Use of inspiratory profiles from patients with chronic obstructive pulmonary disease (COPD) to investigate drug delivery uniformity and aerodynamic dose emission of indacaterol from a capsule based dry powder inhaler

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Abbreviations: ACI, Andersen Cascade Impactor; DPIs, Dry powder inhalers; FPD, Fine particle dose; FPF, Fine particle fraction; HPLC, High performance liquid chromatography; PIF, Peak inhalation flow; IT, Inhalation time; Vin, Inhaled volume; LOD, Limit of detection; LOQ, Limit of quantification; MMAD, Mass median aerodynamic diameter; PIL, Patient information leaflets; TRA, Total residual amount; TED, Total emitted dose; TRD, Total recovered dose

ABSTRACT

Most patients using dry powder inhalers (DPIs) are unable to achieve the inhalation parameters recommended for pharmacopoeial in-vitro dose emission testing. The dose emission characteristics of indacaterol Breezhaler (IB) have been measured using COPD patients' inhalation profiles (IPs) when using IB and replayed in-vitro using a breath simulator attached to an Andersen Cascade Impactor. The peak inhalation flow (PIF) of the profiles ranged from 28.3-87.8 L/min and inhaled volumes (Vin) from 0.7-3 L. The indacaterol total emitted doses (TED), fine particle dose (FPD) and mass median aerodynamic diameter (MMAD) were measured. TED varied between 61% to 83% of the 150 µg nominal dose, the FPD was found to vary between 19% and 30% and the MMAD from 3.7 µm to 2.3 µm with the increase of the profiles' PIF and Vin. The mean (SD) values were 113.4(8.9) µg, 39.7(5.0) µg and 2.7(0.5) µm, respectively. The quantity and the quality of the emitted dose from the indacaterol Breezhaler® are dependent on the capability of a patient generating an optimal inhalation profile. Therefore, when using the IB patients should be encouraged to inhale as fast as they can from the start of their inhalation and for as long as possible.

Keywords:

Andersen cascade impactor (ACI), Peak inhalation flow (PIF), Inhalation volume (Vin), Acceleration rate (ACIM), Breath simulator (BRS) and Indacaterol Breezhaler® (IB)

1. Introduction

The number and type of dry powder inhalers (DPIs) is steadily increasing and the trend is for these devices to be more widely used then metered dose inhalers (MDIs). This growth is because they do not contain propellants, their dose emission is breath actuated and overall they are easier to formulate (Haughney et al., 2010; Muralidharan et al., 2015). The quality of the emitted dose from DPIs depends on different factors such as device design and formulation (Chrystyn, 2003), inhaler device's intrinsic resistance (Laube et al., 2011) and the inhalation profile (IP) characteristics of patient inhalations (Azouz et al., 2015a). The patient's inability to achieve the required optimum inhalation effort will result in poor drug delivery to the lungs (Haidl et al., 2016). Each patient IP is composed of 3 inhalation manoeuvre parameters namely, the peak inhalation flow (PIF), the inhaled volume (Vin) and the initial acceleration rate of the inhalation (ACIM) (Bagherisadeghi et al., 2017; Chrystyn et al., 2015; Olsson and Asking, 1994). The IP characteristics depend on the patient's body mass index (BMI), gender, age, disease status and their inspiratory muscle capability (Chrystyn and Price, 2009), which results in a large inter-patient variability of the inhalation manoeuvre parameters when inhaling through the same DPI device. These result in different drug doses delivered to the lungs of patients during routine use (Buttini et al., 2016).

The compendial method for testing the dose emission form DPIs is an in-vitro method using a vacuum pump to generate an inhalation flow corresponding to a pressure difference of 4 kPa in the device and an inhalation volume of 4 L (EP, 2013; USP, 2014). However, most patients are not able to achieve these parameters when inhaling through a DPI device (Al-Showair et al., 2007; Azouz et al., 2015a). Furthermore, the IP generated by the vacuum pump produces a square wave with the instant acceleration of the flow to the set PIF which is maintained until 4L are drawn through the DPI. No human can replicate this type of IP (Copley et al., 2014). In contrast, the IP of the patient is bell shaped with a gradual increase in the

