A Fourier Transform Infrared Spectroscopy Study of the Effect of Temperature on Soy Lecithin-Stabilized Emulsions

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ABSTRACT: The effect of temperature on soy lecithin-stabilized emulsions was studied using Fourier transform infrared spectrosocpy (FTIR). Oil-in-water (o/w) 4% (wt/vol) soy lecithin emulsions were prepared in 6% (vol/vol) medium-chain triglycerides and 94% (vol/vol) water using a two-stage homogenizer set at a pressure of 3000 psig. Three types of emulsions were used in this study: emulsions containing Lecigran and Lecimulthin as emulsifiers and a control emulsion, with no emulsifier added. After preparation, the emulsions were cooled to 4°C, held at this temperature, and spectra were collected after 1 h. The emulsions and reference water were raised to room temperature (22°C) and held at that temperature for 1 h and the spectra collected. The temperature was raised 15°C over the temperature range of 22 to 82°C, and spectral data were collected similarly. The four regions used for this determination in the subtracted spectra of the emulsion were those contributing to –OH vibration, –CH_2 stretching, H–O–H bending vibrations, and P=O, C–O–C, and P–O–C vibrations. The control emulsion was greatly affected at temperatures other than room temperature. This was due to the lack of lecithin as an emulsifier, resulting in a destabilization of the emulsion with temperature increases. The vibrational peaks for the emulsion containing Lecimulthin were found to be lower than those for the emulsion made with Lecigran due to greater water bonding. The control had the highest peaks at the –OH regions because of reduced interaction at the oil–water interface. Both of the emulsions with phospholipids remained stable throughout the temperature range. FTIR is a potentially powerful tool that could be used in the rapid determination of emulsion stability in food systems by measuring emulsifier–water interactions.

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KEY WORDS: Fourier transform infrared spectroscopy, medium-chain triglycerides, oil-in-water emulsions, soy lecithin.

Most food emulsions are subject to processing operations (pasteurization, sterilization, spray-drying, evaporation, freezing, crystallization, and cold storage) that involve a variation in temperature. With some foods (milk, cream liquor, etc.), it is important that the emulsion be stable over a wide range of temperatures, whereas for others (butter, margarine, etc.), the controlled emulsion destabilization is an integral part of the production. It is important to understand the effect of heat on the molecular and colloidal aspects of food emulsions. However, there are few studies on the effect of processing conditions on emulsion properties (1).

Lecithin is a natural emulsifier that has been used in foods for centuries (2). Owing to its polarity, soy lecithin acts as a surfactant and confers emulsifying, viscosity, wetting, and dispersive properties to food emulsions (3). Soy lecithin has been used to study the rheology and stability of phospholipid-stabilized emulsions (4). It migrates to the interface, reducing interfacial tension by wetting. These hydrophilic and dispersive properties of lecithin can be modified by changing the relative concentration of any one component or by altering its chemical structure, thereby causing a change in the physical and chemical properties of the lecithin.

Stability is important in food dispersions, as consumer perception of quality is largely influenced by appearance. A stable emulsion is defined as one in which the inevitable process of separation has been slowed to an extent that it is not of practical importance over 2 or 3 yr, during which time the emulsions are usually handled (5). An emulsion should retain its appearance, viscosity, particle size, and odor during this period of time. Emulsion stability is dependent on the properties of surface components present in the emulsion system.

Fourier transform infrared spectroscopy (FTIR) has been used to determine microstructures formed in phosphatidylcholine (6), water penetration in lipid bilayer membranes (7), and the orientation of synthetic peptides in phospholipid bilayers (8). The partially unrealized potential of the FTIR technique is perhaps responsible for the lack of application in characterizing the wide variety of oil-in-water (o/w) emulsion systems (9).

The objective of this research work was to investigate the use of FTIR for the rapid analysis of the physical chemistry and stability of soy lecithin-stabilized emulsions.

