Drug Development and Industrial Pharmacy, 26(3), 337–342 (2000)

COMMUNICATION

Validation of Tablet Dissolution Method by High-Performance Liquid Chromatography

Jian-Hwa Guo,* W. W. Harcum, G. W. Skinner, P. R. Dluzneski, and D. E. Trumbull

Hercules Incorporated, 500 Hercules Road, Wilmington, DE 19808

ABSTRACT

Dissolution is a qualitative and quantitative tool that can provide valuable information about biological availability of a drug, as well as batch-to-batch consistency. It is considered one of the most important quality control tests performed on pharmaceutical dosage forms, and validation of dissolution methods is an important part of good manufacturing practices (GMP). Hydroxypropylcellulose (HPC) was formulated with acetaminophen (APAP) and hydrochlorothiazide (HCTZ). Dissolution methods and limits are reported in the USP/NF. Standard operating procedures (SOP) for the HP 8452A spectrophotometer and Vanderkamp 600/6010 Dissolution Tester were followed according to the GMP Manual. A dissolution method was developed for each formulation based on the above. The only discrepancy between high-performance liquid chromatography (HPLC) and standard dissolution testing occurred when comparing the results of the HPC/HCTZ formulation. The ultraviolet (UV) samples were filtered through a 10-μm filter, and the HPLC samples were filtered through a 0.2-μm filter. When the HPC/HCTZ samples were filtered through a 10-μm filter for both UV and liquid chromatography (LC), the results were equal. Filter pore size and area have a large effect on concentration of HPC/HCTZ. The smaller the pore size and the smaller the diameter of the filter, the more HPC/HCTZ is filtered out. HCTZ has a greater tendency to interact with HPC in the filter than other active ingredients tested. HPC and HCTZ levels have little or no effect on the amount of HCTZ lost.

Key Words: *Dissolution; HPLC; Hydroxypropylcellulose; UV; Validation.*

* To whom correspondence should be addressed. B.F. Goodrich Company, 9921 Brecksville Road, Brecksville, OH 44141-3289. Telephone: (216) 447-5377. Fax: (216) 447-6361. E-mail: guo@brk.bf′g.com

INTRODUCTION

Dissolution is the process by which a solid solute enters a solution. In the pharmaceutical industry, it may be defined as the amount of drug substance that goes in solution per unit time under standardized conditions of liquid/solid interface, temperature, and solvent composition. The USP/NF has a dissolution limit for most tablet and capsule monographs.

Dissolution behavior of drugs has a significant effect on their pharmacological activities. In vitro/in vivo correlation demonstrates a direct relationship between bioavailability of a drug and its in vitro dissolution rate (1–6). Dissolution rates can be affected by many factors: disintegrants, type, particle size and concentration of binders, lubricants, coatings, method of granulating, compression force, pH and temperature of dissolution medium, and the particle size of the drug. In spite of in vitro/in vivo correlation, dissolution is not a predictor of therapeutic efficiency. Rather, it is a qualitative tool that can provide valuable information about biological availability of a drug, as well as batch-to-batch consistency. Dissolution is considered to be one of the most important quality control tests performed on pharmaceutical dosage forms. Validation of dissolution methods is an important part of good manufacturing practices (GMP).

The objective was to validate the Aqualon Pharmaceutical Laboratory dissolution apparatus and ultraviolet (UV) spectrophotometer using high-performance liquid chromatography (HPLC).

EXPERIMENTAL

The materials used were hydroxypropylcellulose (HPC; Klucel® EXF, Pharm, Hercules Inc., Aqualon Division, Wilmington, DE); acetaminophen (APAP) powder USP (Rhone-Poulenc, Inc., Specialty Chemicals, Cranbury, NJ); hydrochlorothiazide (HCTZ) USP (Abbott Lab., Chem. and Agriculture Products Div., North Chicago, IL); croscarmellose sodium (Ac-Di-Sol®, NF, FMC Corp., Food and Pharmaceutical Division, Newark, DE); magnesium stearate NF (Witco Corp., Organics Division, Chicago, IL); lactose, regular grind, NF (Wisconsin Dairies, Formost Ingredient Group, Baraboo, WI); calcium sulfate, hydrous, NF (U.S. Gypsum Co., Industrial Gypsum Division, Chicago IL); and calcium sulfate dibasic USP (Di-Tab, Rodia, Cranbury, NJ).

