Evaluation of Different Particle Size Reduction Techniques in Application of Formulation Preparation

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ABSTRACT--- ***Several commonly used particle size reduction techniques were evaluated in this study. Two excipients Avicel PH 101, calcium phosphate dihydrate and a poorly soluble compound naproxen were used for evaluation. All the techniques tested in this study: tissue grinder, glass beads, Glen Mills high speed mixer, polytron and sonication showed different degree of particle size reduction. The polytron in combination with the sonicator proved to be the most suitable technique for particle size reduction in discovery with limited API available. The processed particle sizes from the polytron technique were comparable to the glass tissue grinders while minimizing the loss of sample. Glass beads created small particle sizes (under 20 microns) depending on the length of time, but it was overly time consuming. Glen-mill needs about hundreds of milligram material to work with despite the comparable particle size reduction.***

Keywords--- Particle size, size reduction, Avicel PH 101, Naproxen, Dibasic calcium phosphate dehydrate

Abbreviation API: Active Pharmaceutical Ingredient, BCS: Biopharmaceutical Classification System, MCT: 0.5% Methylcellulose and 0.2% Polysorbate 80

1. INTRODUCTION

Particle size and size distribution are important parameters for oral solid dosage form development, since they have significant impact on dissolution rate, and subsequently bioavailability, particularly for BCS class II compounds[1-6]. For early drug discovery stage, majority of the compounds are BCS class II. In order to maximize oral exposure and minimize the variation of bioavailability of active pharmaceutical ingredient (API), particle size reduction is critical to control the particle size of the formulations for drug discovery support. Many particle size reduction techniques have been used in pharmaceutical field; common techniques include a variety of milling methods (jet-mill, ball-mill, and hammer-mill), sonication, homogenization, grinding, and glass beads.[6]

The purpose of this study was to investigate several common particle size reduction techniques and to identify the most suitable method in discovery stage when API supply is limited. The techniques evaluated include homogenization, sonication, and the use of tissue grinders, glass beads, and a high speed mixer mill. Besides final particle size and distribution after size reduction by using different techniques, API needs and time consumption are the other two key parameters taking into consideration.

2. MATERIALS AND METHODS

Materials

Three different compounds were used in the assessment of the available techniques. Avicel pH101 microcrystalline cellulose was obtained from FMC Biopolymer, Philadelphia, PA and Dibasic Calcium Phosphate Dihydrate was a free sample from JRS Pharma, Patterson, NY. Both of them are commonly used excipients in solid dosage form. Naproxen or (+)-2-(6-Methoxy-2-naphthyl)-propionic acid was purchased from TCI America, Wellesley Hills, MA.

Methods

Techniques for Particle Size Reduction

The common techniques evaluated in this study included sonication (VWR water bath sonicator 150D, VWR), glass beads(0.5 mm to 3mmm, Corning Inc.), Polytron homogenization (Polytron PT 1200 E, Kinematica), grinding using glass tissue grinder (2ml, 7ml, 24ml, 40ml, Corning Inc.) and plastic tissue grinder (15ml, 35ml, 50ml, Fisher Scientific), and ball milling (Gen Mill High speed mixer model MM301 and 3mm zirconium oxide beads, GlenMills Inc.). Combination of different techniques was also assessed in order to maximize the efficiency of size reduction.

Samples Preparation

Formulations for nonclinical PK, efficacy and toxicology, suspensions are the commonly used oral formulations especially for high doses. In this study, the majority samples were prepared in suspension. Avicel PH 101 and calcium phosphate dihydrate were simply suspended in Milli-Q distilled water and naproxen was suspended in 0.5% Methycellulose and 0.2% Polysorbate 80(MCT) due to the poor wettability of this compound. Various concentrations of samples ranging from 5mg/ml to 100mg/ml were investigated depending on the techniques. Dry samples were also used for glass beads and Glen mill (ball mill) techniques.

Particle Size Measurement

The Malvern Mastersizer 2000 was used to analyze the particle size and size distributions of treated samples. The particle size and distribution was based on volume in this study, which is more relevant to the weight of compounds in a formulation. The particle size values were the average of the triplets or more. d10 (10 percent of the particles based on volume is less than the value), d50 (50 percent of particles based on volume is less than the value) and d90 (10 percent of the particles based on volume is less than the value) are used to report particle size data in the results.