EXPERIMENTAL PROCEDURES

Materials and methods. Medium-chain triglycerides (MCT) were obtained from Abitec Corp. (Janesville, WI). Lecigran 5750 (Riceland Foods, Stuttgart, AR) and Lecimulthin 100 (Lucas Meyer, Decatur, IL) were used in this study. These lecithins are granular deoiled products from soybean lecithin,

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contain no additives, and have a phospholipid concentration of approximately 97%.

The o/w emulsions were prepared with 6% (vol/vol) MCT, 94% (vol/vol) water, and 4% (wt/vol) soy lecithin (four replicate samples were prepared). The MCT and emulsifier were mixed and heated to 70°C with stirring. Distilled water was heated separately to the same temperature. The two phases were introduced into a two-stage 15M lab homogenizer (Gaulin Corporatin, Wilmington, MA) maintained at 3,000 psig pressure and a temperature of 70°C. To prevent separation of oil and water phases in the reservoir, a whisk was used to mix the two phases together. The product was collected and recycled twice. The homogenizer was washed with alkaline and acidic solutions and finally with hot water before and after sample preparation. Approximately 250 mL of the final product was packed in amber bottles and left at room temperature for approximately 48 h. A control emulsion (no soy lecithin) was prepared and consisted of a homogenized mixture of MCT and water, the same weights used for the lecithin-stabilized samples, and stored similarly.

Sample treatment. The emulsions were stored at 4°C for 1 h before collection of FTIR spectra. The samples were then placed at room temperature (~22°C) for 1 h and spectra were collected, after which they were placed in a water bath and heated to 37, 52, 67, and 82°C. After every 15°C increase in temperature, the samples were held for 1 h at this temperature, after which spectra was collected.

FTIR analysis. An Impact 410 FTIR instrument (Nicolet, Madision, WI) was used for spectra collection. Horizontal attenuated total reflectance (HATR) with a standard Ge plate was used. Approximately 0.75 mL of emulsion sample was introduced onto the Ge plate. Typically, 100 scans of sample and background were separately co-added at a nominal resolution of 4 cm^{-1}. The Ge plate was cleaned with ethanol and distilled water between runs.

RESULTS AND DISCUSSION

Figure 1 shows the spectra of the control (A), 4% Lecigran (B), and 4% Lecimulthin (C) samples 48 h after preparation. Each spectrum was examined with reference to the –OH vibration (3,600–3,200 cm^{-1}), $-CH_2$ vibration (3,000–2,800 cm^{-1}), –C=O vibration (1,800–1,600 cm^{-1}), and P=O, C–O–C, and P–O–C vibrations (1,200–1,050 cm^{-1}).

–OH vibration bands. The main vibrational peaks that were used in this determination were those due to H–O–H (water) stretching vibration at 3,400 cm^{-1}; the O–H hydrogen bonded stretching vibration evident at 3,323 cm^{-1}; and the bound X–O–H stretching vibration (where X is C–OH or P–OH groups) at ~3,225 cm^{-1}. Figure 2 shows the spectra of the control (A), 4% Lecigran (B) and 4% Lecimulthin (C) emulsions at 4, 22, 37, 52, 67, and 82°C at the H–O–H vibration region (3,000–4,000 cm^{-1}). Low temperature (4°C) had the effect of breaking the emulsion by destroying the membrane of the emulsifying agent surrounding each globule, thereby causing coalescence. As the temperature was increased from 4 to 22°C, the emulsion spectra were similar to the original emulsions (Fig. 2). A 15°C increase to 37°C showed the same behavior as seen for the emulsion at 4°C, even though the peaks at 37°C were more pronounced. As the temperature was raised, the initial emulsion separation was evident by the bands at 3,390 and 3,225 cm^{-1}. An additional 15°C increase to 52°C gave rise to three peaks at 3,390, 3,300, and 3,225 cm^{-1}, due to the presence of free H–O–H, –OH hydrogen bonded, and X–O–H groups, respectively.

