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Compatibility studies of lapachol with pharmaceutical excipients for the development of topical formulations

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Abstract

Among naphthoquinones, lapachol and many heterocyclic derivatives have been investigated during the past years, mainly due to their antibacterial, antifungal and anticancer activities. Assessment of possible incompatibility between an active component (i.e. lapachol) and different excipients along with the evaluation of thermal stability are crucial parts of a normal study prior to the final formulation setting of a medicine. DSC study was used as an important and complementary tool during pre-formulation to determine the compatibility of drug-excipients with the purpose of developing a lapachol gel-cream formulation. The DSC curves of lapachol (Lpch) and binary mixtures with excipients (propylparaben, methylparaben, edetic acid, isodecyl oleate, trietanolamine, glyceril monoesterate, cetiol, glicerin, carbopol, cetomacrogol and mineral oil) were obtained. The results showed that Lpch only exhibited interaction which could influence the stability of the product in the binary mixtures of Lpch/methylparaben, Lpch/cetostearyl alcohol and Lpch/glyceryl monostearate. There was a significant shift or absence of lapachol melting peak in both cases. Photovisual DSC was used to confirm the results obtained with conventional DSC because it is a more sensitive method.

Keywords: Lapachol; DSC; Gel-cream; Compatibility studies

1. Introduction

Lapachol is a naphthoquinone (2-hydroxy-3-(3-methyl-2-butenyl)-1,4-naphthoquinone) extracted from Pau d'arco (Family Bignoneaceae). A great spectrum of therapeutic activities has been attributed to lapachol or its derivatives, such as prevention of cercarial skin penetration of *Schistossoma mansoni* [1,2], trypanosomicidal [3], antiinflammatory [4] and antineoplasic activity; antimalarial against erytrocytic stages of *Plasmodium falciparum* [5] and against enteroviruses [6–8].

Assessment of possible incompatibility between an active component (i.e. lapachol (Lpch)) and different excipients along with the evaluation of thermal stability are crucial parts of a normal study prior to the final formulation setting of a topical dosage form [9]. Excipients are known to facilitate the administration and release of active components as well as to protect them from the environment. Excipients are considered pharmaceutically inert, but physical and chemical interactions with an active component are possible [9].

The development of pharmaceutical formulations requires previous knowledge of the physicochemical properties of the drug, excipients and analytical instrumentation that can be applied efficiently with swift results. Thermal analysis is used in the pharmaceutical industry as a rapid technique that is appropriate for quality control and the development of new medicines [10–12]. In particular, differential scanning calorimetry (DSC) allows for the evaluation of possible incompatibilities by revealing changes in appearance, shift or disappearance of melting or other exothermic processes, and/or variations in the corresponding enthalpies of reactions [13,14]. However, differences in the DSC curves of binary mixtures compared to the individual components

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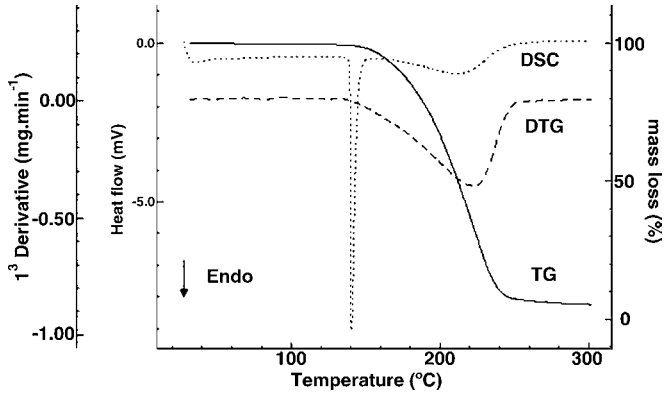

Fig. 1. DSC and TG/DTG curves of lapachol obtained in dynamic nitrogen atmosphere (50 ml min^{-1}) and at a heating rate of 10 °C min^{-1}.

may arise for reasons other than chemical incompatibility [9].

