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MICROENCAPSULATION OF MATRICINE BY A DEHYDRATING LIQUID AND ASSESSMENT OF ITS RETENTION

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ABSTRACT

Matricine of flowers of cultivated *Matricaria chamomilla L.* was isolated and identified by TLC, IR, UV and ^{1}H-NMR and quantified by HPLC. One of the lipophylic materials of this plant (matricine) has been used as antispasmodic and anti-inflammatory. Retention of matricine by microencapsulation technique was one of the objectives of this study. Encapsulation was carried out by cold dehydrating liquid method and effects of the various process parameters on retention of the matricine were evaluated. To achieve high retention values it was necessary to employ low core to shell material ratio, high solid concentration, high viscosity of the emulsion continuous phase, the use of absolute ethanol as desiccant, short contact times between capsules and desiccant, and low air pressure in the formation of microcapsules. Results suggested that the process might be much more efficient if continuous coextrusion of the emulsion and desiccant were used.

Keywords: Microcapsules, Microencapsulation, Cold dehydrating liquid method, Retention of matricine, Acacia, *Matricaria chamomilla L.*

INTRODUCTION

Attention to herbal medicine is increasing nowadays and as a result manufacturers try to prepare more effective dosage forms. Volatile oils are the most important and unstable chemical constituents of herbs with therapeutic effects (1). However microencapsulation of such substances in polymeric shell could offer a solution to these drawbacks (2).

Many kinds of polymers have been used by a number of procedures for encapsulation of volatile oil droplets including coacervation phase separation, solvent evaporation, atomization and so on (3).

The choice of any method depends on the drug physicochemical characteristics and the desired final results (4). For the purpose of this study since matricine is volatile and water insoluble, perhaps the most appropriate method is cold dehydrating liquid, whereby liquid droplets "core" are entrapped in a continuous film of polymer "wall" (5). This method involves two major unit operations: emulsification and dehydration (5).

In the present study, *Matricaria Chamomilla L.* flowers which are rich in various lipophylic and hydrophylic materials were used (1). Matricine is a lipophylic substance with anti-inflammatory activity, which is more effective than its derivatives such as chamazulene. The aims of this study were: reducing of evaporation, leaching and solidifying of liquid matricine, stabilizing against environmental degradation (light, air, humidity, etc), and masking its odor.

MATERIAL AND METHODS

Material

Matricaria chamomilla L. flowers collected from the botanical garden of school of pharmacy and pharmaceutical sciences, Isfahan, Iran in April on May 1997. Chloroform, methanol, acetonitril, dichloromethane, ethylacetate, acetone, absolute ethanol, isopropanol, potassium hydrogen carbonate, silicagel 100, silicagel GF_{254} and some other chemicals used in this study were purchased from Merck Chemical Company (Germany). Methanol and Acetonitril were HPLC grade and other chemicals were analytical grades.

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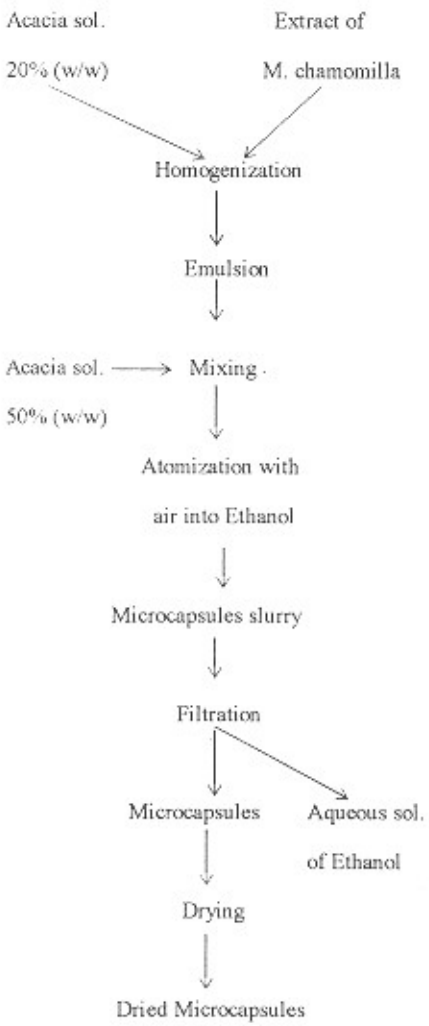
Acacia powders were supplied by Evans Medical Supplies, LTD (England).