acceleration rate to reach the peak inhalation flow (Chrystyn and Price, 2009). The in-vitro compendial methods, therefore, fail to reliably assess how a DPI would perform during routine use by patients especially those with severely impaired lung function and very severe COPD (Al-Showair et al., 2007; Copley et al., 2014). Recently, researchers have adapted a new methodology to replace the use of the vacuum pump with in-vivo generated patients' IPs that was previously measured when the patient inhaled through the DPI being tested (Bagherisadeghi et al., 2017; Chapman et al., 2011; Chrystyn et al., 2015; Colthorpe et al., 2013; Olsson et al., 2013). The patient's IP is replayed using a breath simulator (BRS) (Olsson et al., 2013). This methodology is more representative than the electronic lung methodology (Brindley et al., 1994). This adapted methodology has opened up the opportunity to provide an insight into the dose that patients inhale during routine use and improve the clinical relevance of in-vitro orally inhaled product (OIP) testing techniques (Copley et al., 2014). This ex-vivo methodology has been used recently to study dose emission and aerodynamic characteristics from different (Bagherisadeghi et al., 2017; Chrystyn et al., 2015) from multidose DPIs formulated with a fixed combination of budesonide and formoterol. Our method uses the Andersen Cascade Impactor (Bagherisadeghi et al., 2017) whereas others use the Next Generation Impactor (Chapman et al., 2011; Chrystyn et al., 2015; Colthorpe et al., 2013; Olsson et al., 2013). Using our method, we have used COPD inhalation profiles, measured when they inhaled through a placebo Breezhaler, to identify the dose emission characteristics of the indacaterol Breezhaler (IB). The use of Patients' IPs to determine the aerodynamic dose emission characteristics from DPIs provide a new insight to the overall dose emission performance of DPIs. The aim of the present study was to examine the impact that real-life IPs obtained from 16 patients with COPD on the dose-emission characteristics of Indacaterol Breezhaler®.

2. Materials and methods

2.1 Patients' inhalation profiles

The IPs of 37 patients with different COPD severity aged 55-79 with a mean age of 66 were recorded using an inhalation profile recorder (Azouz et al., 2015b). Ethics approval was obtained and all patients gave signed informed consent. The profiles were recorded using an empty Onbrez Breezhaler® inhaler device. The patients were given the patient information leaflet (PIL) to read and trained to inhale as hard and fast as they can from the start of the inhalation manoeuvre and for as long as they could. The inspiratory parameters (e.g., PIF, ACIM and Vin) were measured for each profile; the peak inhalation flow (PIF) for each profile is the highest flow the patient can achieve from the start to the finish of the inhalation time. The inhalation volume (Vin) was calculated from the area under the curve of the inhalation flow against the time profile (Azouz et al., 2015b). The acceleration rate (ACIM) was determined by firstly calculating the slope of the flow values against the inhalation time, and then the value with a high linear regression (R^2) was chosen. In the case of Onbrez Breezhaler the linear regression was almost $R^2=1$ for all the values, therefore the acceleration rate was determined as the values with a high slope.

Since the ACI is calibrated to work at a maximum PIF of 90 L/min then profiles with a PIF above 89 L/min were not chosen in this study Their PIF ranged from 2.3 to 151.3 L/min with a mean (SD) of 88.9 (29.5). Twenty patients inhaled greater than 89L/min and of the remainder, the inhalation profiles of 2 were similar so only one was chosen leaving 16 inhalation profiles.

2.2 Experimental set-up

The ex-vivo methodology, shown in Figure 1 has been described by Bagherisadeghi et al. (2017). Flow drawn through the Andersen Cascade Impactor (ACI) (Copley Scientific Ltd, UK), by the Critical Flow Controller (TPK 2000, Copley Scientific Ltd, UK) linked to the vacuum pump (HCP5, Copley Scientific Ltd, UK), and is matched with supplementary air provided by a compressed air supply introduced via the side arm of the mixing inlet (Copley Scientific Ltd, UK). This ensures a constant set flow through the ACI during each dose emission determination. This matched flow when the adult version of the Alberta Idealised Throat (AIT) (Copley Scientific Ltd, UK), with the DPI in situ, is attached to the mixing inlet ensures that there is no flow through the DPI. A breath simulator (BRS 3000; Copley Scientific Ltd, UK) is programmed with the inhalation flow/time data of the measured inhalation manoeuvre. When this programmed profile is played it draws this from the supplementary air. Since the vacuum pump is set to maintain a constant flow through the ACI then the inhalation flow profile is subsequently drawn through the DPI. The flow through the ACI and provided by the supplementary air source is set at either 60 or 90 L/min so that when using the appropriate ACI stages the cut-off diameters are not changed. Also, this means that an inhalation profiles with a PIF above 90 L/min cannot be used.