This study evaluated the thermal stability of lapachol (Lpch) and mixtures of Lpch/excipients by conventional DSC and photovisual system, and excipients used in the prototype formulation of gel-creams were selected. Gel-creams, which are relatively new preparations, are oil–water systems containing gum as a thickener in the external aqueous phase. They can be made from a gelified preparation in which an emulsifying agent is incorporated in conjunction with an oily phase, which can be a mineral or vegetable oil [15]. The use of a thickener increases the viscosity of the continuous phase, reducing the frequency of collision and coalescence of drops, thereby increasing the stability of the preparation. Carbopol is commonly used as a thickener in these types of formulations as its structure contains a small lipophilic in addition to a large hydrophilic portion, which act as oil in water emulsifiers. The lipophilic part of this polymer is located on the oil–water interface, and the hydrophilic portion swells in the presence of water to form the gel that envelops the tiny drops of oil. This way, it is possible to obtain more stable formulations that do not require a large quantity of oil phase to achieve a good level of viscosity.

2. Experimental

2.1. Materials

Lapachol was donated by Laboratório Farmacêutico do Estado de Pernambuco (LAFEPE). Propylparaben, methylparaben, edetic acid and isodecyl oleate were purchased from Henrifarma. Trietanolamine and ethanol were obtained from Merck. Glycerilmonoesterate was provided by Croda, and the other excipients were acquired from Galena. The mixed samples consisted of equal weights of lapachol, and each excipient was individually weighed into amber glass vials to give composite weights of 20 mg. The physical mixtures were prepared using an agata mortar with pestle for approximately 10 min.

2.2. Measurements

The DSC curves of the lapachol drug, excipients and binary mixtures were obtained with a Shimadzu calorimeter, model DSC-50, coupled to a photovisual system consisting of an Olympus microscope connected to a Sanyo camera, model VCC-D520, under a nitrogen flow of 50 ml min^{-1}, at a heating rate of 10 °C min^{-1}, up to 400 °C. These parameters were based on the conventional laboratory program and appropriate method. Transition temperatures were recorded from a plot of heat flow versus temperature (30–400 °C). The peak temperature (T_p) of phase transitions was determined from the DSC curve with Tasys software from Shimadzu. Reaction heat was determined by using the area of the peaks between the onset temperature (T_o) and the end-temperature from the DSC curve. Samples (2.0 mg) were weighed to the nearest ±0.01 mg, and sealed aluminum crucibles were used. The images were captured by means of DSC coupled to the photovisual system under similar conditions of conventional DSC. Medium and standard deviation (S.D.) values were determined from triplicates of DSC curves. The DSC cell was calibrated with indium (mp 156.6 °C; $\Delta H_{fus.}$ = 28.5 J g^{-1}) and zinc (mp 419.6 °C). TG/DTG curves were obtained with a thermobalance model TGA 50 (Shimadzu) in the temperature