Formulation

Tablets from the formulations in Table 1 were used for dissolution testing.

Development of Ultraviolet Method

APAP and HCTZ were the active drugs in the tablet formulations. Dissolution methods and limits are reported in the USP/NF. In this study, dissolution tests were carried out using the USP/NF 23 dissolution test method on a Vanderkamp VK6010 and VK600 Dissolution Apparatus (Vankel Industries, Cary, NC) using a fully automated 8452A diode array UV spectrophotometer (Hewlett Packard Co., Palo Alto, CA). The UV spectrophotometer measures absorbance at a set wavelength, compares the measurements to stored standards, and calculates the percentage released based on label weight. Standard operating procedures (SOP) for the HP 8452A spectrophotometer and Vanderkamp 600/6010 dissolution tester were used in this study. A dissolution method was developed for each formulation based on the above.

Table 1

Tablet Formulations

Ingredient	Amount (mg)	Ingredient	Amount (mg)
APAP, USP	500	HCTZ, USP	50
Lactose regular grind, NF	30.5	Dibasic calcium phosphate , NF	208.75
Calcium sulfate, NF	30.5	Hydroxypropylcellulose, NF	30
Hydroxypropylcellulose, NF	24	Ac-Di-Sol, NF	9
Ac-Di-Sol, NF	12	Magnesium stearate, NF	2.25
Magnesium Stearate, NF	3	Total	300
Total	600		

Calibrator tablets (50 mg prednisone disintegrating tablets and 300 mg salicylic acid nondisintegrating tablets, USP) were used to calibrate the dissolution apparatus for USP dissolution methods 1 and 2.

Acetaminophen

Acetaminophen, an analgesic and antipyretic (pain and fever), is the active ingredient used in wet granulation binder studies. Dissolution tests were done on tablets compressed at 15 kN after they were stored 24 hr. The USP test method for APAP tablets was as follows: Apparatus 2 (paddles) was used at 50 rpm in 900 ml of pH 5.8 deaerated phosphate buffer for 75 min at 37°C. The ultraviolet absorbance of the wavelength was set at 243 nm. The method was a single-component analysis made with three standard solutions having a known concentration in the same medium. USP tolerance for APAP is not less than 80% dissolved in 30 min (T_{80} value $\geq$ 80% dissolved in 30 min). The dissolution timetable was set at: 5, 10, 15, 20, 30, 45, 60, and 75 min. The UV absorbance of the wavelength was set at 243 nm.

Two different concentrations of APAP solutions were prepared by adding different HPC levels. The APAP concentrations were 0.3 and 0.55 mg/ml, and the HPC levels were around 0.02 and 0.04 mg/ml. Those solutions were filtered with different pore size or area size filters, and the filtered solutions were analyzed by UV or HPLC methods for APAP concentrations.

Hydrochlorothiazide

HCTZ is the active ingredient used in direct compression (DC) binder studies. Dissolution tests were done on HCTZ tablets compressed at 15 kN after they were stored 24 hr. The USP test method for HCTZ tablets was as follows: Apparatus 1 (baskets) was used at 100 rpm in 900 ml of 0.1 N HCl for 75 min at 37°C with not less than 60% dissolved in 60 minutes (T_{60} value). The method was a single-component analysis made with four standard solutions having a known concentration in the same medium. The UV absorbance of the wavelength was set at 272 nm. The dissolution timetable was set at 5, 10, 15, 20, 30, 45, 60, and 75 min.

Development of High-Performance Liquid Chromatography Method

Samples for HPLC were taken at the same time interval as the fully automated diode-array UV spectrophotometer. The HPLC method used to analyze APAP, phenylpropanolamine, and HCTZ for dissolution studies is described briefly below.