The spectra of lecithin-stabilized emulsions at 67 and 82°C were similar to those at 52°C. However, they were of lower intensity, probably due to water starting to change phase at these higher temperatures by reducing H-bonding. This subsequently reduced the vibrations at the –OH region considerably.

$-CH_2$ vibration bands. The asymmetric (ν_a) and symmetric (ν_s) methylene vibrations were at 2,924 and 2,857cm^{-1}, respectively. The $-CH_2$ bands were contributed by the aliphatic hydrocarbon chains of MCT and the phospholipids in lecithin. Figure 3 shows the variation in the methylene vibration for the control (A), Lecigran (B), and Lecimulthin (C) emulsions with respect to temperature. The vibration for the emulsions containing 4% emulsifier were constant with the temperature increase between 22 and 67°C. This effect was due to the presence of the emulsifier, which appears to hold the emulsion together within this temperature range. Weete *et al.* (10) found that heating lecithin under controlled time and temperature conditions greatly improved its properties as an emulsifier for o/w emulsions. At extreme temperatures (4 and 82°C), there was an increase in the $-CH_2$ vibration, which is consistent with a previous study (11). This explains the increased vibration with increased concentration of $-CH_2$ groups due to reduced emulsifier–oil phase interaction. The oil separation led to an increased adsorption of oil droplets on the Ge surface, hence the increased vibration. The $-CH_2$ vibration of the control emulsion was very inconsistent, showing that the absence of an emulsifier greatly affected this emulsion as temperature was increased. The $-CH_2$ vibration (ν_a and ν_s) was evident at 4 but not at 22°C. There was increased vibration at 4°C caused by a change in the conformation of the methylene chains. Above 82°C, the increase in $-CH_2$ vibration can be attributed to the phase behavior of water as it changed from liquid to vapor. This reduced stability at the w/o interface resulted in the falling out of oil droplets onto the Ge surface, therefore increasing the group vibration. There was increased intramolecular bonding within the MCT matrix, which slightly increased vibration of the group.

Maroncelli *et al.* (12) found that changes in the relative bands could be used to estimate conformational disorder and changes in the packing of solid hydrocarbons as a function of temperature. The contributions of the hydrophobic effect and conformational entropy increase with increasing temperature, and the effects of hydrogen bonding, electrostatic interactions, and van der Waals forces decrease with increasing temperature. This is seen most strongly in the spectra of the control emulsion, whereas in the lecithin-stabilized emulsions, there are no significant changes in this region.