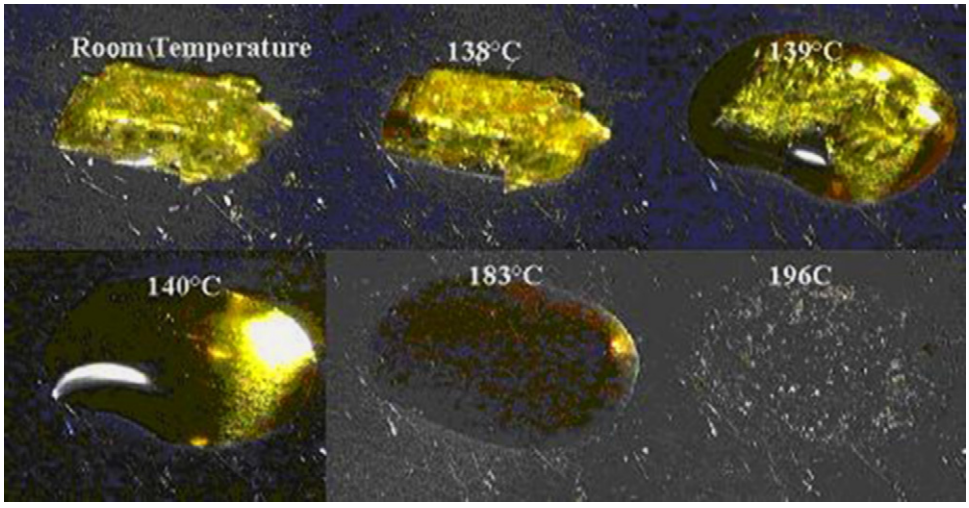

Fig. 2. DSC photovisual of lapachol: (a) room temperature, (b) 138 °C, (c) 139 °C, (d) 140 °C, (e) 183 °C and (f) 196 °C.

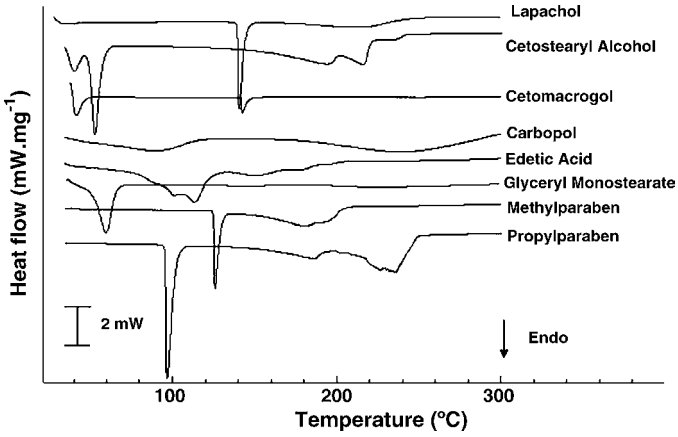

Fig. 3. DSC curves of lapachol and solid excipients obtained in dynamic nitrogen atmosphere (50 ml min^{-1}) and at a heating rate of 10 °C min^{-1}.

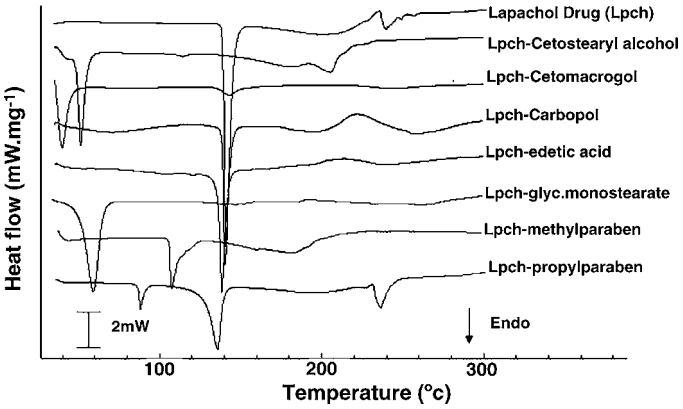

Fig. 4. DSC curves of binary mixtures of lapachol and excipients obtained in dynamic nitrogen atmosphere (50 ml min^{-1}) and at a heating rate of 10 °C min^{-1}.

range 25–900 °C, using platinum crucibles with ~3 mg of samples, under dynamic nitrogen atmosphere (50 ml min^{-1}) and at a heating rate of 10 °C min^{-1}.

3. Results and discussion

DSC curves of lapachol showed a sharp endothermic peak that corresponded to melting of Lpch at a temperature onset of 138.5 °C ($\Delta H = 105.6$ J g^{-1}). After this event, thermal decomposition of material was observed at a temperature of approximately 141 °C (Fig. 1). The thermal behavior of Lpch using DSC photovisual can be seen in Fig. 2. TG/DTG curves indicate that the thermal decomposition process of Lpch occurs in one stage of mass loss in the temperature range of 141–260 °C.

