DRUG DEVELOPMENT AND INDUSTRIAL PHARMACY, 20(12), 1957-1969 (1994)

FREEZE-DRIED PREPARATIONS OF KETOPROFEN AND HEPTAKIS-(2,6-O-DIMETHYL)-ß-CYCLODEXTRIN

O. Funk, L. Schwabe, K.-H. Frömming

Institut für Pharmazie der Freien Universität Berlin
Kelchstraße 31, 12169 Berlin, Germany

ABSTRACT

The purpose of this study was to examine the inclusion formation in freeze-dried preparations of ketoprofen and heptakis-(2,6-O-dimethyl)-ß-cyclodextrin. The products obtained were amorphous mixtures of the two components. X-ray diffractography, differential scanning calorimetry, ^{13}C-CPMAS-NMR spectroscopy and thermofractography did not give any proof of an inclusion of ketoprofen in the cyclodextrin cavity.

INTRODUCTION

Freeze-drying is one of the methods for the preparation of cyclodextrin (CD) complexes [1, 2, 3]. This procedure is used primarily for the preparation of complexes with easily water soluble CD-derivatives. Previous papers confirmed the formation of pure inclusion compounds in freeze-dried preparations of

nicotinic acid/ß-CD [4] and ketoprofen/ß-CD [5], respectively.

The purpose of this investigation is to examine how far an inclusion compound can be obtained between keto-profen and heptakis-(2,6-0-dimethyl)-ß-CD (DIMEB) in freeze-dried preparations. DIMEB is easily water soluble whereas the solubility of ß-CD is about 1.8 g·100 ml^{-1} water. The experiments were performed by using X-ray diffractometry, IR-spectrometry, thermo-analytical methods and ^{13}C-CPMAS-NMR spectroscopy for inclusion confirmation.

MATERIALS AND METHODS

Materials

Ketoprofen (Klinge Co., München, Germany), Heptakis-(2,6-di-0-methyl)-ß-cyclodextrin (Lehmann & Voss Co., Hamburg, Germany)

Preparations

The physical mixtures of ketoprofen and DIMEB (2:3) were prepared during 15 min mixing in a Turbula mixer, type T2C (Bachofen Co., Basel, Switzerland).

Freeze-dried preparations: 125 mg ketoprofen and 1 g DIMEB were dissolved in 2 ml water and frozen as follows: Procedure **1**: Deep-freeze chest (-40°C) on non-prechilled shelves of 20 cm diameter and 18 mm height for 24 hours; **2**: like **1**, but on prechilled shelves; **3**: Freezing bath (-30°C); Freezer, type K, with thermo-stat F3 (Haake Co., Berlin, Germany); **4**: Aceton/dry ice mixture (-85°C) on prechilled shelves; **5**: Liquid nitro-gen (-196°C). The frozen samples **3** to **5** are stored over night at -40°C before the freeze-drying starts. Freeze

dryer, Gamma 2-20 (Christ Co., Osterode, Germany). Main drying 24 h, 0.1 to 0.05 hPa on non-heated shelves. Secondary drying 48 h, 0.1 to 0.05 hPa, shelf temperature 50°C.

Physical and Physical Chemical Determinations

Solubility studies: The solubilities were evaluated according to Higuchi and Connors [6] by shaking aqueous mixtures of excess amounts of ketoprofen and various concentrations of ß-CD for 7 d at 25°C. Ketoprofen was assayed by UV-spectrometry at 260 nm in aqueous solution. UV-spectrometer, type Uvikon 710 (Kontron Co., Eching, Germany).

IR spectra were scanned with the IR-spectrometer SP 1000 (Pye-Unicam, Cambridge, England), KBr method.

^{1}H-NMR spectra were scanned using a spectrometer AC 300 (Bruker Co., Karlsruhe, Germany) with Na-3-trimethylsilyl-propionate-2,2,3,3-d_4 as internal standard.

^{13}C-CPMAS-NMR spectra were recorded at 62,9 MHz using a BRUKER AC-250 (Karlsruhe, Germany) spectrometer.

X-ray diffractograms were obtained using a powder diffractometer 1700 (Philips, Eindhoven, Netherlands), Cu-$K_{\alpha 1}$ radiation.

The specific surface areas were determined by the BET method with an Areameter (Ströhlein Co., Düsseldorf, Germany).