3. RESULTS AND DISCUSSION

Tissue grinder

Both glass and disposable plastic tissue grinders with different sizes were used to evaluate the effectiveness of this technique. Avicel PH 101 was used as a model compound to compare different tissue grinders. The original size of Avicel PH 101 with d50 of 60um, after grinding by using different tissue grinders, the size can be reduced to d50 of 13 to 30 um, depending on the size and type of the tissue grinder (Figure 1). Concentrations of 5mg/ml, 10mg/ml and 20mg/ml of Avicel PH 101 were tested with and without sonication, the average particle sizes were based on all the measurements.

The glass tissue grinders were more effective compared to plastic tissue grinder. The difference is due to the design of the plastic tissue grinder. Instead of grinding the compound with vertical motion, the plastic tissue grinders utilize a rotational motion by turning the handle like a key, which results in lower shear forces and contact surface area, therefore less reduction of particle size. The sizes of the tissue grinder also affect the efficiency of particle size reduction. The 24mL one was the most effective glass tissue grinder among all the tissue grinders tested. The 40mL glass tissue grinder showed comparable particle size reduction compared to 24mL glass tissue grinder, but it is more difficult to operate due to the suction and friction of the larger tissue grinders, which also gave a wider standard deviation (Figure 1).

In order to investigate more on compound concentrations impact to particle size reduction, dibasic calcium phosphate dihydrate was also used in this study. Besides the size of tissue grinders, the concentration of the compounds also contributed to the effectiveness of size reduction (Figure 2). Three sizes (2mL, 7mL and 24mL) of glass tissue grinders were tested, the 40mg/ml dibasic calcium phosphate dihydrate showed smaller particle sizes, especially for d50 and d90 in comparison with those of 20mg/ml for all three glass tissue grinders. The 24mL glass tissue grinders worked the best among the three tested glass tissue grinders for both concentrations of this compound similar to Avicel pH 101. The particle size could also further reduced by sonication for both compounds if needed.