The Onbrez Breezhaler (Novartis, CH) was the DPI used in this study. For each inhalation profile, 3 capsules (nominal containing indacaterol maleate equivalent to 150µg of indacaterol) were separately aerosolised into the ACI and three separate dose emission determinations were made (n=3). Amounts deposited on each stage of the ACI, the final filter, the pre-separator, the mixing inlet and in the AIT were measured using a validated High Performance Liquid Chromatography (HPLC) method. The total emitted dose (TED), fine particle dose (FPD) (which represent particles with an aerodynamic diameter $\leq$ 5µm), the mass median aerodynamic diameter (MMAD) and geometric standard deviation (GSD) were

obtained. After each dose had been emitted the amount left in the capsule and left in the device were also measured using the validated HPLC method to identify the total residual amount (TRA)

2.3 HPLC assay

The HPLC method (Table 1) used was validated using Dexamethasone as an internal standard. The limit of quantification and limit of detection for indacaterol were 0.17 and 0.07 µg/mL respectively.

2.4 Data Analysis

The Copley Inhaler Testing Data Analysis Software (CITDAS version 2.0, Copley Scientific Ltd, UK)) was used to calculate the aerodynamic dose emission parameters.

The TED was obtained from the cumulative amounts deposited in the Alberta throat (AIT), the pre-separator (PS) and all the stages of the ACI. The FPD was the mass associated with particles < 5 µm and expressed as a % of the label claim. The fine particle fraction (FPF) was the FPD divided by the TED. The MMAD was the size corresponding to the 50th percentile of the cumulative mass-weighed distribution of the amount deposited in the ACI. The total recovered dose (TRD) was calculated as the sum of the total emitted dose (TED) and the total residual amount (TRA).

SPSS version 16.0 software (SPSS Inc., Chicago, USA) was used for the statistical analysis. A two-way analysis of variance (ANOVA) was used to determine any significant differences between the TED, FPD, %FPF, MMAD between different inhalation PIFs at the same Vin and also between different Vins at the same PIFs.

3. Results

The 16 selected inhalation profiles are presented in Figure 2 and the inhalation parameters of these 16 COPD patients together with their demographic details and lung function are shown in Table 2. The mean (SD) PIF, Vin, ACIM and Ti were 64.9 L/min (18.9), 2.1 L (0.7), 4.2 L/s^2 (1.30) and 3.4s (1.0) respectively (Table 2). A summary of the indacaterol aerodynamic dose particle size distribution and the dose emission of each IP is shown in Table 3. The results are arranged in ascending order according to the profile's PIF and the effect of each profile on the TED, FPD and MMAD of inhaled indacaterol are shown in Figures 3, 4 and 5 respectively. This table and the figures show that the TED (as % of 150 µg indacaterol nominal dose) ranged from 61% to 83% with a mean (SD) of 75.6 % (5.9) (Figure 3). The fine particle dose (FPD) of indacaterol ranged from 19% to 30% (of the nominal dose) with a mean (SD) of 26.5 % (3.3) (Figure 4). Figure 5 highlights a marked reduction in the indacaterol MMAD from 3.7 µm to 2.3 µm with the increase in the PIF of the profiles. Figure 6 shows that the total residual amount (TRA) decreased as the PIF of the patient profiles increased which is consistent with the increase in the TED for the profiles that are shown in Figure 3. The TED, FPD and %FPF increased significantly ($p < 0.05$) when the PIF of the IPs increased. In contrast, TRA and MMAD showed a significant decrease with the PIF of the IPs.