A Zorbax Rx C-18 column (15 cm $\times$ 4.6 mm $\times$ 5 μm) was used in the liquid chromatographic method. The isocratic mobile phase was set at 96% acetonitrile and 4% aqueous phosphate, which had a flow rate of 1.0 ml/min. The aqueous portion of the mobile phase contained 0.025 M potassium phosphate and 0.025 M phosphoric acid. The injection volume was 20 μl, and the column temperature was set at 40°C. With a diode-array detector, UV absorbance was monitored simultaneously at two different wavelengths, which were selected according to the maximum absorptivity of the three compounds involved in the study. HCTZ and phenylpropanolamine were monitored and quantified using absorbance at 205 nm, while APAP was monitored at 254 nm. All three peaks are well resolved with this method and eluted in less than 5 min.

In preparing for this study, a series of calibration solutions containing the appropriate standards were made by serially diluting concentrated stock solutions. The typical concentration range of these calibration solutions was from 0.009 to 0.075 mg/ml, which covered the expected range of concentrations in the study. The sample solutions involved in this study were loaded into the autosampler in the instrument with the calibration series at spaced intervals.

RESULTS AND DISCUSSION

UV and HPLC analysis were done on the following formulations. The tablets were compressed at 15 kN and tested after they were stored 24 hr. Dissolution profiles of HPC and APAP for UV and HPLC are equivalent. Filter pore size and area size had no effect on HPLC results for this formulation (Fig. 1). Filtration tests also indicate filter pore size and area size have no effect on HPLC results for APAP solutions at different HPC levels (Fig. 2).

Dissolution profiles of HPC and HCTZ for UV and HPLC are equivalent when both are filtered through a 10-μm filter. Filter pore size and area size have a large effect on HPLC results for HPC/HCTZ formulations (Fig. 3). The smaller the pore size and the smaller the diameter of the filter, the more HCTZ is filtered out. Figure 4 shows the effect of filter pore size with different HPC/HCTZ combinations. The smaller the pore size and the smaller the diameter of the filter, the lower the HCTZ concentration.

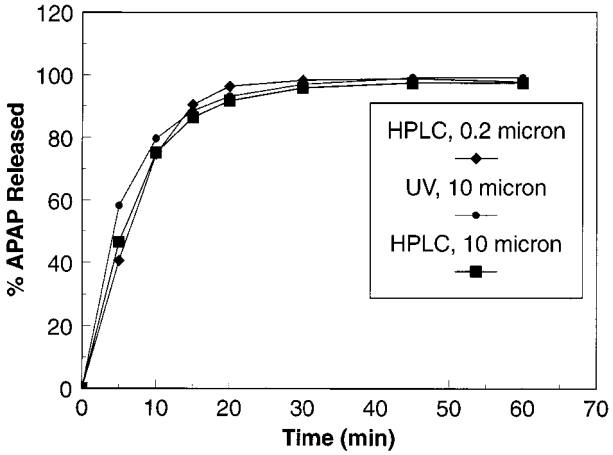

Figure 1. Dissolution results for acetaminophen tablets, UV versus HPLC.

Figure 5 shows that the HPLC analysis of HPC samples yielded a lower than expected HPC concentration. In the case of the lower concentration, the HPC peak was near the detection limit for this method. This increases the possibility for error in quantitation due to baseline noise. For all samples, HPLC analysis yielded a lower estimate than expected for HPC concentration. In comparing the results, it appears that filters with a smaller pore size filter out more HPC.

HPLC results have been used to validate the UV dissolution methods. The only discrepancy occurred when comparing the results of the HPC/HCTZ formulation. The UV samples were filtered through a 10-μm filter, and the HPLC samples were filtered through a 0.2-μm filter. When the HPC/HCTZ samples were filtered through a 10-μm filter for both UV and HPLC, the results were equal. Filter pore size had a large effect on concentration of HPC/HCTZ. The smaller the pore size and the smaller the diameter of the filter, the more HPC/HCTZ was filtered out. HCTZ has a greater tendency to interact with HPC in the filter than other active ingredients tested. HPC and HCTZ level have little or no effect on the amount of HCTZ lost.