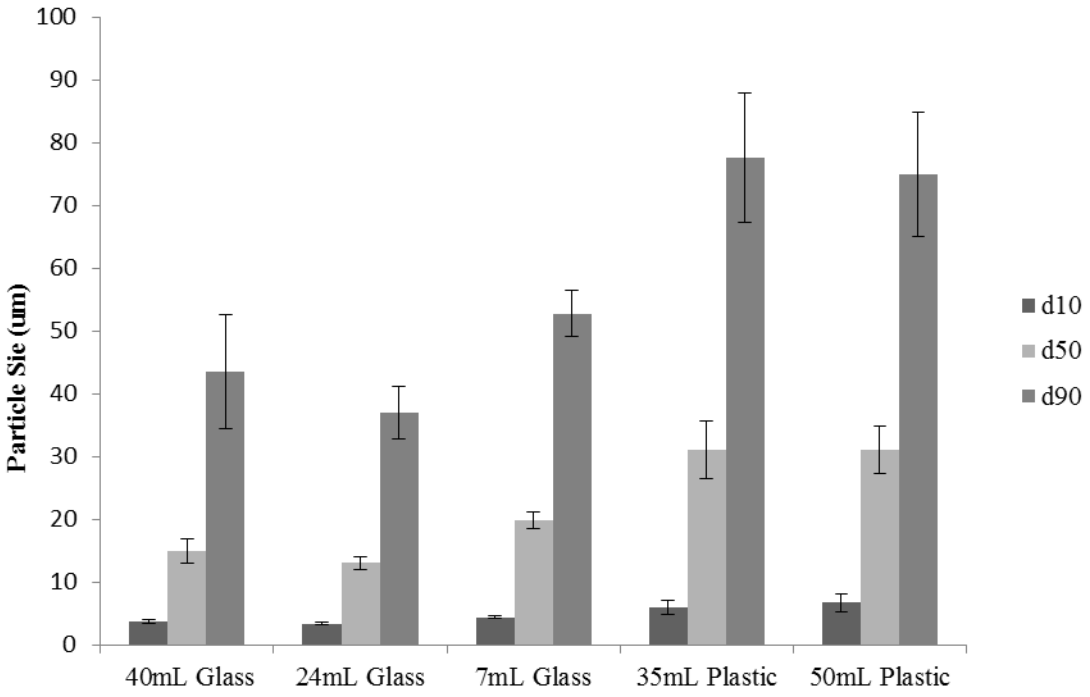

Figure 1: Particle size reduction of Avicel PH101 by tissue grinders

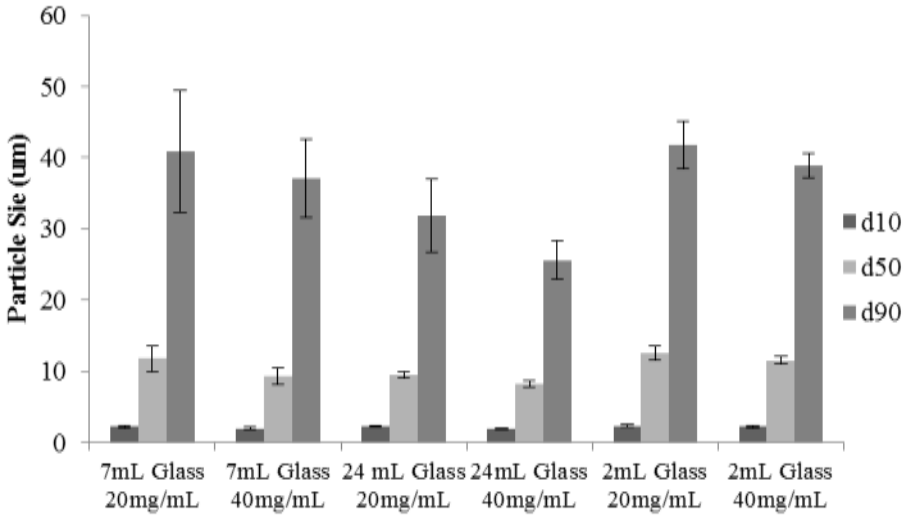

Figure 2: Particle size reduction of dibasic calcium phosphate by glass tissue grinders

Glass Beads

The glass beads technique was the most effective one in terms of small volume of samples and it produced the smallest particle size in this study. Both dry powder and suspension were tested by this technique, the glass beads work better on suspension than dry powder. The vehicle in the suspension, which facilitated the movement of the glass beads, produced more collisions between beads and the compound and thus bringing down the particle size (Figure 3). The particle size of Avicel pH 101 was reduced by two fold if treated with glass beads as dry powder and by almost ten folds if stating as a suspension. The concentration also played a role; 10mg/ml was the optimal concentration among the tested three concentrations. The data used in Figure 3 were four hour treatment, which particle size reached plateau. It suggests that the ratio of glass beads and the total volume of the particles play an important role of size reduction efficiency. For Avicel PH 101, 10mg/ml may give the best ratio of the glass beads added to the total volume of the compound, which provides the most efficient contact and mixing of glass beads and Avicel PH 101. The mechanical property is another factor may impact the size reduction. [7-8]