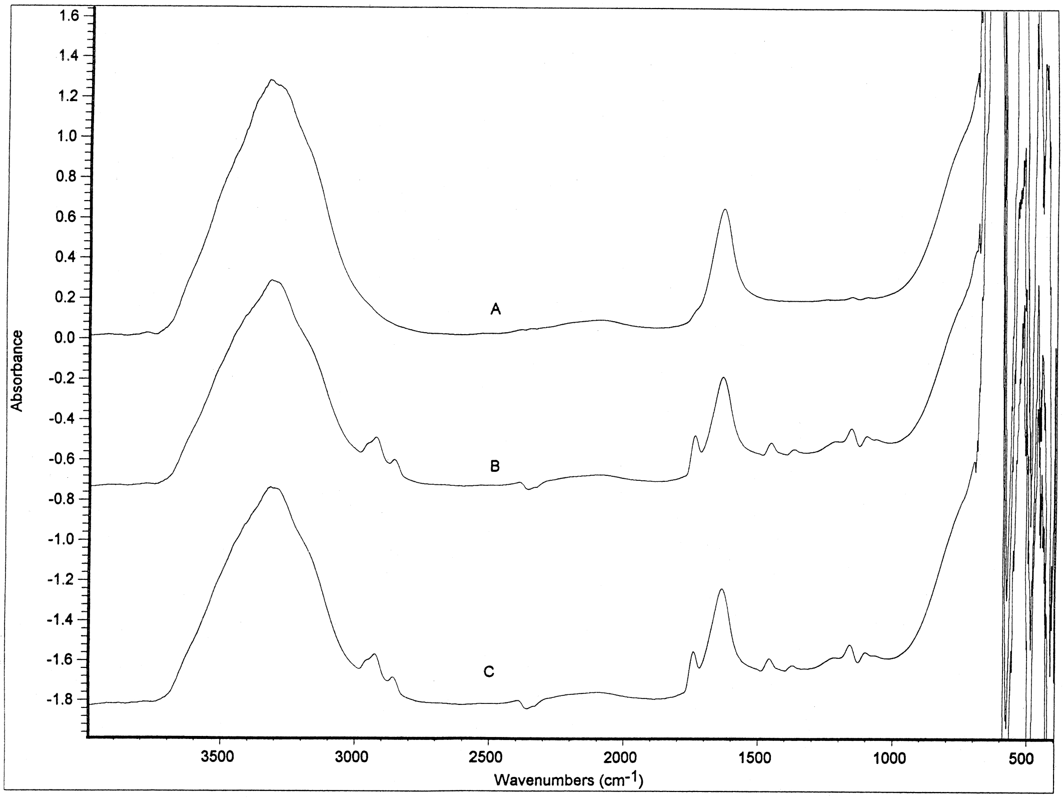

FIG. 1. Fourier transform infrared spectroscopy (FTIR) spectra of control (A), 4% Lecigran (B), and 4% Lecimulthin (C) emulsions after preparation. One hundred scans were co-added at a nominal resolution of 4 cm^{-1} (y is not to scale).

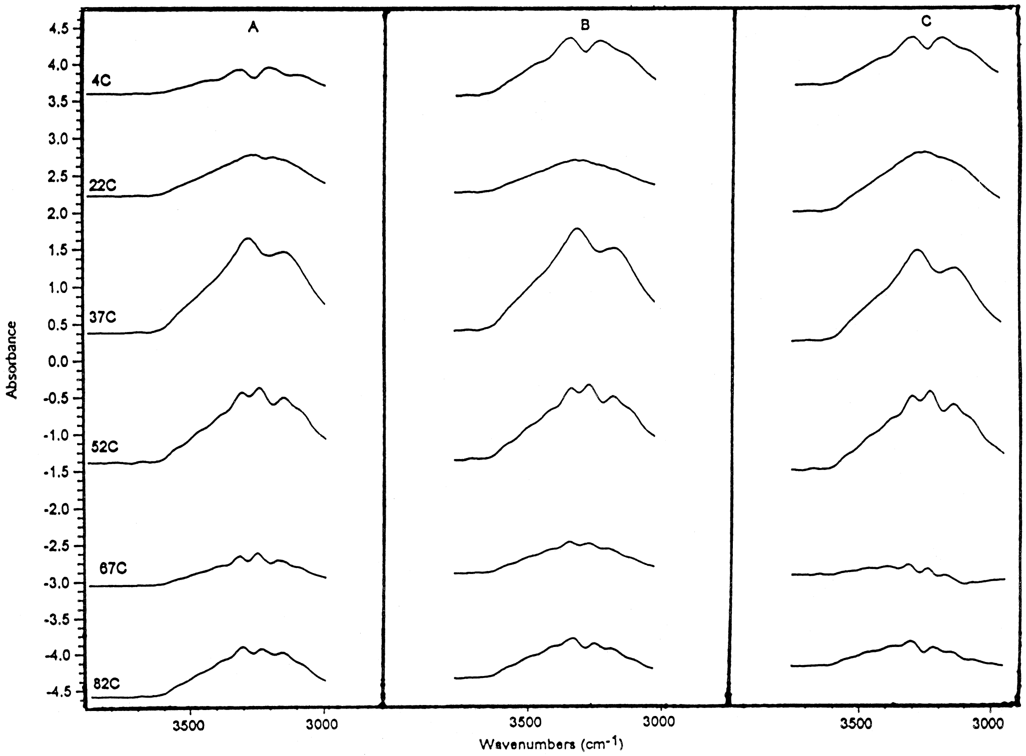

FIG. 2. FTIR spectra of –OH stretching vibration for control (A), 4% Lecigran (B), and 4% Lecimulthin (C) emulsions with increasing temperature. One hundred scans were co-added at a nominal resolution of 4 cm^{-1} (y is not to scale). See Figure 1 for abbreviation.

