Investigation of Primary and Secondary Modifiers for the Subcritical Extraction of Lovastatin from MEVACOR Tablets with Carbon Dioxide

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Abstract

The subcritical fluid extraction of lovastatin from tablet powder mixtures prepared in this laboratory and MEVACOR® tablets is successfully demonstrated. Methanol modifier percentage, additive type (acidic, basic, or neutral), and additive concentration on the extraction efficiency are examined. The extraction recoveries of lovastatin from MEVACOR tablets are shown to be highly dependent on methanol concentration and additive type. Isopropylamine is shown to be the most successful additive investigated. An optimized extraction method is developed, and lovastatin recoveries of 99.5% were achieved with a relative standard deviation of 1.2% from MEVACOR tablets with 15% (v/v) (1.0% [v/v] isopropylamine) methanol-modified CO_2.

Introduction

Past studies in both sub- and supercritical fluid extraction (SFE) and chromatography (SFC) have primarily utilized CO_2 as the supercritical fluid (SF) because it is relatively inert, highly pure, nontoxic, exhibits readily attainable critical parameters (critical temperature [T_c], 31°C; critical pressure [P_c], 71 atm), and has a solvent power equivalent to common organic solvents such as hexane. Super- and subcritical CO_2 has been proven to be an efficient medium in the extraction of nonpolar and moderately polar compounds; however, its solvating power may be insufficient for the extraction of highly polar compounds such as most pharmaceuticals. In fact, the nonpolar nature of CO_2 has been one of the major obstacles preventing its acceptance in the pharmaceutical industry as an extraction fluid. This limitation may be overcome by either using a more polar SF like ammonia or by adding small amounts of polar organic solvents (i.e., modifiers) to CO_2. A polar SF such as ammonia is rarely used due to its toxicity, reactivity, and extreme critical parameters (T_c = 132°C, P_c = 111 atm). As a result, most pharmaceutical applications have utilized modified CO_2 (1–4).

Modifiers generally serve two functions: to increase the solvating power of the SF and facilitate the disruption of analyte–matrix interactions (5). For instance, felodipine, an antihypertensive drug with considerable basicity, was found to be soluble in 100% CO_2; however, when extracting a sustained-release tablet containing felodipine, only 60% was recovered with pure CO_2 under similar conditions (3). To achieve quantitative extractions (97–103%), 8% (v/v) methanol-modified CO_2 was apparently needed to disrupt analyte–matrix interactions. Alternatively, the modifier may be more effectively used by introducing it directly to the matrix prior to extraction with CO_2.

The addition of a modifier to either the SF or to the matrix prior to SFE may not be sufficient for the extraction of multifunctional, highly polar compounds (i.e., salts). A secondary modifier (i.e., additive), however, may be added to the primary modifier, in this case, to achieve successful analyte extraction. The additives typically consist of relatively strong organic acids or bases and are usually added directly to the primary modifier (0.1–5% [v/v]) rather than to the fluid or matrix (6). Additives have also been used for several years to improve peak shape and enhance separation in SFC of polar compounds (6–8). Berger and Deye have demonstrated that compounds containing more than two carboxylic acid groups on a benzene ring could not be eluted from a sulphonic acid column with less than 20% methanol-modified CO_2; however, when an additive such as citric acid was added to methanol-modified CO_2, benzene mono-, di-, and tricarboxylic acids could be separated and eluted (9). The general guideline for use of additives in SFC is that acidic analytes require acidic additives and basic analytes require basic additives. Although the solubility of an analyte may be high in CO_2, successful extraction of the analyte from a complicated matrix such as a tablet may be problematic due to analyte–matrix interactions. It was, therefore, our objective to investigate the role of secondary modifiers (i.e., additives) for the extraction of a commonly used pharmaceutical compound from a complicated matrix such as a tablet. To our knowledge, no papers to this date have been published about general guidelines

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Table I. Extraction and Trapping Conditions Used for Phases A–D

CO_2 pressure:	400 atm
Oven temperature:	40°C
Liquid flow rate:	2.0 mL/min
Restrictor temperature:	50°C
Solid-phase trap:	50/50 (w/w) Porapak Q/Glass Beads
Liquid tandem trap:	methanol
Liquid tandem trap volume:	5 mL, room temperature; 7 mL, room temperature (Phase D, reproducibility)
Collection temperature (solid-phase):	40°C
Desorption temperature (solid-phase):	40°C
Solid-phase trap rinse solvent:	2.0 mL methanol* (Phases A–D) 5.0 mL methanol (Phase D, reproducibility)
Rinsing flow rate:	1.0–2.0 mL/min (Phases A–D)

* The solid-phase trap was rinsed directly into the liquid tandem trap following each dynamic extraction step when constructing extraction profiles.

