efficacy, a bioequivalence study may be sufficient; 2) same conditions as in “1”, but FDC product is going to be used in novel dose or new therapeutic indication and therefore a phase III clinical trial is required; 3) the combination contains one or more new active ingredients and phase I, II and III clinical trials are required to gain approval. In general, there is not a globally applicable guideline for

FDC product registration, but for specific therapeutic classes and four general cases that are described in a WHO technical report, aiming at guiding pharmaceutical companies for development, approval, and marketing FDC products under less developed jurisdictions [61]. Although generic and hybrid submission pathways seem to be sufficient under most jurisdictions, preclinical and clinical data for novel combinations will always be needed if individual components in FDC products are either known or they are new investigational drugs. However, the idea still persists among regulated entities that different jurisdictions around the world should give more importance to convenience/compliance as a rationale for developing FDC products either containing authorized/new drug entities or authorized drugs only bearing in mind patient´s satisfaction or reduced/contained health costs [85]. If generic development is allowed, a BE study design for a FDC product should consider the same principles as if the drugs were given alone, looking for the achievement of equivalence in PK profiles for each FDC active ingredient and their respective either reference FDC or reference mono products. At this point, it is important to realize that PK interactions may have more critical consequences with FDC products than the same drugs given as mono products concomitantly. To conclude, when comparing jurisdictions to obtain FDC product approval, it seems necessary that a balance should be reached between an overcautious registration approach and the potential large public health benefits that would arise from affordable FDC products of proved efficacy. The achievement of broad harmonization in the understanding and application of existent technical guidelines and requirements for FDC product development and registration is still a pending matter.

Formulation Design, Challenges, and Development Considerations for Fixed Dose Combination (FDC) of Oral Solid Dosage Forms – Divyakant Desai, PhD

For formulation scientists without prior experience of the FDC development, two decision trees were discussed to select the most suitable formulation development strategy. The first decision tree was related to the formulation design for an FDC product (Figure 4).

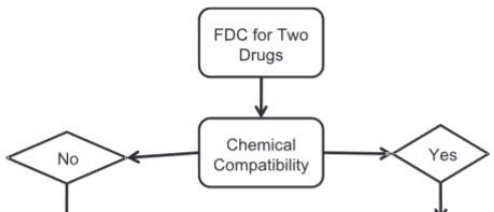

Figure 4: Decision tree for the formulation design of a FDC. Figure adopted from Desai and colleagues [86]. Copyright Taylor and Francis 2013.

If two drugs are chemically incompatible, multi-layer tablet or a drug-specific multi-particulate system was proposed. If they are compatible, then a monolithic system was proposed unless there is a need to keep them apart in order to maintain the dissolution profiles comparable to the respective single entity product. The second decision tree was about the selection of the manufacturing process for an FDC product (Figure 5).

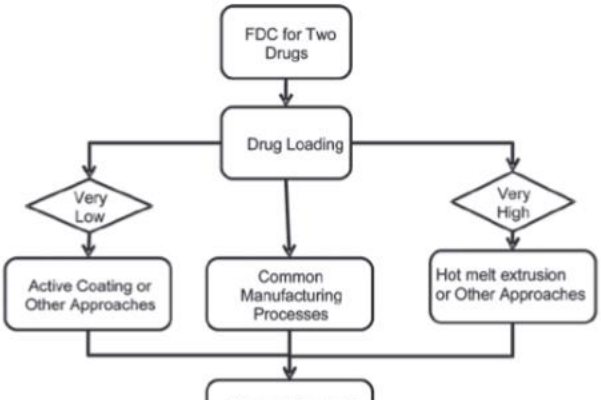

Figure 5: Decision tree for the manufacturing process selection of a FDC. Figure adopted from Desai and colleagues [86]. Copyright Taylor and Francis 2013.