4. Discussion

The dose emission and aerodynamic particle size distributions from the indacaterol Breezhaler were found to be related to the peak inhalation flow of the patient inhalation profiles. The influence of the inhaled volume and the inhalation acceleration of the inhalation manoeuvre cannot be identified from these results. Profile 1 was the slowest profile and also the lowest inhaled volume and slowest acceleration whereas profile 16 had the fastest PIF, the largest volume and one of the fastest acceleration. Nevertheless, the results suggest the

flow dependent dose emission of this product which is consistent with this phenomenon demonstrated by all DPIs (Azouz and Chrystyn, 2012).

Figure 1 describes the ex-vivo methodology to generate the dose emission data for indacaterol from an Onbrez Breezhaler. The recorded COPD patient inhalation profiles in-vivo were replayed by the breath simulator (BRS 3000) to aerosolise the indacaterol 150 µg from an Onbrez Breezhaler into an Andersen cascade impactor. The results reflect the dose that patient would have received during real life, routine use of their DPI (Chrystyn et al., 2015). The current method set-up used in this study is similar to that reported by (Bagherisadeghi et al., 2017). The standard USP induction port is deployed routinely for aerodynamic particle size distribution although there is a widespread recognition that the later fails to accurately reflect deposition behaviour in the upper airway of adults (Zhou et al., 2011). Thus, the Alberta Idealised Throat was used in place of the USP inhalation port to ensure a more realistic representation of the upper airway.

The benefits of using the ACI over NGI when studying the aerodynamic characteristics of inhaler devices are that conversion kits (stages and pre-separator) are available for the operation of the ACI at different flow rates, namely 28.3, 60 or 90 L/min. Whereas with the NGI, the change in the applied flow rate is accompanied with an adjustment to the cut-off diameter of the stages. Thus, using an ACI enables a direct comparison between the amounts of inhaled drug particles deposited on each stage. Furthermore, the internal dead volume of the ACI (1.155 L) is smaller than the NGI (2.025 L) and the use of the ACI enables the determination of low volume profiles (Mohammed et al., 2012). Therefore, our study was carried out using ACI instead of NGI to investigate the overall dose emission performance of Indacaterol Breezhaler® using COPD patients' IPs. The Breezhaler® is a low resistance device (0.017 $kPa^{0.5}$ min/L) and an inspiratory flow rate exceeding 100 L/min is required to generate 4 kPa pressure drop through this device (Dal Negro, 2015). The recorded patients' IPs (Table 2

and Figure 2) show that most of the COPD patients are not able to generate a pressure drop corresponding to 4kPa and an inhaled volume of 4 L recommended by the pharmacopoeia (USP, 2014) when they inhaled through Breezhaler® device.

DPI formulation de-aggregation, dispersion and emission are dependent on the inhalation manoeuvre characteristics PIF, Vin and ACIM (Bagherisadeghi et al., 2017; Chrystyn et al., 2015). The inspiratory effort generated by the patient during the inhalation manoeuvre creates a pressure drop inside the device that releases the dose from the DPI and de-aggregates the formulation. This pressure drop is directly related to the inspiratory effort generated by the patient (Clark and Hollingworth, 1993). DPIs are breath actuated devices and it has been reported that peak inhalation flow rate (PIF) has an effect on the dose emission as well as lung deposition from DPIs, especially low resistance devices such as Aerolizer® and Breezhaler® where dose emission is mainly dependent on the generated PIF (Alaboud, 2011; Chew and Chan, 2001; Colthorpe et al., 2013; Nielsen et al., 1997).

In the present study, the inspiratory inhalation manoeuvre parameters: PIF, Vin and ACIM have shown an effect on the dose emission as well as on the aerodynamic particle size distribution of indacaterol Breezhaler®. However, the impact of the PIF was more noticeable when compared to the effect of ACIM and Vin. When using IPs, all inhalation profile parameters act together and it is difficult to know the extent of the contribution of each inhalation manoeuvre parameter with regard to the TED and APSD. Increasing the PIF have resulted in a significant ($p < 0.05$) increase in TED from 92.0 (4.2) µg to 125.1 (1.4) µg and FPD from 28.7 (1.6) µg to 45.6 (0.8) µg of indacaterol 150 µg dose (Figures 3 and 4). In contrast, the MMAD and TRA decreased significantly ($p < 0.05$) with increasing the PIF (Figures 5 and 6). Suggesting that increasing the PIF of the profiles has led to a subsequent increase in the turbulent energy inside the device resulting in a more dose of indacaterol

being delivered from the Onbrez Breezhaler®. The present results demonstrate the flow rate dependency of Onbrez Breezhaler® as reported in previous studies (Abadelah et al., 2017a; Pavkov et al., 2010). Furthermore, the particles dispersion of the inhaled bolus is obvious from the significant ($p < 0.05$) reduction in the MMAD of all profiles ranging from 3.7 µm to 2.3 µm (Figure 5).