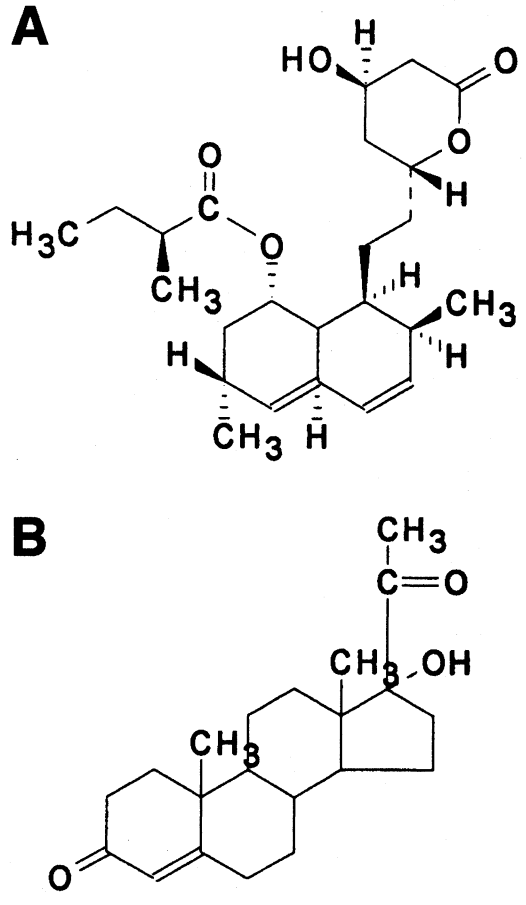


Figure 1. Chemical structures for (A) lovastatin and (B) 17-α-hydroxyprogesterone.

on how to use additives in SFE.

The study was divided into four parts with lovastatin (active ingredient in MEVACOR® tablets) as the prototype drug. Phase A determined the effect of methanol on the extractability of lovastatin from a tablet mixture prepared in this laboratory. The role of additive type (acidic, basic, and neutral) was then investigated in Phase B. The effect of additive and modifier concentration was next investigated in Phase C. Once an optimum additive concentration was chosen (Phase D), the usefulness of the additive with methanol versus methanol-modified CO_2 alone was examined in terms of overall extraction recovery and time needed to extract lovastatin directly from commercially available MEVACOR tablets. Finally, the reproducibility of the optimum SFE method was demonstrated.

Experimental

All extractions were performed on the Isco Suprex Prepmaster (Lincoln, NE). CO_2 (SFE–SFC grade) with helium headspace was donated by Air Products and Chemicals (Allentown, PA). For all tablet extractions, either approximately 100 mg of tablet powder prepared in this laboratory (Phases A–C) containing 10 mg of lovastatin (Merck Research Laboratories, West Point, PA) or a crushed MEVACOR tablet (Phases D–E) containing 10 mg lovastatin was placed into an extraction vessel (5-mL volume [Phase A–B] or 3.5-mL volume [Phase C–D], Keystone Scientific, Bellefonte, PA) containing cotton balls. Cotton balls were used to reduce the dead volume of the vessel. The extraction and trapping conditions for this study are found in Table I.

Extract analysis

After the extraction, trapping, and recovery steps, approximately 0.4 mg of 17-α-hydroxyprogesterone (Figure 1) was added to the combined liquid and solid-phase trap rinses as an internal standard. A portion of each solution was then transferred to SFC vials for analysis.