Instrumentation

The HPLC (Jasco, 970/975, with two pumps, model PU-980, 20 µl loop and Jasco/JMBS LC Net II control, software Borwin and µ Bondapak C18 columns, Jasco Company, Japan), IR-Spectrophotometer (1420, Perkin Elmer, Germany), UV-Vis spectrophotometer (550 SE, Perkin Elmer, Germany), NMR (400, Varian, USA), Spray Drier (SD-05, Lab Plant, England), homogenizer (L4R, Silverson, England) and light microscope (HM-LAUX3, Leitz, Germany) were used in this study.



Scheme 1. Flow chart of the cold dehydration of microencapsulation process

Methods

Matricine of *Matricaria chammomilla L.* flowers was extracted by chloroform, and then the extract was mixed with petroleum ether to separate the non polar portion. The petroleum ether layer was mixed with potassium hydrogen carbonate solution to separate the matricine. After extraction of the aqueous layer by ether, matricine was crystallized and identified by TLC, UV (6,9), IR (8) and ^{1}H-NMR methods (6,9). TLC was performed on silicagel GF_{254} percoated plates (Merck), using some pure sample (matricine), that was obtained from fresh flowers of *Matricaria chamomilla L.* by stahl's method (6). The mobile phase was chloroform:ethylacetate (5:1), and spots were detected by UV at 254 nm, and then R_f were determined (7).

Core material determination

The amount of matricine was determined by HPLC. The mobile phase consisted of: acetonitril, methanol and water (12:38:50). In this study programming was as follows: Detector was UV and λ_{max} was 243 nm, rate of mobile phase was 1ml/min, and injection volume was 20 µl for each run (8).

Microencapsulation

The flow sheet of the preparation process has been shown in scheme 1. Emulsions were prepared by homogenizing one gram of concentrated extract of *Matricaria chamomilla L.* in a 20% (w/w) acacia solution, by homogenizer, until an emulsion of globules smaller than 1 micrometer was obtained. The globule size was verified by using a light microscope. The final wall material concentration normally 40% (w/w) was reached by addition of 50% (w/w) acacia solution. Microcapsules were obtained by atomizing the emulsion (20 ml/min) at 1 atm air pressure, using a fluid nozzle sprayer (spray drier), into stirred 90-100% ethanol. The resulting microcapsule slurry was filtered and, then dried.

The ratio of feed material to ethanol was (1:10), the wall material concentrations were 35% and 40% (w/w), the starting ethanol concentration was 100%, and the dehydration contact time was approximately 2 min. Each experiment was repeated three times.

Retention

In the encapsulation process of this study retention is defined as the ratio of core-material in the final dried microcapsules to that in the emulsion (all on a dry weight basis of solids) i.e., R=Core material in microcapsules (g/100g solids)/Core material in emulsion (g/100g solids)

RESULTS AND DISCUSSION

R_f of matricine was equal to that of standard (R_f= 20). All spectra (UV, IR and NMR) were in agreement with matricine reference spectra (9).

It was established that the most suitable solvent for extraction of matricine and other volatile constituents was ethylacetate with the level of 80 percent (Fig 1). In extraction, the best ratio for flowers to solvent was (1:10) (Fig 2).

The effect of desiccant (ethanol) to emulsion ratio on core material (matricine) retention for two concentrations of wall-material in the emulsion continuous phase, 40% and 35% (w/w) have been shown in Fig 4. For 10:1, ethanol-to-emulsion ratio, the retention was 22% for 40% acacia and 15.5% for 35% one. Lowering the ratio below the (10:1) caused reduction in retention. Although a higher than 10:1 ration decreased the moisture content in the formed microcapsules, it did not affect retention very much.