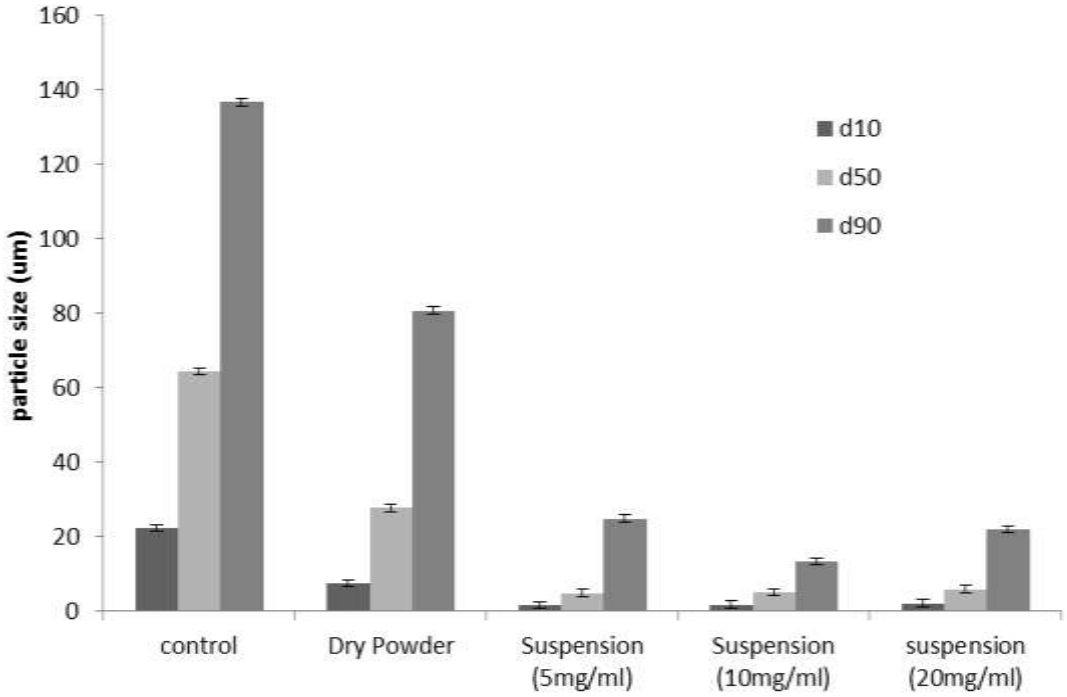

Figure 3: Particle size reduction of Avicel PH101 by glass beads

Glen-mills High Speed Mixer

A high speed mixer type MM301 is a laboratory scale ball mill. The grinding jars of the MM301 perform radial oscillations in a horizontal position. The inertia of the grinding balls causes them to impact with high energy on the sample material at the rounded ends of the grinding jars and pulverizes it. Also, the movement of the grinding jars help bending and mixing of the powder and contact of powder with the grinding balls.

This technique turns out to be an efficient size reduction method if the API can be provided as hundreds of mg. The particle size of Avicel PH 101 with 100mg/ml suspension in water can be reduced about four fold with d 90 of 30 um (Figure 4). The treated time was found to affect the effectiveness of particle size reduction by high speed Glen-mills. With the longer treatment within four hours, the smaller of the final particle sizes was generated. However, the particle size reached the plateau around four hours (Figure 4).

Similar to glass beads, Mixer MM 301 works better on suspension in terms of particle size reduction compared to dry powder (unpublished data, Ran)

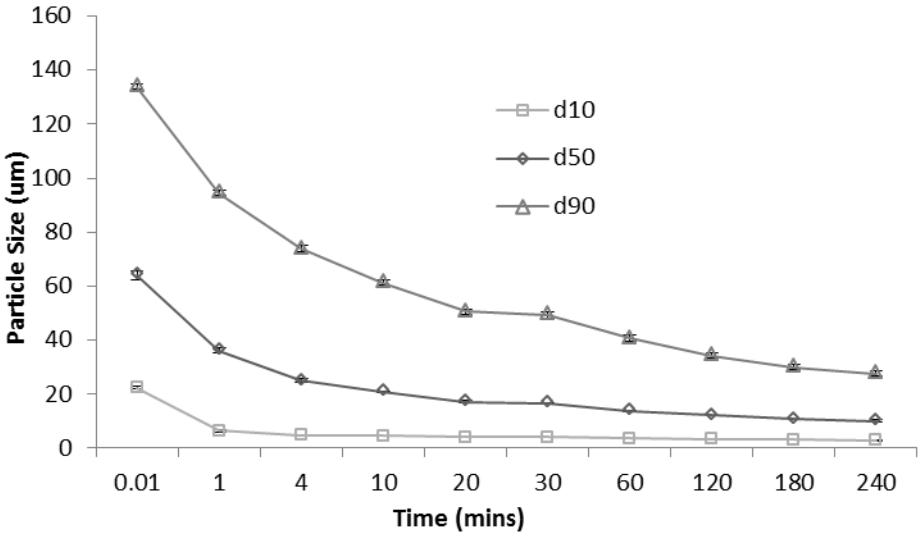

Figure 4. Particle size of Avicel PH 101 (100mg/ml) vs time of milling by Glen-Mill

Polytron

Polytron homogenizer has been used quite often in formulation preparation to homogenize the suspension and reduce the particle size. Polytron handheld generators work via rotor/stator system, in which samples are drawn into the center of the generator tip. The sample is vigorously mixed, accelerated, and processed through the narrow gap between the rotor and stator and finally forced out between the teeth of the stator. Two compounds Avicel PH 101 and calcium phosphate dihydrate were tested in this study by homogenization with both stainless steel and disposable generators of Polytron PT 1200E.

Table 1. Particle size of Avicel PH 101 and dibasic calcium phosphate by homogenization of Polytron

Sample name	d10 (um)	d50 (um)	d90 (um)
Avicel PH 101 control	22.53	64.72	137.15
5 mg/ml Avicel PH 101, Polytron with metal probe	5.00	26.45	69.89
10mg/ml Avicel PH 101, Polytron with metal probe	5.01	26.48	69.64
20mg/ml Avicel PH 101, Polytron with metal probe	4.80	25.22	67.85
5 mg/ml Avicel PH 101, Polytron with disposable probe	5.41	25.72	59.73
10mg/ml Avicel PH 101, Polytron with disposable probe	5.60	26.17	60.10
20mg/ml Avicel PH 101, Polytron with disposable probe	4.66	22.14	51.99
Dibasic calcium phosphate dihydrate control	9.06	135.88	287.41
20mg/ml dibasic calcium phosphate dihydrate, Polytron with metal probe	5.24	35.25	105.26
20mg/ml dibasic calcium phosphate dihydrate, Polytron with disposable probe	4.80	19.96	46.34