A prototype of the Hewlett Packard model G1205 SFC system (Little Falls, DE) was used for Phases A, B, and D (15% methanol-modified CO_2 mixtures and reproducibility) analyses. All other analyses were performed on the Gilson SF3 SFC system (Middleton, WI). All separations were performed isocratically with a Hypersil silica column (25 cm × 4.6-mm i.d., 3-µm particle diameter, Keystone Scientific, Bellefonte, PA) and a mobile phase consisting of 6% (v/v) (0.5% [v/v] trifluoroacetic acid [TFA]) methanol-modified CO_2 at a pressure of 230 bar and a liquid flow rate of 2.0 mL/min. The purpose of the TFA was to eliminate peak tailing of a possible degradation product, hydroxy acid lovastatin. The peak shape of lovastatin was not affected by the addition of the acidic additive to the mobile phase (J.T.B. Strode, L.T. Taylor, A.L. Howard, D. Ip, and M.A. Brooks. Analysis of lovastatin by packed column supercritical fluid chromatography. Submitted for publication.). The column was maintained at 45°C. The injection solvent consisted of methanol, and the volume used was 5 µL. Detection was ultraviolet (UV) at 230 nm. A UV flow cell maintained at room temperature with a 10-µL volume was used.

Traditional liquid extraction

Approximately 100 mg of the in-house tablet powder mixture or a MEVACOR tablet was placed into a 50-mL volumetric flask.

Then 10 mL of an acetic acid–sodium acetate buffer (pH 4.0) was added to the flask, and the solution was sonicated until the tablet powder or tablet was fully disintegrated (15 min). Next, 35 mL of acetonitrile was added to the flask, and the solution was sonicated for 20 min. After cooling to room temperature (30 min), the flask was diluted to 50 mL with acetonitrile. Analyses were performed by SFC on the resulting solutions.

In-house tablet powder mixture

The in-house tablet powder mixture was prepared by mixing all the ingredients except for lovastatin into a round bottom flask. Lovastatin was next dissolved in methanol (50 mL) and added to the flask with stirring. The tablet mixture was allowed to sit overnight in a refrigerator. The flask was then roto-evaporated to remove the methanol. The concentration of lovastatin in the in-house tablet powder mixture was 10 mg lovastatin per 100 mg tablet powder. Four samples were taken to test tablet formulation uniformity (Table II) using the traditional liquid extraction method followed by SFC analysis.

Table II. In-House Tablet Powder Mixture Uniformity with Liquid Extraction Method

Sample	Claim* (%)
1	97.8
2	93.3
3	95.0
4	96.0
Average	95.5
RSD (%)	2.0

SFC conditions used for tablet powder uniformity: column, Hypersil silica (25 cm × 4.6-mm i.d., 3-µm particle diameter); mobile phase, 6% (v/v) (0.5% [v/v] TFA) methanol-modified CO_2; pressure, 230 bar; column temperature, 45°C; liquid flow rate, 2.0 mL/min; injection solvent, methanol; injection volume, 5 µL; detection, UV at 230 nm; UV flow cell volume and temperature, 10 µL, room temperature. For liquid–solid extraction procedures, see Experimental section.

* 100% = 10 mg lovastatin/100 mg prepared tablet powder mixture.

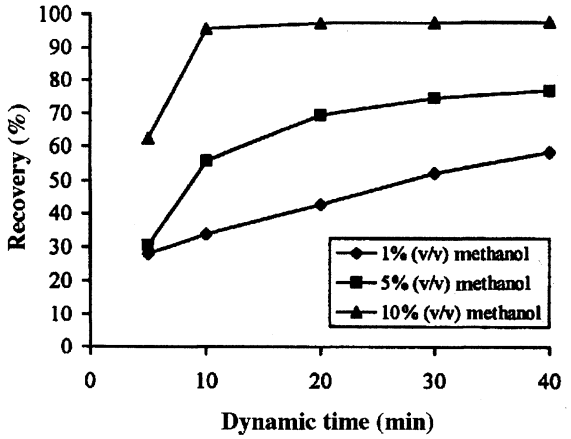


Figure 2. Effect of methanol-modifier concentration on lovastatin recoveries (three replicates) from the tablet powder mixture prepared in-house. SFE conditions: CO_2 pressure, 400 atm; oven temperature, 40°C; liquid flow rate, 2.0 mL/min; restrictor temperature, 50°C; solid-phase trap, 50/50 (w/w) Porapak Q/Glass Beads; liquid tandem trap, methanol; liquid tandem trap volume, 5 mL; collection temperature (solid-phase), 40°C; desorption temperature (solid-phase), 40°C; solid-phase rinse solvent and volume, 2.0 mL methanol; solid-phase rinsing volume, 1.0 mL/min; initial static time, 3.0 min; dynamic time, 40.0 min (total of five dynamic ministeps); static time during trap-rinsing, 2.0 min. Solid-phase trap rinsed directly into tandem liquid trap following each dynamic step.