CONCLUSIONS

The HPLC results validated the dissolution UV methods for APAP and HCTZ. The only discrepancy occurred when comparing the results of the HPC/HCTZ formulation. The UV samples were filtered through a 10-μm filter, and the HPLC samples were filtered through a 0.2-

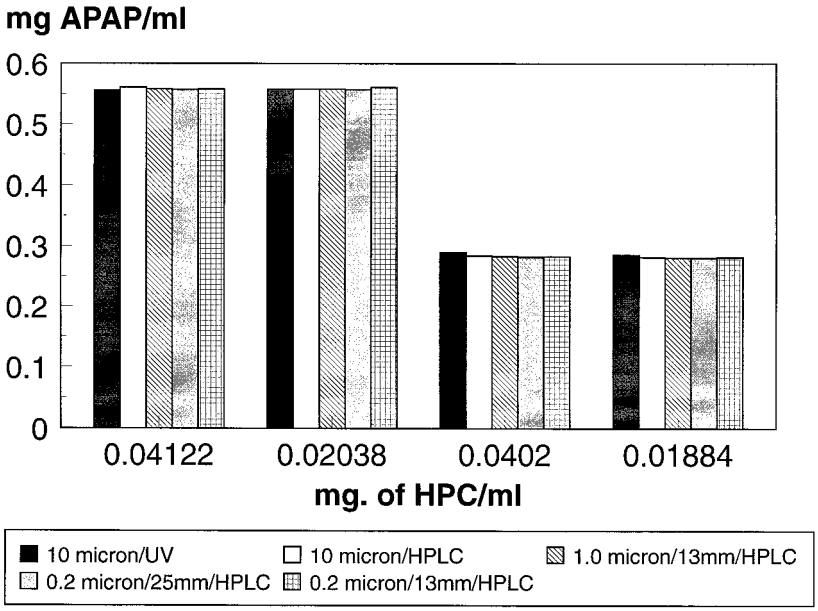

Figure 2. Filtration results of acetaminophen solutions with different levels of hydroxypropylcellulose, UV versus HPLC.

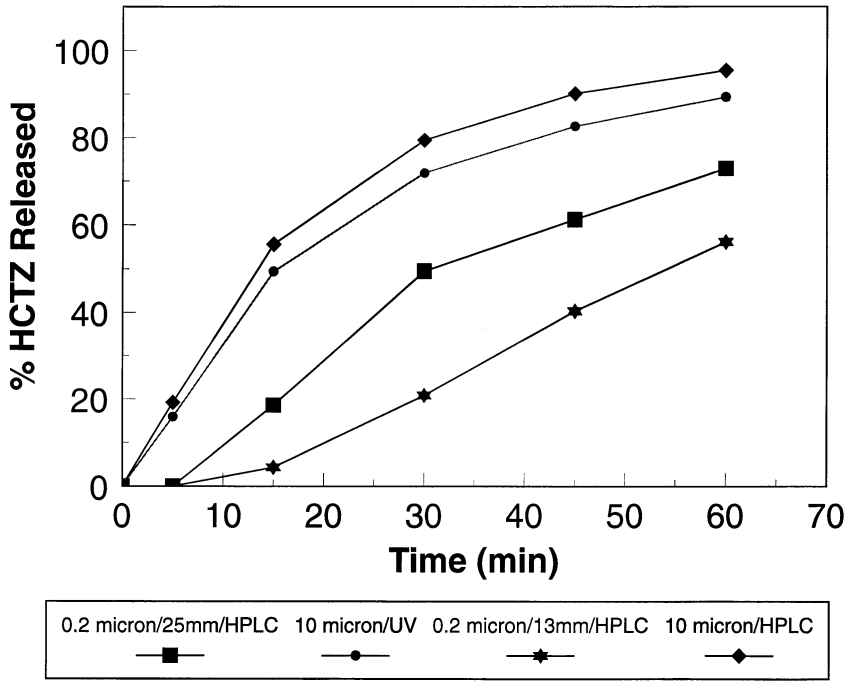

Figure 3. Dissolution results for hydrochlorothiazide tablets, UV versus HPLC.