The drug loading in the formulation dictated the selection of the manufacturing process. If the drug loading is high, a hot melt extrusion (HME) or a bi-layer method of manufacturing was proposed. For a formulation with a low drug loading, an active coating approach was proposed. One of the crucial factors in the manufacturing process selection is a pharmaceutical scientist prior experience with the manufacturing process under consideration. A monolithic formulation system, where two drugs are incorporated in a single dose unit, is considered the most simple formulation approach. However, a case study was presented where a second drug, hydrochlorothiazide (HCTZ), was added to the existing formulation of a hypertensive drug [87]. It was shown that povidone (a binder) and poloxamer (a wetting agent) triggered HCTZ degradation

584 under accelerated storage conditions by solubilizing HCTZ in available moisture. Replacement of povidone
585 by Starch 1500, resolved the stability issue and removal of poloxamer did not impact the BE study
586 adversely. For a bi-layer tablet formulation approach, which is normally used to keep two incompatible
587 drugs apart or to maintain two drug release profiles, few critical formulation factors were presented. Those
588 factors include the selection of excipient with high fragmentation tendency such as lactose in the first layer,
589 more deformable material such as microcrystalline cellulose in the second layer, the weight ratio of not
590 more than 1:6 for two layers. It was also emphasized that the tamping force for the first layer should be able
591 to reduce the volume without sacrificing the surface roughness which is essential for the adhesion of the
592 second layer. Two case studies were presented on the bi-layer formulation approach. In the first case study,
593 the compressibility of an extended release metformin formulation was improved by the addition of 1% w/w
594 silicon dioxide. In the second case study, two different grades of fumed silica behaved differently in a bi-
595 layer tablet formulation [88]. Aerosil 200 did not cause layer separation but Aeroperl 300 did. Aeroperl can
596 adsorb relatively large amounts of moisture at any humidity level due to its greater surface area, but it does
597 not retain moisture when the humidity decreases. In contrast, Aerosil adsorbs relatively smaller amounts of
598 moisture but it retains moisture due to its large pore sizes. It was hypothesized that the moisture not retained
599 by Aeroperl could be available for interactions with other layer excipients such crospovidone. The third
600 formulation technique presented was an active coating technology. An active coating can also be used to
601 maintain two separate release profiles and to separate two incompatible drugs. A case study was presented
602 to show how acid and base sensitive molecule was stabilized selecting and minimizing the excipients in a
603 coating material API come in intimate contact with. For example, 1 mg drug is placed with 99 mg of
604 excipients for a 100 mg tablet, the 1 mg drug can react with 99 mg of excipients. However, if 1 mg drug is
605 placed with 9 mg of coating material, the amount of available for a reaction is reduced drastically. It is also
606 a useful technology to make a tablet for a compression sensitive molecule. Although the active coating is
607 useful, it is not as widely used as other technologies because it presents two big challenges. The first
608 challenge is how to detect coating endpoint so that tablets with correct potencies can be manufactured. If a
609 coating process is stopped early, tablets may be sub-potent. On the other hand, if the coating is stopped

late, tablets may be super potent. The second challenge is content uniformity (active coat uniformity). The content uniformity can be influenced by various process parameters such as pan load, coating time, number of coating guns, and spray quality. A mathematical model was presented in which model parameters were linked with the process parameters for scale-up. It was shown that the model correctly predicted coating uniformity of tablet weighing 200 mg to 1450 mg in different shapes at a 450 kg commercial scale. In summary, the decision trees are very useful to explore the most suitable formulation and manufacturing process for an FDC formulation. Each formulation approach for an FDC will have its own unique challenges but as illustrated by various case studies, it is possible to overcome these challenges to develop a rugged formulation and a commercially viable manufacturing process using various process analytical technologies (PAT).