MEVACOR tablet crushing method

Each commercially prepared MEVACOR tablet (Merck Research Laboratories, West Point, PA) was placed on top of a piece of weighing paper that was sitting in a mortar cup. A pestle was placed on top of the tablet, and pressure was applied until the tablet particles appeared evenly dispersed as a powder. The weighing paper was carefully removed, and the complete crushed tablet was poured into the extraction vessel filled approximately three quarters of the way with a cotton ball. The weighing paper, mortar, and pestle were wiped clean with an additional small piece of cotton. This particular piece of cotton was then placed on top of the other cotton ball inside the extraction vessel. More cotton was added to fill approximately 90% of the vessel volume. The extraction vessel was then sealed.

Results and Discussion

Phase A

Lovastatin, an antihypercholesterolemic drug (Figure 1), was chosen as the test analyte because it is relatively polar and exhibits marginal solubility (0.04% [w/w] at 5000 psi and 40°C) in 100% CO_2 (4). Larson and King found that the solubility was dramatically increased to 0.4% (w/w) with the incorporation of 5% (w/w) methanol-modified CO_2 from a premixed tank (4). Consequently a series of extractions (Table I) were performed to determine the extractability of lovastatin from the tablet powder mixture prepared in-house with the methanol-modified CO_2. Experiments were designed in such a way that an extraction profile could be constructed from the data (Figure 2) in order to examine the effect of methanol concentration and to learn something about the extraction kinetics of lovastatin. A series of dynamic extraction steps followed by trap-rinsing and assay was employed. A tandem solid-phase–liquid trap was employed to ensure quantitative trapping recovery. Lovastatin recoveries were found to be low over the first 40 min where only 58% was extractable with 1% (v/v) methanol-modified CO_2, and 77% was extractable with 5% (v/v) methanol-modified CO_2. When utilizing 1% and 5% (v/v) methanol-modified CO_2, the extraction profile suggested that most of the extractable lovastatin was removed during the first 20 min of dynamic extraction. During this period, the extraction appeared to be dependent on the solubility of the analyte in the methanol-modified CO_2. After 20 min, the extraction process appeared to be limited by the diffusion of the analyte from the matrix into the SF (10). However, quantitative recoveries greater than 97% were achieved from the tablet powder mixture prepared in-house with 10% (v/v) methanol-modified CO_2 employing dynamic extraction ministeps (Figure 2). Because trapping becomes more difficult with modifier con-

centrations greater than 2%, it was of interest to learn if the addition of a secondary modifier could cause reduction of the primary modifier concentration.

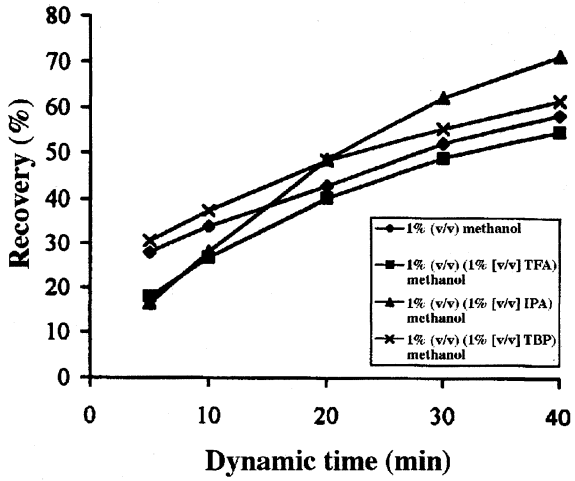


Figure 3. Effect of additive type on lovastatin recoveries (three replicates) from in-house tablet powder mixture. SFE conditions: CO_2 pressure, 400 atm; oven temperature, 40°C; liquid flow rate, 2.0 mL/min; restrictor temperature, 50°C; solid-phase trap, 50/50 (w/w) Porapak Q/Glass Beads; liquid tandem trap, methanol; liquid tandem trap volume, 5 mL; collection temperature (solid-phase), 40°C; desorption temperature (solid-phase), 40°C; solid-phase rinse solvent, 2.0 mL methanol; solid-phase rinsing volume, 1.0 mL/min; initial static time, 3.0 min; dynamic time, 40.0 min (total of five dynamic ministeps); static time during trap-rinsing, 2.0 min. Solid-phase trap rinsed directly into tandem liquid trap following each dynamic step.