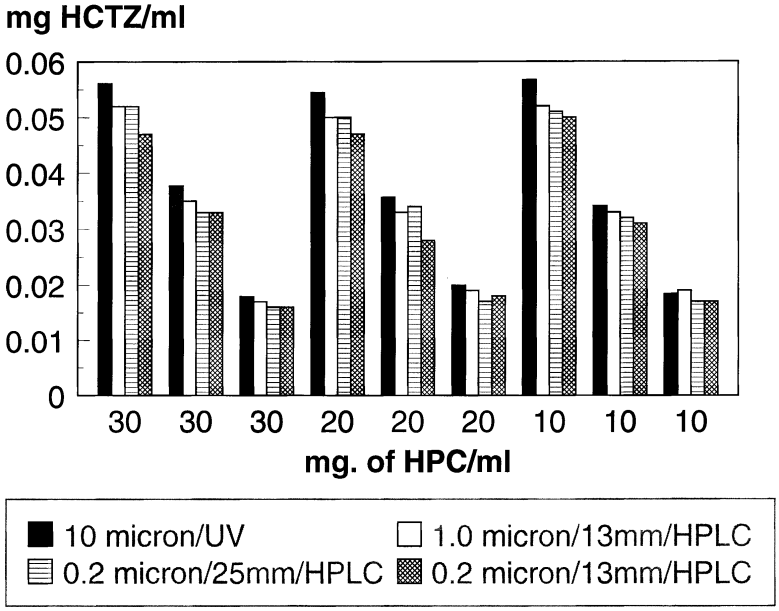

Figure 4. Filtration results for hydrochlorothiazide solutions with diferent levels of hydroxypropylcellulose, UV versus HPLC.

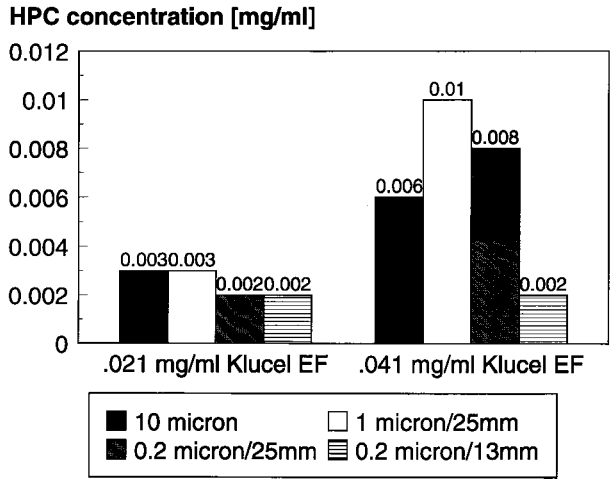

Figure 5. Filtration results for different concentrations of hydroxypropylcellulose solutions.

μm filter. When the HPC/HCTZ samples were filtered through a 10-μm filter for both UV and LC, the results were equal. Filter pore size had a large effect on concentration of HPC/HCTZ. The smaller the pore size and the smaller the area of the filter, the more HPC/HCTZ is filtered out. HCTZ has a greater tendency to be trapped with HPC in the filter than APAP. HPC and HCTZ level have little or no effect on the amount of HCTZ lost. This discrepancy was not seen with HPC/APAP combinations. Dissolution rates can be affected by many factors: disintegrants, type and concentration of binders, lubricants, coatings, method of granulating, compression force, pH and temperature of dissolution medium, and the particle size of the drug. Validation of dissolution methods is necessary for formulation research and development.

REFERENCES

1. L. W. Ditter et al., J. Pharm. Sci., 57, 1146 (1968).
2. L. W. Ditter et al., J. Pharm. Sci., 57, 1269 (1968).
3. A. J. Hussain, Pharm. Sci., 61, 811 (1972).
4. D. C. Monkhouse and J. L. Lach, Can. J. Pharm. Sci., 7, 29 (1972).
5. J. Blanchard, Am. J. Pharm., 150, 132 (1978).
6. L. Z. Benet, G. Levy, and B. L. Ferraiolo, *Pharmacokinetics—A Modern Review*, Plenum, New York, 1984.