The DSC curves of the isolated solid excipients and binary mixtures are presented in Figs. 3 and 4. The melting endotherm

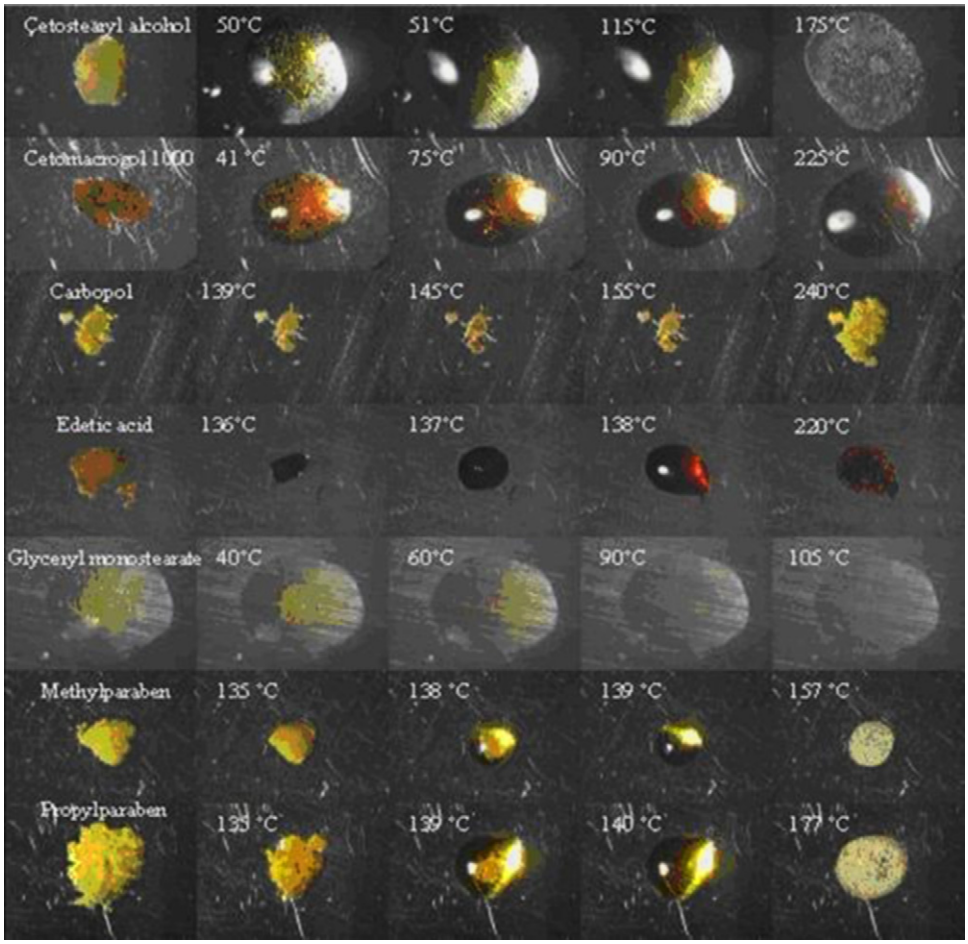

Fig. 5. DSC photovisual of binary mixture of lapachol and excipients obtained in dynamic nitrogen atmosphere (50 ml min^{-1}) and at a heating rate of 10 °C min^{-1}.

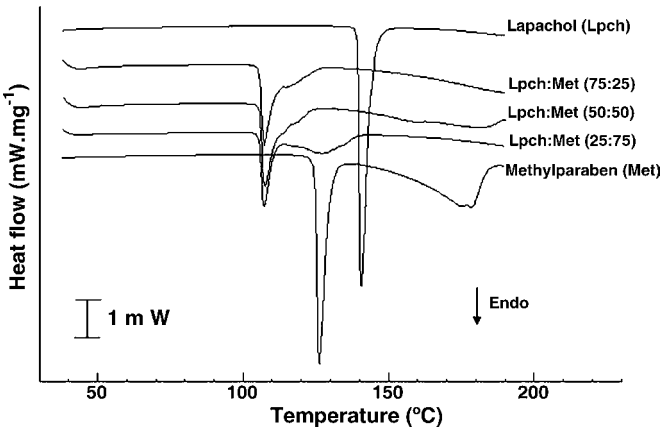

Fig. 6. DSC curves of Lpch:methylparaben mixtures in ratios (w/w) of 100:0, 75:25, 50:50, 25:75, and 0:100.