Table 1 shows the results of size reduction of Avicel PH 101 and dibasic calcium phosphate dihydrate. Three concentrations (5, 10, and 20 mg/ml) of Avicel PH 101 were used for both generators with no significant difference observed. After three to ten minutes of homogenization, all the suspensions of Avicel PH 101 showed a similar degree of size reduction regardless of the concentration and generator. The plastic probe tip seems work better to minimize the particle size of dibasic calcium phosphate dihydrate, particularly for the particles larger than 100um of d90..

Sonicator

Sonication is the easiest method for particle size reduction. Both probe sonicator and water bath sonicator are applicable for size reduction of suspension. Due to the extreme local heat that may be produced by a probe sonicator, a water bath sonicator is generally preferred in the process of formulation preparation. The power intensity limitation of a water bath sonicator allows more effective particle size reduction in combination with other techniques like polytron

homogenization. Naproxen, a typical Class II poor water soluble compound, can be reduced to a limited level of less than two fold compared to original particle size by applying Polytron alone, which may be contributed to Naproxen's poor wettability and moderate elasticity. By combining the sonication step with Polytron homogenization, the particle size can be further reduced about 3 fold for d10, d50 and d90 compared to applying Polytron alone (Figure 5).

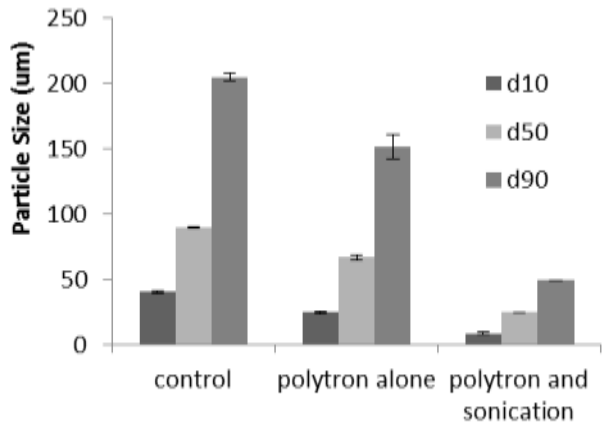

Figure 5. Particle size reduction of naproxen by combination of polytron and sonication.

Summary

It can be concluded that all the evaluated techniques can reduce particle size by a significant amount. The effectiveness is also affected by the properties of the compound and the time a particular method is applied. Keeping ease of use, consumption of time, and efficiency in mind, it can be affirmed that the polytron in combination with sonication is the most effective reduction technique (Table 2). The 24mL glass tissue grinder technique produced lower average particle sizes compared to Polytron, but it is time consuming and labor intensive, requiring the operator to manually grind the sample. Glass beads worked very well for small volumes of sample with the drawback of potential high sample loss and time consumption. Glen-Mills high speed mixer offers the advantage if the materials can be provided in large quantity

Table 2. Particle size reduction techniques summary

Technique	API need and recovery	Time Consumption	Particle size reduction	Narrow Particle size distribution	Easy to work
Plastic tissue grinder	+	−	+	+	−
Glass tissue grinder	+	−	+	+	−
Polytron with metal tips	+	+	+	+	+
Poytron with plastic tips	+	+	+	+	+
Glass beads	−	−	+	+	+
Gen Mills High speed Mixer	−	+	+	+	+
Polytron +Sonicator	+	+	++	++	+

* + effective, −not effective or show disadvantage

4. CONCLUSIONS

For practical use for early drug discovery support, the amount of API is very limited and precious, considering effectiveness and efficiency at the same time; Polytron homogenizer is the best choice for particle size reduction in order

to improve dissolution and bioavailability of poorly soluble compounds. However, the physico-chemical properties play an important role of size reduction too. For example, the elastic compounds (e.g. Naproxen) are harder to break down compared to brittle compounds such as Avicel PH 101. In general, all the techniques tested in this study turned out to work better on suspensions than dry powders. The choice of the technique selection should also rely on the availability of each lab or company and experience of the researchers.

5. ACKNOWLEDGEMENT

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