Thermofractographic Measurements

The thermofractographic measurements were performed in a TAS oven (Desaga Co., Heidelberg, Germany); sample weight 1 mg, heating rate 3 min at each temperature grade from 50°C to 320°C, 100 $ml \cdot min^{-1}$ N_2. TLC conditions [7]: Silica gel 60 F_{254}; formic acid, acetone, toluene, 1:39:60 (v/v/v), as mobile phase. Detection under UV at 254 nm.

Dissolution Behavior

The dissolution rate of ketoprofen was determined by the paddle method DAB 9 with the automatic dissolution tester Dissograph (Hanson, Northridge, Ca., USA): 900 ml 0.1 N HCL, 37°C, 50 rpm; sample weight 9 mg ketoprofen equivalents.

RESULTS

Complex Formation in Solution

There is a considerable solubilizing effect of DIMEB on ketoprofen. The phase-solubility diagram can be classified as A_L-Type (Fig. 1). The complex stability constant was calculated from the ascending line of the solubility isotherm according to Higuchi and Connors [6] assuming 1:1 stoichiometry. The value of the stability constant is 3700 M^{-1}.

^{1}H-NMR spectroscopy proves the inclusion of ketoprofen in the CD cavity because of the shifts of the inner protons H3 and H5 to a higher field. The $\Delta\delta$ values of the protons H1 to H6 are -0.05, -0.02, -0.10, -0.03, -0.16, -0.08 ppm, respectively. The effect of the shift is greater for H5 than for H3.

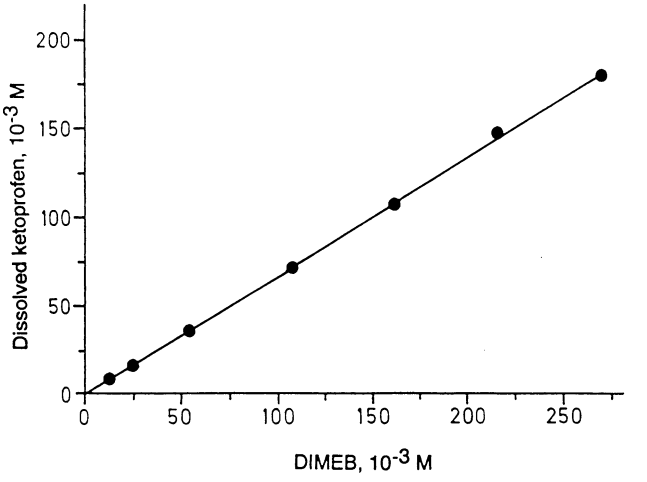

FIGURE 1

Phase-solubility diagram of ketoprofen and DIMEB

Freeze-Dried Preparations of Ketoprofen and DIMEB

Aqueous solutions of ketoprofen and DIMEB were prepared in the molar ratio of 2:3 and frozen with 5 different freezing temperatures. The freezing rates are in the rank order **1<3<2<4<5**. Faster freezing results in a greater surface area of the freeze-dried product (Fig. 2).

The X-ray diffractogram of the physical mixture of ketoprofen and DIMEB (2:3) is characterized exclusively by the peaks of DIMEB (Fig. 3). The amount of ketoprofen is too small for a peak to appear. All ketoprofen/DIMEB freeze-dried products are amorphous, independent of the freezing procedure. Short heating results in a recrystallization. The diffractograms of all 5 recrystallized freeze-dried products are of the

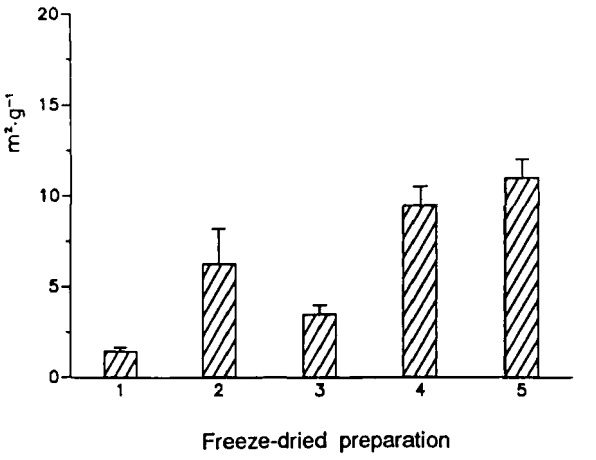

FIGURE 2

Specific surface areas of freeze-dried ketoprofen/DIMEB preparations

same type. They do not differ from that of the physical mixture.