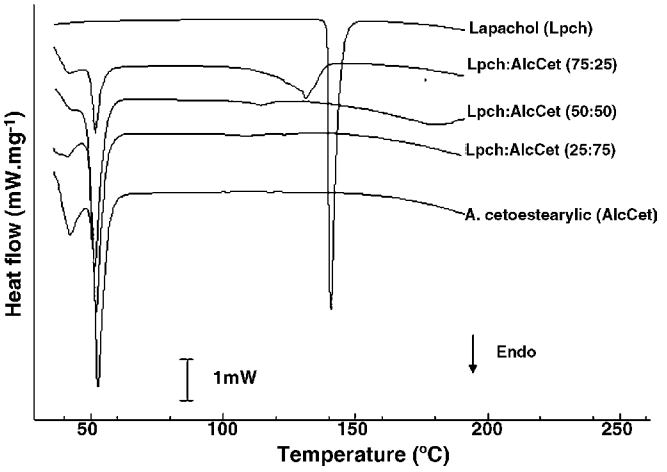

Fig. 7. DSC curves of Lpch–cetostearyl alcohol mixtures in ratios (w/w) of 100:0, 75:25, 50:50, 25:75, and 0:100.

of the drug was well preserved in the majority of cases. However, there were slight changes in the peak shape with little broadening or shifting towards the lower temperature. The DSC curves of the physical mixtures of Lpch and edetic acid, propy-

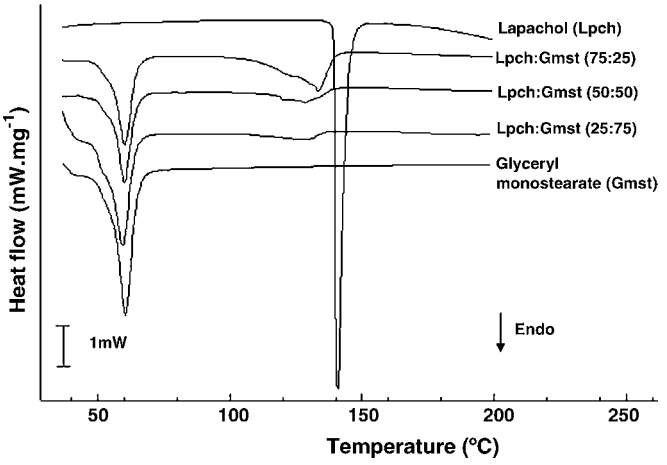

Fig. 8. DSC curves of Lpch–glyceryl-monostearate mixtures in ratios (w/w) of 100:0, 75:25, 50:50, 25:75, and 0:100.

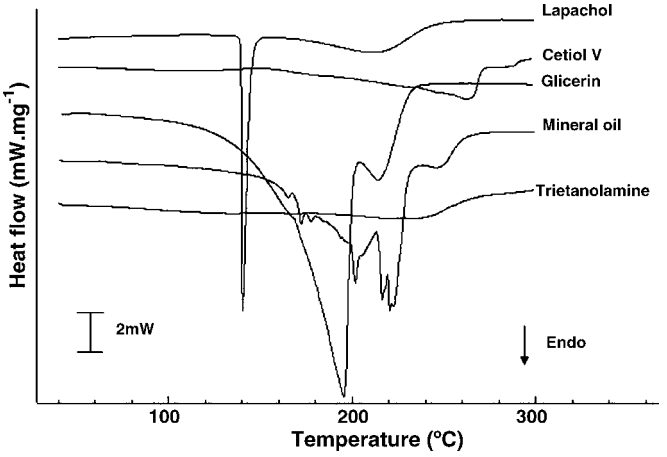

Fig. 9. DSC curves of lapachol and liquid excipients obtained in dynamic nitrogen atmosphere (50 ml min^{-1}) and at a heating rate of 10 °C min^{-1}.