Clinical Pharmacology Aspects of Fixed-Dose Combination Drug Development – Dakshina Murthy Chilukuri, PhD

Combination products are defined in the Code of Federal Regulations [21 CFR 3.2 (e)]as categories of drug-drug combination products. These products could be two or more approved drugs or investigational drug(s) developed along with an approved drug(s) or two or more investigational drugs developed together. The final products can be FDCs, co-packaged products or separate individual products administered together. Among the reasons why these products are developed are the additive/synergistic effects of drugs for the same disease (*e.g.,* anti-viral and cough/cold drug products). Sometimes when two drugs have complementary mechanisms of action they are developed for the same disease as an FDC product. For instance, combining a beta-lactam with a beta-lactamase inhibitor allows for selective killing of bacteria that would otherwise be resistant to the beta-lactam. There are examples of FDCs where one component is included to reduce the adverse events of the other component (e.g. naproxen/esomeprazole delayed-release tablets). Most FDCs are oral but there are examples of inhalational (*e.g.*, tiotropium/olodaterol for Chronic Obstructive Pulmonary Disease, (COPD)) and ophthalmic products (*e.g.,* netarsudil/latanoprost for lowering intraocular pressure). The purpose of this presentation was to provide an overview of the clinical pharmacology considerations in FDC development. FDC development offers interesting challenges to drug

developers. If two or more new molecular entities (NMEs) are being developed as an FDC then dose-finding studies of the drugs are generally required to determine the appropriate dose of each drug to be combined. If the FDC product contains drug component(s) not included in an approved combination therapy, then a factorial design clinical efficacy/safety study may be required to demonstrate the contribution of each drug component. Drug administration challenges such as the effect of food on the FDC will generally need to be addressed. This scenario could get more complicated when the various drugs proposed in the FDCs have different requirements for administration under fed and fasted conditions or when the drugs have different dosing frequency. These scenarios generally require a closer look at the FDC formulation and potential for additional BA studies. Dose adjustments of FDCs in specific populations are potentially problematic given the formulation inflexibility. The typical study conducted as part of the development program of an FDC is a relative BA study. The purpose of the BA study of an FDC is to compare the rate and extent of absorption of each active drug ingredient or therapeutic moiety in the FDC to the rate and extent of absorption of each active drug ingredient or therapeutic moiety administered concurrently as separate, single-ingredient preparations [21 CFR 320.25(g)]. Generally, a two-treatment, single-dose, fasting study of the FDC versus single-ingredient drug products at the highest strength of the combination product with matching doses of individual drug products is recommended [89]. Alternative study designs such as a three-treatment study design comparing the combination drug product versus single-ingredient drug products administered separately may be appropriate. A single-dose, food-effect study on the FDC is usually conducted to evaluate the effect of food on the FDC. Case studies related to BA studies conducted to support approval of FDCs were presented along with examples of FDCs approved based on factorial design studies for the FDCs in comparison versus the individual components administered separately. The FDA guidance entitled "*Codevelopment of Two or More New Investigational Drugs for Use in Combination*" lays out the scenarios where a factorial design study may be appropriate to establish the contribution of the individual components in the FDC.

Concluding Remarks and Future Perspectives

Market access for FDC products is challenging in terms of achieving bioequivalence to co-administration of the individual mono-products, but also because of formulation challenges (compatibility of API's, doses). However, we should not neglect the impact of GI physiology on oral drug behavior which can result in intersubject differences in systemic outcome, potentially leading to failures in bioequivalence studies. Therefore, it's important to finalize a clear link between formulation strategy and clinical evaluation, supported by guidelines of regulatory authorities. In addition, the contribution of *in vitro* predictive dissolution testing can help assist regulatory decisions with respect to the approval of FDC products in a sense that these models identify the underlying GI variables playing a crucial role in the absorption process inside the GI tract. From an academic point of view, these clinically-relevant dissolution models can be optimized and validated when pharmaceutical companies would share their non-BE formulations (*i.e.*, clinical failures). When they do so, the underlying problems can be unraveled which will be taken into account by formulations scientists when formulating FDC products.

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