CHEMICALLY-SPECIFIC CHARACTERIZATION OF PARTICULATE IN INHALABLE DRUG PRODUCTS USING RAMAN AND LASER-INDUCED BREAKDOWN SPECTROSCOPY

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SUMMARY

The impact of foreign particulate and agglomerates on dissolution profile and permeability of the inhalable drug particles within the sites of action (i.e. patient's lungs) is critical. In this study, Raman Chemical Imaging (RCI) and Raman/ Laser-induced breakdown spectroscopy (LIBS) were used to investigate the Active Pharmaceutical Ingredients (API) Particle Size Distribution (PSD), degree of particles aggregation and presence of foreign particulate in commercial inhalable products. Automated Raman/LIBS analysis was used to investigative the contamination issues and to establish the size and chemical makeup (metallic, organic or inorganic) of foreign particulates. The particulate population results obtained by Raman/LIBS analysis are shown to be advantageous in an investigation of non-conformance/safety of the inhalable products.

INTRODUCTION

Since the approval of the dry powder inhalers (DPI) containing fluticasone propionate (FP) and salmeterol xinafoate (SX) in 1999, combination inhaled therapies dominated COPD market. Due to the lack of specific guidance of the Office of Generic Drugs (OGD) as to the Abbreviated New Drug Application (ANDA) submission there was very little attempts of generic manufacturers to come up with bioequivalent DPI formulations. Success of an ANDA submission to OGD as it is stated in recently published draft Guidance on FP and SX [2] depends on ability of the manufacturer to prove that their test product is qualitatively and quantitatively the same as the reference product, as it relates to both the formulation and the device. Accurate and precise data on the PSDs of the individual ingredient and possible contaminants is required for the drug product submission. Standard methods such as conventional cascade impaction (without the use of chemically-specific assay procedure) and optical microscopy are not chemically-specific, thus cannot be applied for combinational product PSD analysis.

Any differences in the PSD of each of the active ingredients and foreign particulate will almost certainly result in differences in deposition, preventing co-deposition of each active in the correct ratio in each region of the lung. Even if the drugs PSDs were identical, final formulations may be affected by numerous mechanical and environmental factors and contaminations. External factors that may cause imbalances/variation on deposition/ inefficacy due to the contribution of the excipients used (i.e. the grade/size of milled lactose which often is acted as a carrier). One active component of a dry powder formulation may have more surface energy or may be more hygroscopic than the other, leading to differences in how each detaches from the carrier or aggregates with other particles.

Because the interactions of the various components depend largely on the surface energies of the particles, some researchers have attempted to gain a better understanding of DPIs aerosolization behavior by using the colloid probe method of Atomic Force Microscopy (AFM) [3]. Such analysis to determine the Cohesive-adhesive Balance (CAB) between each of the components of a dry powder formulation allows direct quantification of the forces of interaction between API and other ingredients, however it may be time consuming and expensive.

RCI and automated Raman/LIBS analysis are shown to have a potential to address the need to accurately identify and size not only primary drug particles but also aggregates and foreign particulate based on their unique spectroscopic properties. With the ability to detect smallest spectral shifts the method is sensitive enough to identify polymorphic forms of the drugs, which is important because different polymorphs/hydrates have different dissolution rates, and the presence of particular polymorphs of the API or specific grade of the lactose (which is used as a carrier) in the formulation can therefore affect product bioavailability.

Mechanisms to create drug particles of desired physico-chemical properties (i.e. engineered particles) as well as the device effect on drug particles de-agglomeration were successfully investigated by this high fidelity imaging method without time-consuming cascade impaction analysis [4].

EXPERIMENTAL METHODS

The FALCON II Wide-Field Raman Chemical Imaging system (ChemImage Corporation, Pittsburgh, PA) combines spectral information gathered by Raman spectroscopy with morphological information obtained by optical imaging. The RCI microscope collects data from hundreds of randomly selected Fields Of View (FOVs) from across each sample. The system then overlays the spectral data with an optical image of the sample, producing a montage of data correlated on a pixel-by-pixel basis that clearly shows which particles consist of excipients or any of the active ingredients. RCI method [5, 6] allows extraction of a Raman spectrum from every pixel in the image (spatial resolution approaching 350 nm can be achieved). Thus every image provides spectral and spatial information about any drug to drug or drug to excipient agglomerates that may form [7]. The population statistics obtained from RCI on adhered particles were correlated to CAB data for analysis.

The SPE-Is raman.id+metal.ID system (RapID, Berlin, Germany) was used to establish the size and chemical identification of foreign particulate. Particle count, size and shape were obtained from image analysis whereas particulate chemical indentify was obtained by using high throughput automated spectroscopic analysis. A spectral library of known contaminants was also used in this analysis. This library provided greater accuracy and assurance in the investigation of the source of the contamination.