The effects expressed above were related to the driving force for mass transformation between the dehydrating capsule and ethanol. High ratios of ethanol to emulsion assured rapid drying and fixation of protective crust that prevented extensive leaching of core material. Higher solids concentration in the emulsion also enhanced rapid crust formation. The higher viscosity of emulsion at higher solids concentration may also play a role in minimizing core material leaching and lead to better microstructural properties of the microcapsules formed (10).

As expected, reducing ethanol concentration in the desiccant had a similar effect to that of reducing the desiccant to emulsion ratio, which is illustrated in Fig 4. A sharp decrease in retention was observed below 95% ethanol and below an initial ethanol concentration of 90%, sufficient dehydration could not be achieved, and the particles agglomerated into a sticky, useless mass. Losses for matricine were mainly due to leaching and evaporation because of high vapor pressure.

Retention in the process was dependent on the initial core material concentration (on a dry basis) in the emulsion sprayed into ethanol which is similar to the study of microencapsulation of paprika oleoresin and aromatic esters (10).

It is reported that at higher core material concentration, the curves deviated more from the 100% retention line. This deviation was more evident where poorer desiccant and more soluble or volatile core materials were employed (Fig 5). Comparison of the retention of paprika oleoresin (10) with the 100% retention line indicates the loss due to leaching and evaporation. Comparison with retention of paprika oleoresin and methyl anthranilate indicates volatility differences. From these findings it can be concluded that matricine will be lost like methyl anthranilate. Retention of matricine is quite sensitive to wall-material concentration in the continuous phase of the emulsion (10). A similar observation is reported (11) for retention of volatiles in freeze-drying. This group showed that increasing the solid concentration enhanced retention up to a limit, which was dependent to the nature of the solids (wall material), core material (volatile), and initial concentration of core material in the emulsion. Higher solids concentration led to a more rapid crust formation. Higher viscosity at a given constant air pressure led to the formation of larger drops by the nozzle. Larger drops have smaller surface area to volume ratios and this also reduced mass transfer between the capsule and ethanol. With acacia, the solids could not be increased above 40% in this process (because of high viscosity), thus an asymptotic retention value could not be reached.

Decreases in retention by increase in amounts of core material could happen by two possibilities. First, at higher core-to-shell material ratios, there was less protection afforded for each core material droplet within the microcapsule. Second, the core material solubilized in the continuous phase may interfere with the drying process and leads to an inferior solid shell (10).

Prolonged contact between the microcapsules and the desiccant produced drier capsules, increased leaching of core material, and lowered retention, which is illustrated in Fig 6. From this and similar experiments, it is obvious that very short contact times were needed to achieve the best results. In

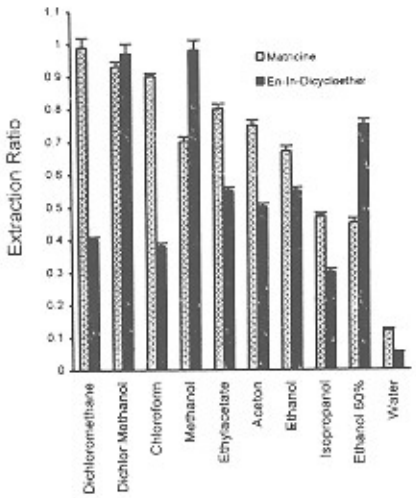


Figure 1. Effect of different solvents on the extraction of Matricine and Dicycloethers.

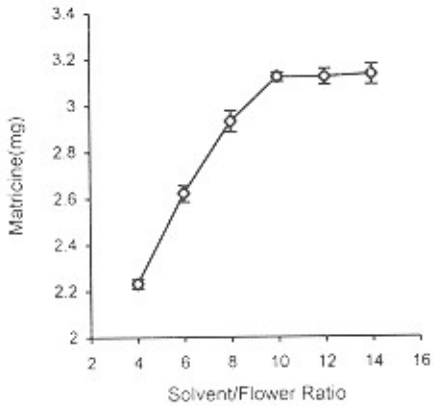


Figure 2. Effect of solvent to flower ratio on the extraction of matricine.