The IR-spectra of the freeze-dried products demonstrate a shift of the carbonyl band of ketoprofen at 1690 cm^{-1} to 1722 cm^{-1}.

The DSC thermogram of the physical mixture shows the melting peak of ketoprofen between 90°C and 95°C (Fig. 4). This endotherm is not present in the freeze-dried product. But the freeze-dried product shows an exothermic peak at 140°C which should be lead back to a recrystallization of the amorphous structure. The thermoanalytic behavior does not differ among the preparations obtained by the different freezing methods.

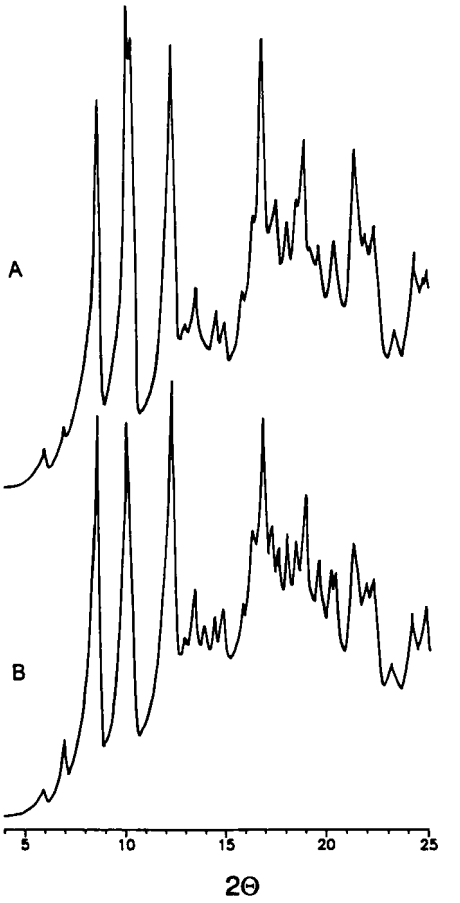

FIGURE 3

X-ray powder diffraction patterns of ketoprofen/DIMEB preparations
A: Physical mixture (2:3), B: Freeze-dried preparation **2**, recrystallized

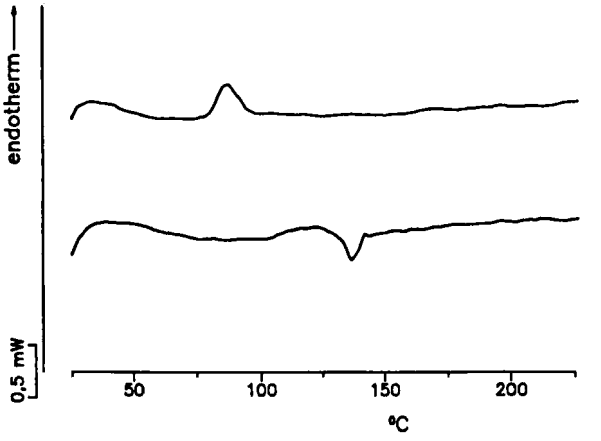

FIGURE 4

DSC thermograms of ketoprofen/DIMEB preparations A: Physical mixture (2:3), B: Freeze-dried preparation 2

Thermofractographic experiments of the physical mixture and the freeze-dried product results in a largely identical result (Fig. 5). The largest part of ketoprofen is released in both cases between 170°C and 230°C. There is no remarkable difference in the thin layer chromatograms among the different freeze-dried products.

Fig. 6 shows the ^{13}C-CPMAS-NMR spectra of the physical mixture and the freeze dried product. The carbon peaks of the CD in the freeze-dried product are unchanged, whereas the carbon peaks of the ketoprofen are broader than in the mixture.