lparaben, carbopol and cetomacrogol 1000 can be considered to be the superposition of the DSC curves of the two individual components. These results show that physical interactions of components did not occur within the mixture. Photovisual DSC was used to confirm the results obtained by conventional DSC (Fig. 5).

In the thermal curve of the binary mixture of Lpch/methylparaben, there was an appreciable downward shift of the drug peak temperature, which can be indicative of some drug-excipient solid interaction. DSC curves showed that the peak at around 139 and 124 °C, which was observed for Lpch and methyparaben, respectively, disappeared in the eutectic mixture (Fig. 4). However, a new peak at around 104 °C, which was not observed for Lpch and methyparaben, appeared in the eutectic mixture. Binary mixtures of Lapch/methylparaben in ratios (w/w) of 100/0, 75/25, 50/50, 25/75, and 0/100, respectively, were prepared for thermal analysis (Fig. 6). Scott et al. [16] used DSC to study the eutectic formation by a model non-stereoidal anti-inflammatory drug with a range of seven terpene transdermal permeation enhancer. They found that DSC studies on the ibuprofen:thymol mixtures indicated that they

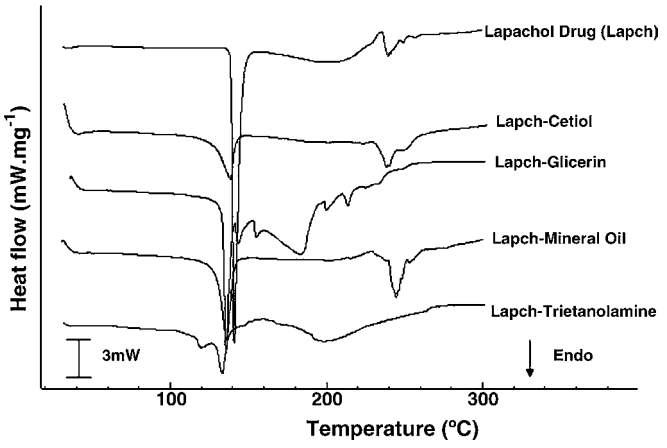

Fig. 10. DSC curves of binary mixtures of lapachol and excipients obtained in dynamic nitrogen atmosphere (50 ml min^{-1}) and at a heating rate of 10 °C min^{-1}.