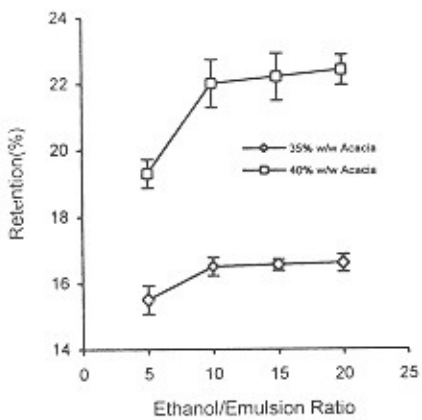


Figure 3. Effect of ethanol to emulsion ratio on the retention of matricine.

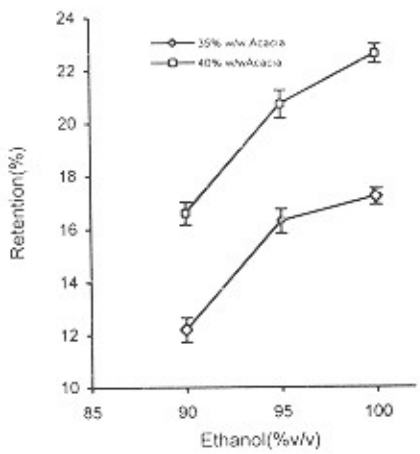


Figure 4: Effect of ethanol concentration on the retention of matricine (Ratio of ethanol to emulsion is 10:1).

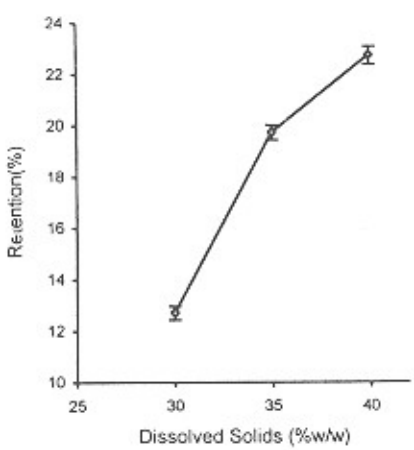


Figure 5. Effect of dissolved solids concentration on the retention of matricine.

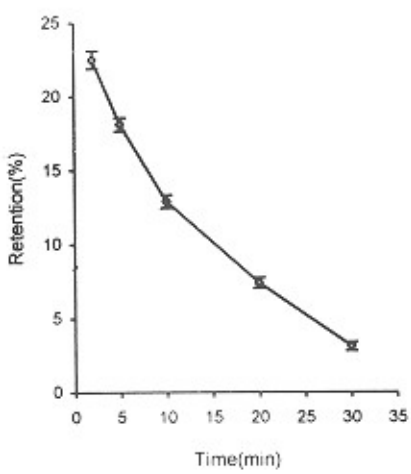


Figure 6. Effect of contact time with ethanol on the retention of matricine.

the batch experimental set-up the total minimum contact time was 2 min. This time can be reduced if a continuous drying process is used.

As reported before (10), the effect of relative humidity on retention of core material during storage (retention is defined here with respect to the product at the beginning of storage) is quite gradual until about 0.75 water activity, when a sharp decrease is observed. This sharp decrease in retention results from a high enough water intake by the wall material to make it semi fluid and then, wall fusion and enhanced solublization of core material and diffusion into atmosphere occurred (10).

The influence of different factors on the retention of matricine, such as solid concentration and viscosity of the emulsion continuous phase, ethanol to emulsion ratio, ethanol concentration, contact time with ethanol, the yield of microparticles and the encapsulation efficiency of matricine in microparticles were evaluated by ANOVA (α= 0.05).

Conclusion

This study indicates the set of parameters that should be selected to design an optimal low temperature microencapsulation process for various core-material (volatile and non volatile) using ethanol as dehydrating agent. For maximum core-material retention some of the relevant conditions are, the use of absolute ethanol, short microcapsule-desiccant contact time, low core to shell material ratio, low air pressure in the spraying process, high concentration of the shell material and drying at normal temperature.

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