Phase B

After determining the effect of methanol on extraction efficiency, the role of the secondary modifier (i.e., additive) type was

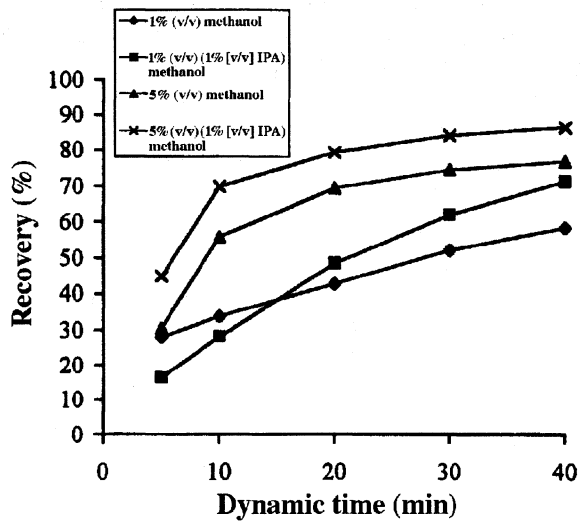


Figure 4. Effect of methanol-modifier concentration on lovastatin recoveries (three replicates) from tablet powder mixture prepared inhouse. SFE conditions: CO_2 pressure, 400 atm; oven temperature, 40°C; liquid flow rate, 2.0 mL/min; restrictor temperature, 50°C; solid-phase trap, 50/50 (w/w) Porapak Q/Glass Beads; liquid tandem trap, methanol; liquid tandem trap volume, 5 mL; collection temperature (solid-phase), 40°C; desorption temperature (solid-phase), 40°C; solid-phase rinse solvent, 2.0 mL methanol; solid-phase rinsing volume, 1.0 mL/min; initial static time, 3.0 min; dynamic time, 40.0 min (total of five dynamic ministeps); static time during trap-rinsing, 2.0 min. Solid-phase trap rinsed directly into tandem liquid trap following each dynamic step.

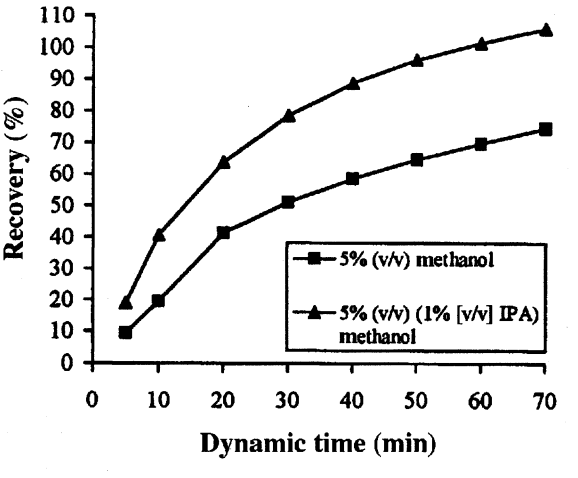


Figure 5. Subcritical fluid extraction (one replicate) of lovastatin from MEVACOR tablets at various additive and modifier concentrations. SFE conditions: CO_2 pressure, 400 atm; oven temperature, 40°C; liquid flow rate, 2.0 mL/min; restrictor temperature, 50°C; solid-phase trap, 50/50 (w/w) Porapak Q/Glass Beads; liquid tandem trap, methanol; liquid tandem trap volume, 5 mL; collection temperature (solid-phase), 40°C; desorption temperature (solid-phase), 40 C; solid-phase rinse solvent, 2.0 mL methanol; solid-phase rinsing volume, 1.0 mL/min; static time, 3.0 min; dynamic time, 70.0 min (total of eight dynamic ministeps); static time during trap rinsing, 2.0 min. Solid-phase trap rinsed directly into tandem liquid trap following each dynamic step.