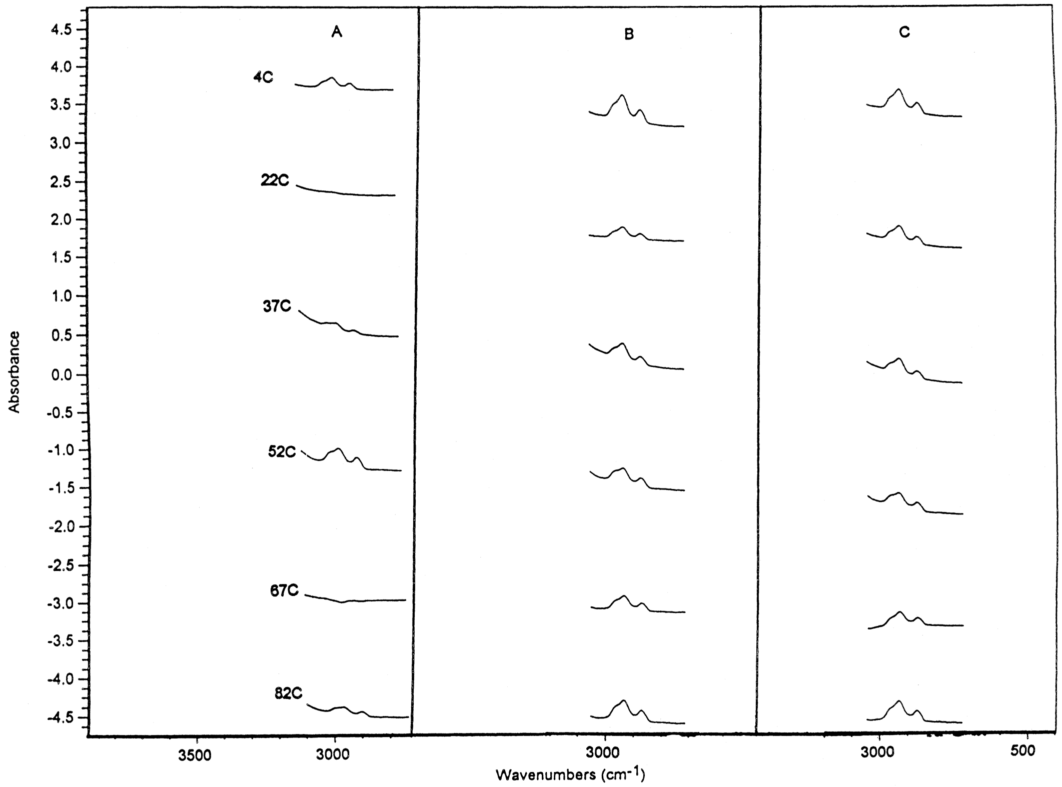

FIG. 3. FTIR spectra of –CH$_2$ stretching vibration for control (A), 4% Lecigran (B), and 4% Lecimulthin (C) emulsions with increasing temperature. One hundred scans were co-added at nominal resolution of 4 cm^{-1} (y is not to scale). See Figure 1 for abbreviation.