Ketoprofen in the freeze-dried products dissolves faster than in the physical mixture (Fig. 7). The

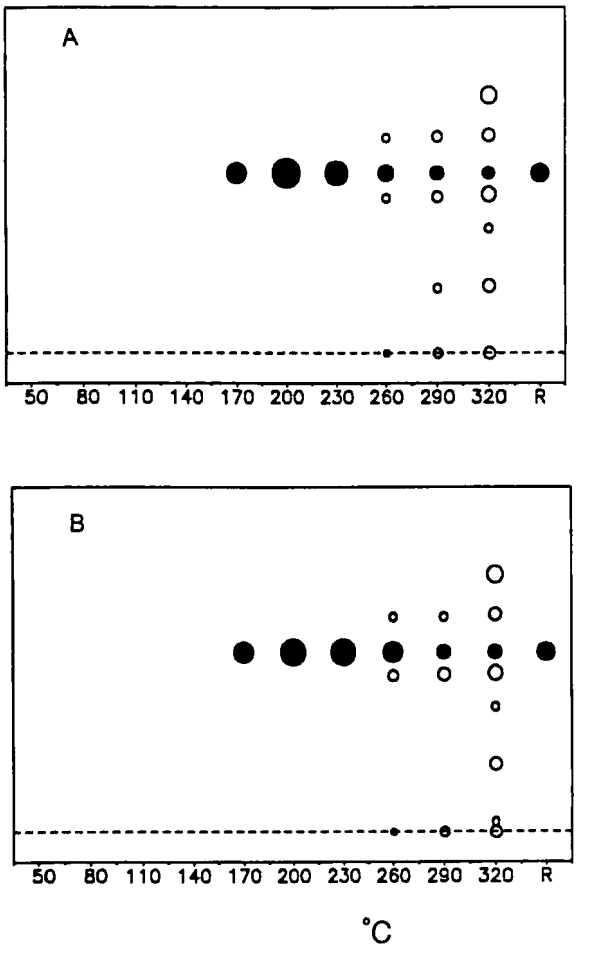

FIGURE 5

Thermofractograms of ketoprofen/DIMEB preparations
A: Physical mixture (2:3), B: Freeze-dried preparation 2

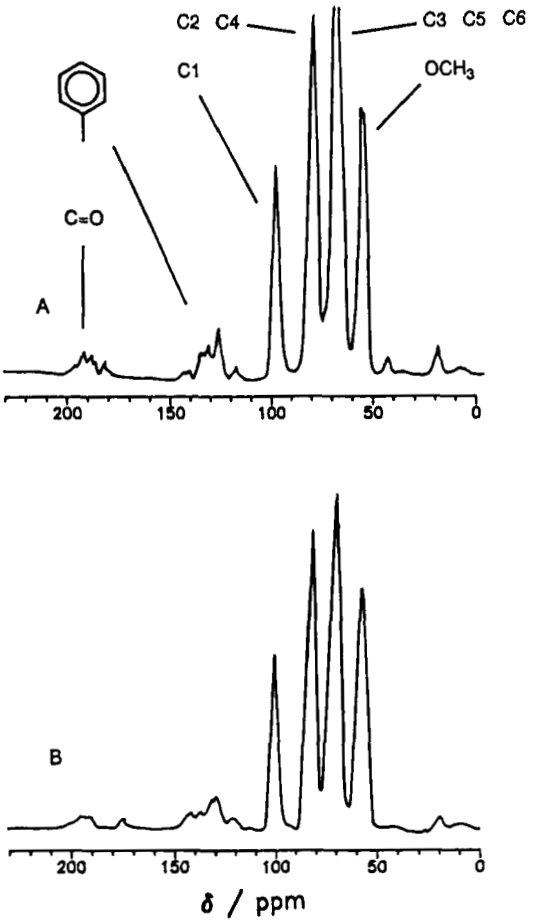

FIGURE 6

^{13}C-CPMAS-NMR spectra of ketoprofen/DIMEB preparations
A: Physical mixture, B: Freeze-dried preparation 2

dissolution process is practically finished after 10 minutes.

DISCUSSION

The phase-solubility behavior and the ^{1}H-NMR experiments prove the existence of an inclusion com-

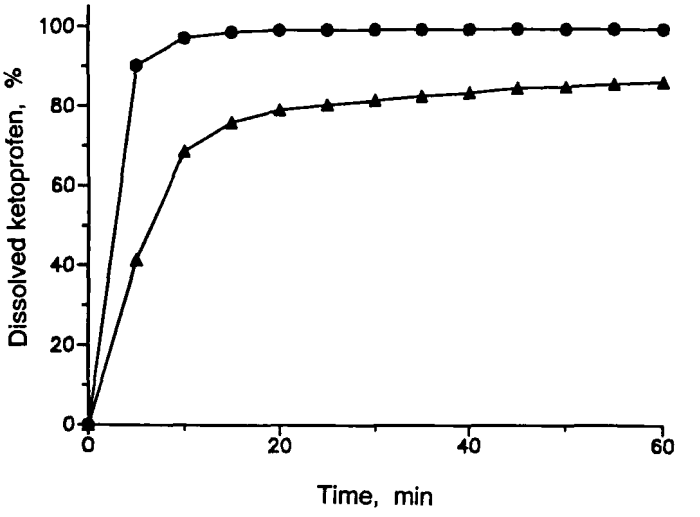

FIGURE 7

Dissolution behavior of ketoprofen/DIMEB preparations
A: Physical mixture, B: Freeze-dried preparation **2**