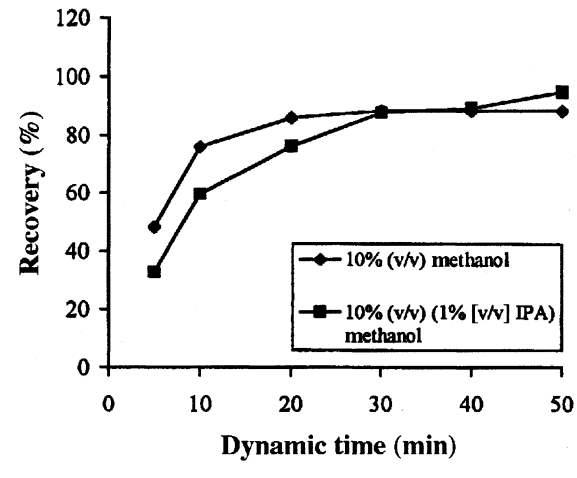
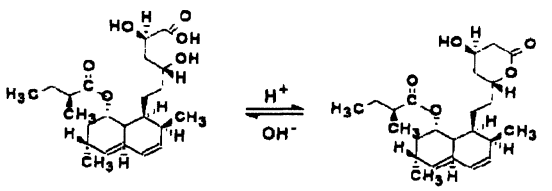


Figure 6. Subcritical fluid extraction (one replicate) of lovastatin from MEVACOR tablets at various modifier concentrations with and without IPA. SFE conditions: CO_2 pressure, 400 atm; oven temperature, 40°C; liquid flow rate, 2.0 mL/min; restrictor temperature, 50°C; solid-phase trap, 50/50 (w/w) Porapak Q/Glass Beads; liquid tandem trap, methanol; liquid tandem trap volume, 5 mL; collection temperature (solid-phase), 40°C; desorption temperature (solid-phase), 40°C; solid-phase rinse solvent, 2.0 mL methanol; solid-phase rinsing volume, 1.0 mL/min; static time, 3.0 min; dynamic time, 50.0 min (total of six dynamic ministeps); static time during trap-rinsing, 2.0 min. Solid-phase trap rinsed directly into tandem liquid trap following each dynamic step.

then investigated. Methanol-modified CO_2 (1% [v/v]) was chosen as the extraction fluid in Phase B due to the limited extractability of lovastatin under these conditions so that the apparent effect of each additive on the extractability could be ascertained. Each additive was introduced directly to the methanol as 1% (v/v). The total additive concentration introduced corresponded to 0.0001%. It can be seen from Figure 3 that isopropylamine (IPA) was the only additive over time that significantly improved the extractability of lovastatin from the prepared tablet powder mixture. In fact, similar extraction recoveries utilizing all three additives (i.e., acid, neutral, and base) were observed during the first 20 min of the extraction. During this time, the extraction was apparently governed simply by the solubility of the lovastatin in the 1% (v/v) methanol-modified CO_2. The extractability of lovastatin, however, increased from 58% with 1% (v/v) methanol-modified CO_2 to 71% with 1% (v/v) (1% [v/v] IPA) methanol-modified CO_2 alter 40 min. T-tests were performed in order to statistically compare the average extraction recoveries after 40 min of all three additives with methanol versus methanol-modified CO_2 alone. With a 95% confidence interval, it was shown that the extraction recoveries of lovastatin (e.g., after 40 min) were statistically greater with the use of IPA than with TFA and tributylphosphate (TBP) or no additive at all.

The increased extractability of lovastatin with the secondary modifier (IPA) after 40 min cannot simply be explained by enhanced solubility, but by a combination of solubility and analyte displacement from the matrix. Excluding the active drug sustance, common tablet ingredients include filling agents such as cellulose and starch as well as lubricants and coloring agents. Cellulose, for example, contains free methoxy and hydroxy acidic sites that contribute to the "activity" of the matrix. Lovastatin, which contains a lactone ring (cyclic ester), may be considered basic due to unshared pairs of electrons on the oxygen in the lactone ring as well as its ability to accept protons. When treated with base, lactone rings are known to open up due to hydrolysis of the cyclic ester. Specifically, Larson et al. reported the conversion of lovastatin in fermentation broth to its hydroxyacid form when in the presence of 3% methanol-modified CO_2 and *t*-butylamine (Equation 1) (4).



Hydroxyacid lovastatin Lovastatin Eq 1

Knowing that lovastatin is basic and that the tablet matrix contains many acidic sites, the enhanced extractability of lovastatin from the tablet powder mixture with the basic additive can be explained by displacement. In this case, when the basic additive was introduced, the stronger base (IPA) preferentially adsorbed to the matrix, thus displacing the basic analyte, lovastatin, from any acidic sites on the tablet powder matrix. The conversion of lovastatin to its hydroxyacid degradate during the extraction with IPA was indeed a concern. However, when the SFC analysis was performed with the capabilities to separate lovastatin and its hydroxyacid degradate, no additional chromatographic peaks were detected. Therefore, it was ensured that the extracted lovastatin was present in the lactonized form. This was expected due to the low amounts of IPA used (0.0001% [v/v]).