The results of our study of indacaterol Breezhaler® are in line with the findings of previous studies using glycopyrronium Breezhaler® (Chapman et al., 2011; Colthorpe et al., 2013). Breezhaler® is a low resistance device and patients were able to achieve an average PIF of 72 L/min (Chapman et al., 2011) and 70 L/min (Colthorpe et al., 2013). The previous studies showed that Breezhaler® was producing higher FPD and also preferred by patients with a range of disease severity, Chapman et al., 2011 reported a mean of 26.8 (5.8) FPD (% nominal dose) for Breezhaler comparing to less than 10% FPD for Handihaler®. Our results showed a mean (SD) of FPD of 26.4 (3.3) (% nominal dose) with an MMAD of 2.7 (0.5).

Drug retention inside the inhaler continues to be a factor plaguing the performance of novel inhalers (Tajber et al., 2009). Drug retention varies between inhaler devices in that some studies have reported between 30 and 50% of the nominal dose being retained within the device. It is important that the complete dose is released from the inhaler so as to maximise the therapeutic effect, minimising drug wastage and avoiding potential dosage errors during the next inhalation (Abadelah et al., 2017b). The TRA is reduced from 45% to 11% of the total nominal dose for profile 1 (PIF (28.3 L/min), Vin (0.7 L), ACIM (1.9 L/sec^2) and 16 (PIF (87.8 L/min), Vin (3 L), ACIM (5.1 L/sec^2) respectively (Table 3), suggesting that all inhalation manoeuvre parameters were involved in device and capsule emptying, supporting our previous study with the Easyhaler (Abadelah et al., 2017b). Irrespective of the PIF, Vin and ACIM for all profiles used, there was always some indacaterol retained in the capsule/

device (Table 3) suggesting that other factors are also involved in dosage emptying such as the length of device inhalation channel and the nature of the coating material inside the device. For instance, the Easyhaler has a short inhalation channel and reported to have less residual amount compared to a device with longer inhalation channel such as the Aerolizer® (Azouz and Chrystyn, 2012; Coates et al., 2004). Our unpublished data with Cyclohaler and Handihaler followed the same trend observed with the Easyhaler and Aerolizer (Azouz and Chrystyn, 2012)

5. Conclusion

This ex-vivo methodology provides a more realistic representation to the way patients use their inhalers and the type and quality of the dose they would receive in real life use. The combined inspiratory inhalation parameters: PIF, Vin and ACIM showed an effect on the dose emission of indacaterol from Onbrez Breezhaler. Optimal profiles with the highest PIFs, provided the highest TED, FPD, the lowest MMAD and RA. Thus, the quantity and the quality of the emitted dose can be achieved by inhaling hard and fast from the start of the inhalation manoeuvre and for as long as patient possibly could to empty the dose and to maximise drug delivery to the lungs from Onbrez Breezhaler.

6. Conflict of interest:

Henry Chrystyn has no shares in any pharmaceutical companies. He has received sponsorship to carry out studies, together with Board Membership, consultant agreements and honoraria for presentation, from several pharmaceutical companies that market inhaled products. These include Almirall, AstraZeneca, Boehringer Ingelheim, Chiesi, GlaxoSmithKline, Innovata Biomed, Meda, Napp Pharmaceuticals, Mundipharma, NorPharma, Norvartis, Orion, Sanofi, Teva, Truddell Medical International, UCB and Zentiva. Research sponsorship has also been received from grant awarding bodies (EPSRC and MRC). He is the owner of Inhalation Consultancy Ltd. He is also a consultant of Research in Real Life, which is subcontracted by Observational and Pragmatic Research Institute Pte Ltd.

The authors declare no conflicts of interest in this work.

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