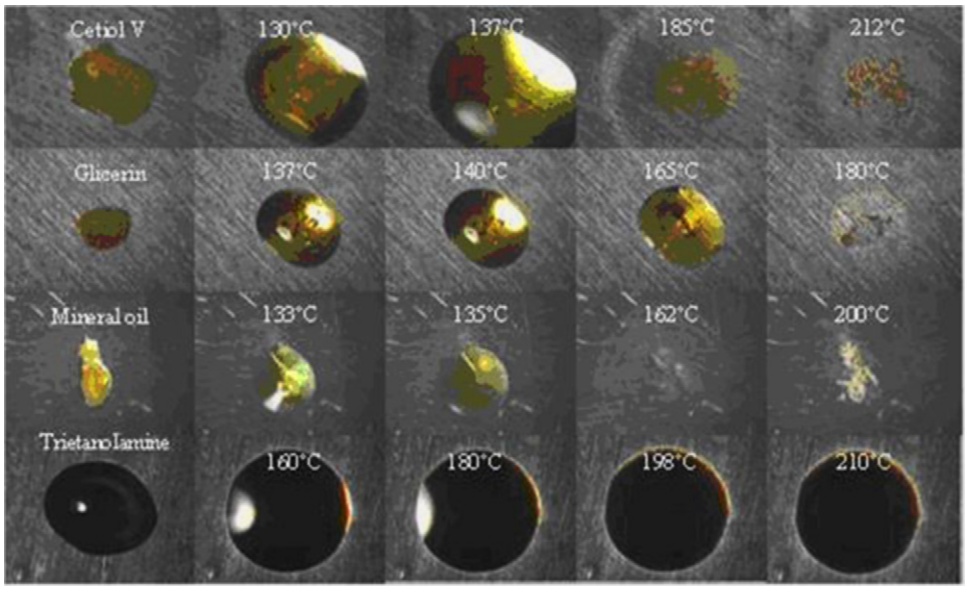

Fig. 11. DSC photovisual of binary mixture of lapachol and liquid excipients obtained in dynamic nitrogen atmosphere (50 ml min^{-1}) and at a heating rate of 10 °C min^{-1}.

provide an example of a simple binary eutectic system with no evidence of solid solution formation. A similar behavior was observed in this study for the Lpch:methylparaben mixture.

In the DSC curves of the Lpch–cetostearyl alcohol and Lpch–glyceryl-monostearate binary mixtures, the endothermic peak of drug was broadened and shifted to a lower temperature (Fig. 4). This occurred because cetostearyl alcohol and glyceryl-monostearate showed a sharp endothermic peak at 53 and 60 °C, respectively, corresponding to melting. Instead, DSC photovisual showed that this effect was mainly due to the partial dissolution of drug in melted excipients (Fig. 5). The disappearance of the melting peak of drug is indicative of a strong interaction, but not necessarily corresponding to incompatibility. The modification in drug thermal behavior was somewhat intense, depending on the different degree of drug solubility in melted component and, for the same excipients, in the mechanical treatment sustained by the sample [17]. In fact, a similar effect was observed for other drugs, such as naproxen [18], piroxicam [19], ketoprofen in mixtures with various PEGs and was attributed to drug dissolution in the melted polymer [20].

Figs. 7 and 8 show the DSC curves of the binary mixtures of Lpch/cetostearyl alcohol and Lpch/glyceryl monostearate in different proportions. As one can see, the DSC scans showed small melting peaks even with such low drug content. These interactions seemed to be very effective in physical mixtures containing 50 and 25 wt% Lpch, and its melting point was not possible to be detected. By applying DSC to look at interaction between felodipine and excipients, Bikiaris et al. [21] explored the physical state of a variety of new solid dispersion of hesperetin or felodipine. In the case of dispersions in PEG as well as the physical mixtures, DSC, though fast rates were used, could not detect the presence of crystalline drug. This was not because the drug was amorphous, but because of the increased solubility of the drugs in the liquid PEG at elevated temperatures.

Fig. 9 presents the DSC curves of mineral oil, isodecyl oleate, trietanolamine and glycerine. It was noted that the excipients were thermally stable up to approximately 150 °C. After this temperature, processes of volatization and thermal decomposition of these materials occur. The thermal behavior of physical mixtures of lapachol and excipients correspond to the superposition of the DSC curves of the two individual components. Lpch demonstrated compatibility with all the liquid excipients studied, as shown in Fig. 10. DSC Photovisual analysis showed that the viscosity of liquid excipients decreases with temperature, therefore, their volume increases (Fig. 11).

4. Conclusion

Thermoanalytical methods were used extensively to evaluate the physical properties of drugs, including melting and vaporization temperatures and with the corresponding enthalpies, glass transitions, vapor pressures, as well as to study the compatibility and stability of the components of pharmaceutical preparations. Possible incompatibilities between active components and different excipients were observed by using DSC. However, differences in the DSC curves of binary mixtures and individual compounds do not necessarily correspond to incompatibility. For further confirmation of a substantial incompatibility, it is important to associate with other analytical techniques. Conventional and photovisual DSC curves of Lpch and excipients showed interactions with glyceryl monostearate, methylparaben and cetostearyl alcohol.

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