Phase C

Because the lovastatin extraction recoveries from the tablet powder mixture were shown to be statistically greater with IPA than the extraction recoveries achieved with the other additives and methanol-modified CO_2 alone, the effect of IPA concentration at various methanol-modified CO_2 concentrations was then investigated further in Phase C. Surprisingly, increased additive concentrations (0.5, 1.0, and 2.0% [v/v] in 1% [v/v] methanol-modified CO_2) at a constant modifier concentration did not affect the extraction recoveries nor the extraction rate. It was believed that all matrix acidic sites were occupied by IPA at a concentration of 0.5% (v/v) in methanol; therefore, increased additive concentrations would not further increase lovastatin extractability.

The usefulness of the IPA additive at various methanol-modified CO_2 concentrations can also be observed in Figure 4. T-tests were performed in order to statistically compare the average extraction recoveries after 40 min with and without IPA in 5% (v/v) methanol-modified CO_2. With a 95% confidence interval, it was shown that the extraction recoveries of lovastatin (e.g., after 40 min) were statistically greater when IPA was used. Once again, lovastatin extraction recoveries at various modifier concentrations were significantly enhanced with the presence of IPA. Overall extraction recoveries over 40 min increased from 77% with 5% (v/v) methanol-modified CO_2 to 86% with 5% (v/v) (1% [v/v] IPA) methanol-modified CO_2.

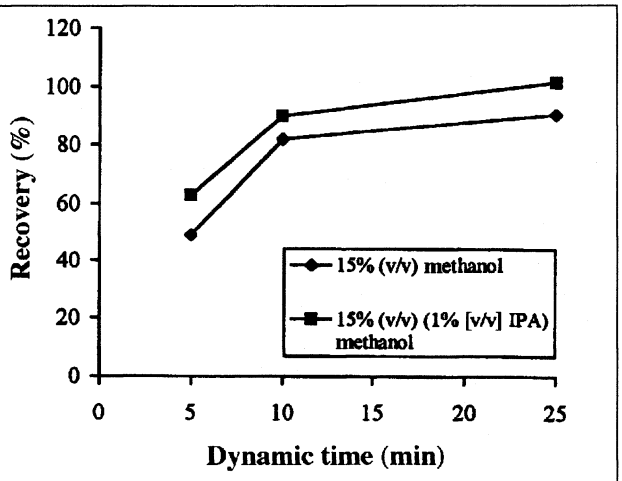


Figure 7. Subcritical fluid extraction (one replicate) of lovastatin from MEVACOR tablets at various modifier concentrations with and without IPA. SFE conditions: CO_2 pressure, 400 atm; oven temperature, 40°C; liquid flow rate, 2.0 mL/min; restrictor temperature, 50°C; solid-phase trap, 50/50 (w/w) Porapak Q/Glass Beads; liquid tandem trap, methanol; liquid tandem trap volume, 5 mL; collection temperature (solid-phase), 40°C, desorption temperature (solid-phase), 40°C; solid-phase rinse solvent, 2.0 mL methanol; solid-phase rinsing volume, 1.0 mL/min; static time, 3.0 min; dynamic time, 25.0 min (total of three dynamic ministeps); static time during trap rinsing, 2.0 min. Solid-phase trap rinsed directly into tandem liquid trap following each dynamic step.

Table III. SFE Reproducibility for Lovastatin (10 mg) from MEVACOR Tablets with 15% (1.0% [v/v] IPA) Methanol-Modified CO_2

Tablet	Recovery (%)
1	98.4
2	101.0
3	98.7
4	98.7
5	100.5
Average recovery (%)	99.5
RSD	1.2

SFE conditions: CO_2 pressure, 400 atm; oven temperature, 40°C; liquid flow rate, 2.0 mL/min; restrictor temperature, 50°C; solid-phase trap, 50/50 (w/w) Porapak Q/Glass Beads; liquid tandem trap, methanol; liquid tandem trap volume, 7 mL; collection temperature (solid-phase), 40°C; desorption temperature (solid-phase), 40°C; solid-phase rinse solvent and volume, methanol, 5.0 mL; solid-phase rinsing volume, 2.0 mL/min; static time, 3.0 min; dynamic time, 25.0 min (total of three dynamic ministeps); static time between dynamic steps: 2.0 min. See Table II for SFC conditions.