pound between ketoprofen and DIMEB in solution. The specific surface areas of the freeze-dried preparations increase with faster freezing rates (Fig. 2).

None of the analytical methods used give any hints on an inclusion formation between ketoprofen and DIMEB in the freeze-dried preparations. The shift of the carbonyl stretching band of ketoprofen at 1689 cm^{-1} to higher wave numbers which can be explained by the breakdown of the intermolecular hydrogen bonds, permits the assumption that ketoprofen exists as monomer.

The absence of the melting peak of ketoprofen in the DSC thermograms of the freeze-dried products is expected because of the amorphous structure. The thermofractographic results definitely prove that ketoprofen is not included in the cavity of DIMEB.

^{13}C-CPMAS-NMR spectra do not let us recognize an influence of ketoprofen on the signals of the carbon atoms of cyclodextrin. The change of the ketoprofen signals in the freeze-dried product compared to the physical mixture, can be established as well by a slow movement of ketoprofen in the cavity of DIMEB as by the amorphous structure.

The higher dissolution rates of the freeze-dried preparations compared to the physical mixture can be explained with their greater specific surface areas and their amorphous state. The different surfaces among the 5 freeze-dried preparations do not influence the dissolution behavior.

In summary it can be said that in freeze-dried products of ketoprofen and DIMEB no inclusion compound exists, but the two components occur as a physical mixture. Comparable results were found in the systems of nicotinic acid/DIMEB [4] and vitamin A acetate/DIMEB [8]. The explanation can be given by the phase-solubility behavior. The phase-solubility diagram is classified as A_L-type. If at the eutectic point with saturated single components the solubility of the inclusion compound is not exceeded, no solid inclusion compound precipitates at the freezing process. In contrast, the phase solubility diagram of the ketoprofen/ß-CD is classified as B_S-Type. Because of the limited aqueous solubility of ß-CD (1.8 g·100 ml^{-1}) the solubility product of the ketoprofen/ß-CD inclusion compound is exceeded at the eutectic point.

ACKNOWLEDGEMENTS

The authors are grateful to Wacker Chemie Co. and Klinge Co. for generous gifts of DIMEB and Ketoprofen,

respectively. The X-ray powder diffractogram measurements were done in the Institut für Anorganische und Analytische Chemie der Freien Universität Berlin (Prof. Dr. H. Hartl, Mrs. I. Brüdgam), the ^{13}C-CPMAS-NMR spectroscopic determinations in the Institut für Organische Chemie der Freien Universität (Prof. Dr. K. Roth). We thank them for valuable discussions.

The work was supported by the Fonds der Chemischen Industrie for which we are very grateful.

REFERENCES

1. M. Koruzumi, N. Nambu and T. Nagai, Chem.Pharm. Bull. 23, 3062 (1975).
2 T. Oguchi, M. Okada, E. Yonemochi, K. Yamamoto and Y. Nakai, Int.J.Pharm. 61, 27 (1990).
3. J. Blanco, J.L. Vila-Jato, F. Otero and S. Anguiano, Drug.Dev.Ind.Pharm. 17, 943 (1991).
4. O. Funk, L. Schwabe and K.-H. Frömming, J.Incl.Phenom., in press.
5. K.-H. Frömming, L. Schwabe and O. Funk, in "Minutes of the 6th Int. Symp. on Cyclodextrins", A.R. Hedges, ed., Editions de Santé, Paris, 1992, p.308.
6. T. Higuchi, K.A. Connors, Adv.Anal.Chem.Instrum. 4, 117 (1965).
7. H. Auterhoff and K.-A. Kovar, Identifizierung von Arzneistoffen, 5. Aufl., Wissenschaftliche Verlagsges.mbH, Stuttgart, 1985, p. 76.
8. T. Gelder, Ph.D. Thesis, Freie Universität Berlin, 1988.