RESULTS & DISCUSSION

RCI method with conjunction to CAB approach to colloid probe AFM was applied to investigate the microstructure and product aerosolization performance of commercial Seretide® Accuhaler®. Assessment of free-standing API particles (including same drug agglomerates) and bound to lactose API particles (including different drugs agglomerates) wasd done on the overlayed/fused BFR/RCI images. By RCI/AFM analysis it was possible to establish the link between the microstructure and the dispersive mechanism of the formulation. For example, comparison of the CAB ratio of FP with respect to lactose and SX with respect to lactose suggested that the adhesion of FP to lactose is significantly ($p<0.05$) greater than SX to lactose. The cohesive salmeterol xinafoate interactions are 1.85 times greater than the adhesive SX-FP interactions, whilst the FP-SX interactions were 1.2 times greater than the cohesive FP-FP interactions. RCI analysis of particles deposited on the NGI suggested that the presence of finer free standing SX than FP and the presence of FP-lactose agglomerates vary (Table 1), which allows correlating adhesion properties of the particles with extend of different types of agglomerates (Figure 1). These differences are related to the surface interfacial properties of the components of the formulation as suggested by CAB-AFM analysis.

API	Free Standing (% of total detected)	Bound to Lactose (% of total detected)	Bound to other API (% of total detected)
Fluticasone propionate	46%	20%	2%
Salmeterol xinafoate	50%	10%	5%

Table 1 - Statistics of free standing versus bound to carrier API particles.

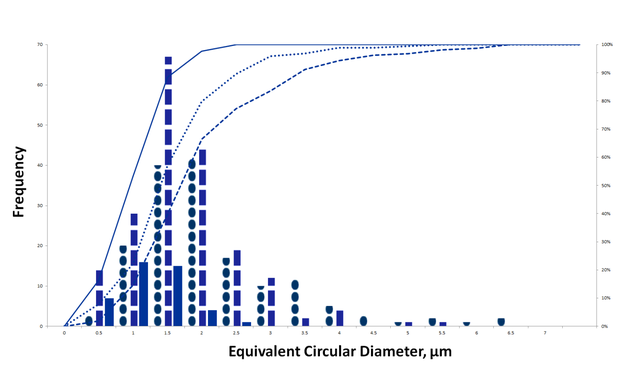

Figure 1 - PSD histogram and cumulative percentage for: fluticasone propionate (dashed line); lactose (dotted line); salmeterol xinafoate (solid line).

RCI and agglomerate-identification image processing analysis was successfully used to differentiate stand-alone fluticasone propionate particles from co-associated fluticasone propionate particles in Flixotide MDI to evaluate product delivery profile of this combinational product (Tables 2-5).

Sample	API Agglomerates (% of total API particles)	D10 (µm)	D50 (µm)	D90 (µm)	Std. Dev. (µm)	Min. (µm)	Max. (µm)
Flixotide 125	9 (26%)	4.8	6.2	8.3	1.5	4.3	9.2

Table 2 - Fluticasone propionate API-API agglomerates in sample based on Equivalent Circular Diameter.

Sample	< 1 (µm)	1-2 (µm)	2-3 (µm)	3-4 (µm)	4-5 (µm)	> 5 (µm)
Flixotide 125	0	0	0	0	2	7

Table 3 - Fluticasone propionate API-API agglomerates size distribution in sample based on Equivalent Circular Diameter.

API	Free Standing (% of total detected)	Agglomerated API (% of total detected)
Fluticasone propionate	74%	26%

Table 4 - Statistics of free standing versus agglomerated API particles (Figure 7).

RCI can easily separate different chemical species or polymorphic/hydrate forms of the same chemical within single particle or aggregate for identification of polymorphic impurity/unwanted phase. Thus, understanding which specific microstructure aids better dispersion of the different drugs using different mechanisms can benefit inhalable drug development on the very early stages. Raman/LIBS analysis provides information about quantity/chemical ID of any of the foreign particles. For example, a cellulose particle would be identified based on morphological parameters (optical image) and associated spectrum and the dimensions of this foreign particle would be reported (Figure 2).

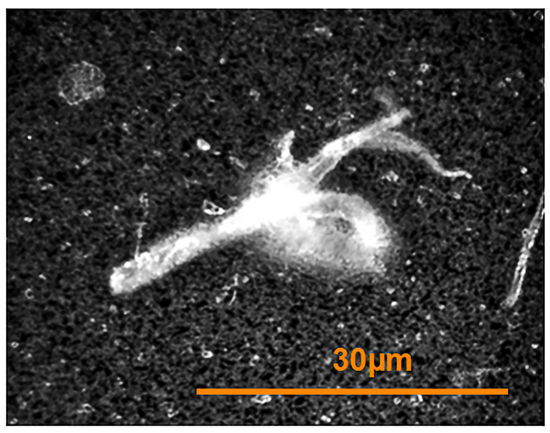

Figure 2 – Foreign particulate material identified in a DPI product. Particle ID: Cellulose (Paper), Rank: 882,S/N: 39.2, Length: 30.6µm; Class: 4, L/W: 2.3.

CONCLUSIONS

Utilizing combined Raman/LIBS as a method for in-depth particle characterization within inhalable drug products has shown to be beneficial for differentiating particles by their chemical make-up, acquiring particle size information, and evaluating the degree of drug-specific aggregates. Therefore, it can be concluded that by using these analytical methods in conjunction with one another, it is possible to identify the presence of foreign particulates in the formulations. Such analysis may aid in understanding dispersion of the drug particles and inhalation product aerosolization performance without time-consuming impaction analysis testing or AFM analysis.

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