–C=O vibration band. The peaks that were used for this determination were at 1,741 cm^{-1} due to carbonyl groups present in the MCT and the lecithin, and at 1634 cm^{-1} due to the H–O–H bending vibration. Significant information can be obtained from the vibrations at these two regions. The shifts in water bands at the –OH vibration region (3,600–3,200 cm^{-1}) and at 1,634 cm^{-1} can be used to determine the state of an emulsion (11). The FTIR spectra for the region 1,800–1,600 cm^{-1} are shown in Figure 4. As temperature was increased from 4 to 82°C, there were very few differences in the lecithin-stabilized emulsions. The largest difference between Lecigran- and Lecimulthin-stabilized emulsions was at 67°C. This can be explained by the fact that Lecimulthin is more hydrophilic, causing an increase in the size of the globules and therefore entrapping more water than Lecigran and reducing the H–O–H bending vibration at 1634 cm^{-1}. The control was highly affected by heat as evidenced by the large differences seen in the FTIR spectra from 4 to 82°C. The absence of emulsifier seems to have affected the emulsion and greatly reduced its stability as the temperature increased, leading to MCT separation and subsequent adsorption onto the germanium. Soy lecithin seems to work by reducing these fluctuations in the amount of oil and water held at any one temperature, thereby keeping the emulsions fairly stable.

P–O–C, P=O, and C–O–C vibration bands. Figure 5 shows the FTIR spectra of the emulsions at the region between 1,200 and 1,000 cm^{-1}. The vibrations at 1,157 and 1,101 cm^{-1} are due to overlapping vibrations of C–O–C and P=O, while that at 1,101 cm^{-1} is due to P–O–C stretching vibrations in the MCT and the phospholipids present in the lecithin. This vibrational region was constant with temperature for the lecithin-stabilized emulsion. However, large differences were found for the control in this region as the temperature was increased from 4 to 22°C and from 52 to 67°C. These changes are due to the absence of an emulsifier and denote a weak emulsion in which oil separates out and gets adsorbed onto the Ge surface.

Soy lecithin-stabilized emulsions were found to be stable under the temperature ranges investigated. However, the control emulsion was found to vary largely depending on the temperature. The peak regions for the group stretching, bending, and wagging vibrations were used to determine the stability of the emulsions, as well as to explain the physical chemistry involved in the emulsion systems. This study shows it is possible to use FTIR to determine the stability of lecithin-stabilized emulsions. The subtraction spectra yield important information at the micro- and macromolecular level in emulsion systems. FTIR is an emerging technique in the study of phase behavior of lecithin in emulsion systems and provides rapid nondestructive analysis of the adsorption behavior in emulsion systems.