Phase D

A MEVACOR tablet containing 10 mg of lovastatin was crushed, placed in an extraction vessel filled with cotton, and extracted under conditions similar to those described in Phases A–C. The total extraction time was extended to 87 min (17 min total static time, 70 min total dynamic time) for 5% (v/v) methanol-modifier with and without IPA (Figure 5). Because the additive concentration in methanol had no statistical effect on recovery, 1% (v/v) IPA was chosen. An overall recovery of only 84% was achieved with 5% (v/v) (1.0% [v/v] IPA) methanol-modified CO_2 within an extraction time of 40 min (dynamic); however, 106% was recovered within 70 min (dynamic). A MEVACOR tablet was also extracted with 5% methanol-modified CO_2 (e.g., no IPA), and only 74% was recovered within 70 min (dynamic). The advantages of the addition of IPA as an additive when extracting from the MEVACOR tablet were clearly shown.

Although quantitative lovastatin recoveries from MEVACOR were achieved with 5% (v/v) methanol (1% [v/v] IPA), the time required for the extraction was 87 min (17 min total static time, 70 min total dynamic time). A dynamic extraction without trap-rinsing between dynamic ministeps as well as an extraction time of approximately 30 min was desired for the final optimized SFE method. Similar to the previous studies, extraction profiles consisting of alternating static and dynamic steps with trap-rinsing in between each dynamic step were performed in order to compare overall extraction recoveries achieved and time needed versus the various modifier and additive percentages. A modifier percentage of 10% (v/v) methanol with and without IPA was next investigated (Figure 6). Overall extraction recoveries (one replicate) of 95 and 88% were achieved with 10% methanol with and without IPA, respectively, but the time needed was 50 min (six dynamic ministeps). Further attempts were made to increase the extraction recovery to 100% and to reduce the time needed to approximately 30 min. Therefore, 15% (v/v) methanol with and without IPA was next investigated (Figure 7). Once again an enhancement was observed when IPA was employed, and 102 and 91% were recovered (one replicate) with and without IPA, respectively, and in this case, the extraction time needed with the IPA was 35 min (static and dynamic).

In the belief that an optimized method had been developed, five MEVACOR tablets were then extracted with 15% (v/v) methanol with 1% (v/v) IPA. The extraction method consisted of three dynamic ministeps, and a 2-min static time was added between each dynamic step to mimic trap-rinsing, as was used when constructing the previous extraction profiles. In this case, the solid-phase trap was not rinsed until the 35-min extraction was completed. Average recovery percentages (five replicates), standard deviations, and relative standard deviations (RSDs) are found in Table III. It can be seen that 10 mg of lovastatin per tablet was fully recovered (99.5%) from the MEVACOR tablets with an RSD of 1.2% with 15% (v/v) (1% [v/v] IPA) methanol-modified CO_2 within 35 min. As compared with the traditional liquid extraction procedure (Table II), the SFE method has been shown to be very advantageous. The use of acetonitrile and buffer has been eliminated, and solvent consumption has been reduced from 95 mL to merely 17.5 mL of methanol used as modifier (7.5 mL), a tandem liquid trap (5 mL), and solid-phase rinsing (5 mL). Also, many laborious and time-consuming steps performed in the liquid extraction such as the addition of buffer and acetonitrile, mixing, sonicating, and cooling steps have been eliminated. As compared with the traditional liquid extraction procedure, the extraction time was reduced from over an hour to merely 35 min by using SFE. Simply all that is required for the SFE method is crushing the tablet, placing it in the extraction vessel, and performing the one-step extraction.

Conclusion

The subcritical fluid extraction of lovastatin from MEVACOR tablets has been demonstrated. Extractability was shown to be dependent on modifier concentration and additive type. IPA was believed to be the most successful additive because of its ability to displace adsorbed lovastatin from the acidic tablet matrix sites versus methanol-modified CO_2 alone. For a series of five MEVACOR tablet extractions, 99.5% recoveries (1.2% RSD) were achieved with 15% (v/v) (1.0% [v/v] IPA) methanol-modified CO_2.

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