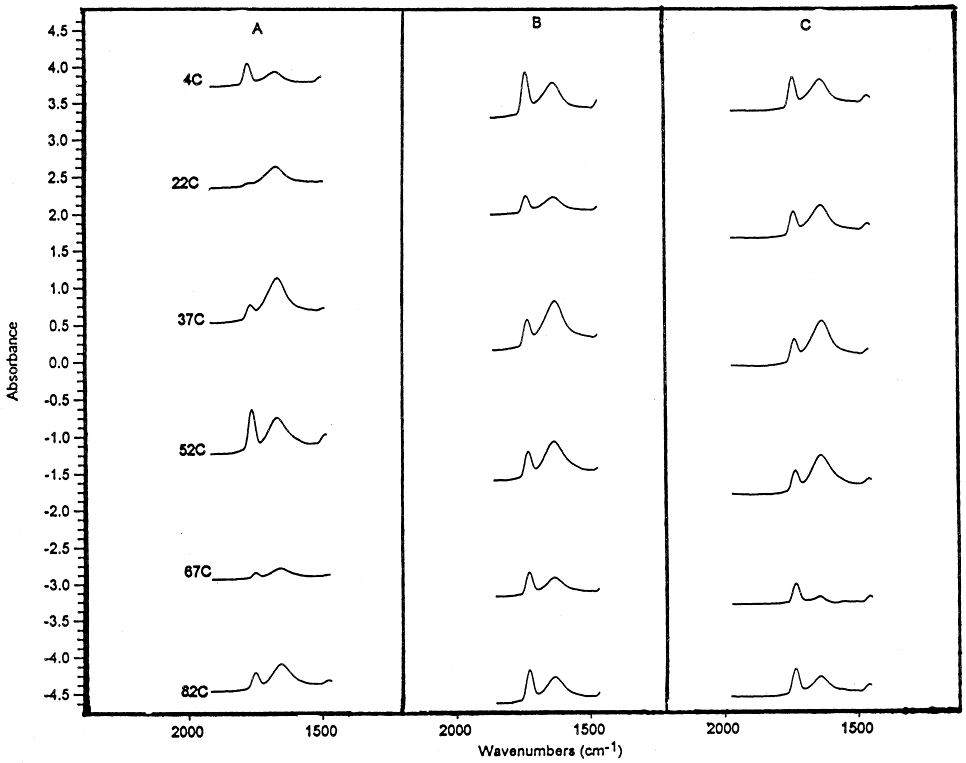

FIG. 4. FTIR spectra of –C=O stretching and H–O–H bending vibrations for control (A), 4% Lecigran (B), and 4% Lecimulthin (C) emulsions with increasing temperature. One hundred scans were co-added at a nominal resolution of 4 cm^{-1} (y is not to scale). See Figure 1 for abbreviation.

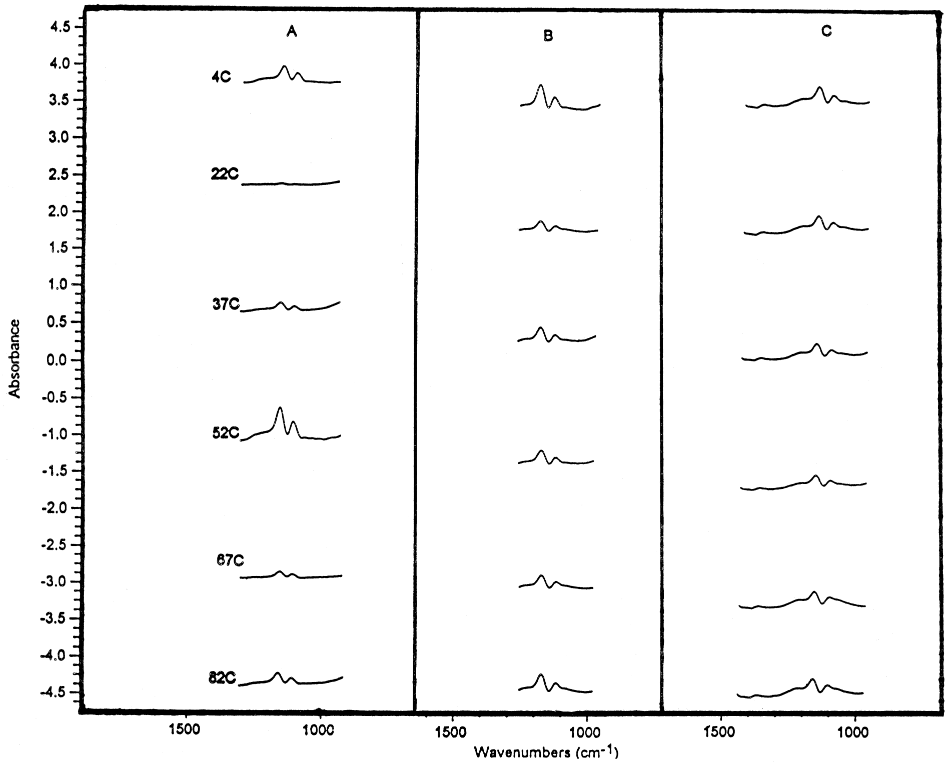

FIG. 5. FTIR spectra of P–O–C, P=O, and C–O–C stretching vibrations for control (A), 4% Lecigran (B), and 4% Lecimulthin (C) emulsions with increasing temperature. One hundred scans were co-added at a nominal resolution of 4 cm^{-1} (y is not to scale). See Figure 1 for abbreviation.

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