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Short Communication

Surface investigation of chitosan film with fatty acid monolayers

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Abstract: The surface pressure- molecular area (π-A) isotherm curves of two fatty acids of different chain lengths, i.e. stearic (C_{18}) and arachidic (C_{20}) acids, were obtained by using Langmuir-Blodgett (LB) technique. Results showed clear isotherm plots with limiting mean molecular area around 21 $Å^2$ for both acids. However, the monolayer was found to collapse at higher than 33 mN m^{-1} and 21 mN m^{-1} for stearic acid and arachidic acid respectively. The effect of Langmuir-Blodgett monolayers of the acids was investigated by atomic force microscopy (AFM). Chitosan film, before and after dipping in water, was also studied by means of AFM so that it could be used for comparison. It was found that the surface of chitosan was more homogeneous and smoother after dipping in water. In addition, more homogeneous surfaces were achieved after transferring a layer of the fatty acid onto the substrate.

Keywords: chitosan, fatty acids, Langmuir-Blodgett, atomic force microscopy

Introduction

Chitosan is an organic polysaccharide usually obtained by deacetylation of the most abundant naturally occurring chitin of crab and shrimp shells [1,2]. This fibril biopolymer is composed predominantly of unbranched chains of β -(1 $\rightarrow$ 4)-2-amino-2-deoxy-D-glucopyranose as shown in Figure 1. It is a non-toxic, biocompatible, and biodegradable polymer [3]. It has been widely used in diverse fields, ranging from waste management to food processing, medicine, and biotechnology [4-9].

Chitosan easily forms a film. In general, its films are clear and flexible, with good mechanical properties. In addition, they do not possess any pores [10]. Since chitosan degrades before melting, it is necessary to dissolve it in an appropriate solvent before casting into films. It is preferable to dissolve chitosan in acetic acid due to its non-toxicity and ease of removal.

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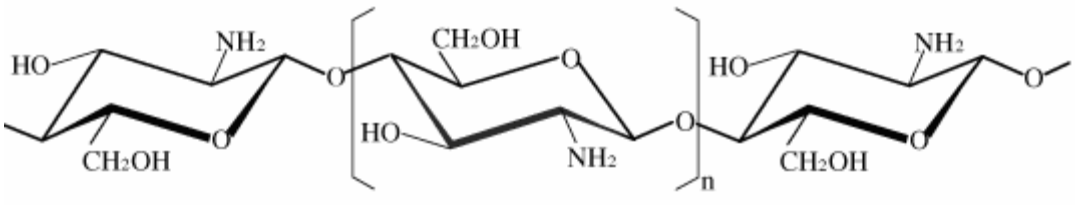

Figure 1. Chemical structure of chitosan

Generally, the film properties of chitosan depend on its morphology, which is effected by molecular weight, degree of N-acetylation, solvent evaporation, and free amine regenerating mechanism. In addition, the solvent used has an influence on the properties of chitosan films [11]. Chitosan coatings have been studied by several researchers to improve the quality and extend the storage life of food products [12-16].

The objective of this work is to characterise some of the surface properties of chitosan film after transferring a monolayer of stearic acid or arachidic acid onto its surface with a view to further explore the properties of the resulting thin film and its applications such as the production of contact lenses. These two fatty acids are known to form good-quality monolayers [17-19]. For this purpose we used Langmuir-Blodgett (LB) technique and atomic force microscopy (AFM).

Materials and methods

Materials

Shrimp-source chitosan was a gift from the chitin-chitosan laboratory of Universiti Kebangsaan Malaysia (UKM). Its degree of deacetylation (DD) was determined to be higher than 93 % by UV method according to the procedure reported by Muzzarelli and Rochetti [20]. Acetic acid (99.5%), stearic acid (99%) and arachidic acid (99%) were purchased from Sigma Chemicals. Chloroform (99.9%) was used as a solvent and deionised water (resistivity =18 MΩ cm) was used as a subphase.

Preparation of chitosan thin film

A certain amount of chitosan was dissolved in 1% acetic acid solution and cast into films at 75^{o}C on microscopic slides (size 25.4 ×76.2 mm, 1-1.2 mm thick), and left for 48 hours. This film was used as substrate for the LB monolayer study.

Methods

The molecular weight of chitosan was found to be about 7.9 $\times 10^5$ g mol^{-1} as determined by gel permeation chromatography (GPC) equipped with a Waters 1515 HPLC pump and Waters 2414 refractive index detector. The column used was PL aquagel-OH 30 (8 μm, 300 × 7.5 mm) and the solvent used was 1% acetic acid.

Surface pressure-area isotherms were recorded in a clean room using a KSV 3000 balance with symmetrical compression of the monolayers. A Whilhelmy plate was used as the surface pressure sensor. Stearic acid and arachidic acid were each dissolved in chloroform (1.0 mg mL^{-1}), and then an appropriate amount of each spreading solution (100 μL) was carefully injected onto the air-water

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interface. After the chloroform was allowed to evaporate for about 10 min, the monolayer film was compressed at a speed of 10 cm^2 min^{-1}. After that, the monolayer was transferred onto the chitosan substrate with a dipping speed of 5 cm^2 min^{-1} at 9.5 mN m^{-1}. Only one layer at the condensed phase was required in this work. All samples as well as the recording of isotherms were prepared or made in the clean room at room temperature.

Atomic force microscopic observation was performed on a digital AFM nanoscope, Dimension 3000 (Digital instruments, Santa Barbara, CA). AFM studies were carried out using the tapping mode under air atmosphere at ambient temperature. The scan size, set point and scan rate are shown in the images. The tapping set-point was adjusted to minimise probe-sample interactions.

Results and Discussion

Langmuir-Blodgett film

Figure 2 depicts the surface pressure-molecular area (π-A) isotherm curve of stearic acid monolayer. The isotherm shows considerable curvature indicating expanded monolayer behaviour. The surface pressure increases gradually, reaches a plateau, and then increases again at lower molecular area before collapsing. In the region of high surface pressure (above 20 mN m^{-1}), the isotherm seems to have a constant slope. The monolayer was found to collapse at higher than 33 mN m^{-1}. The limiting molecular area observed, i.e. the area occupied per molecule when the monolayer is closely packed, was 21 $Å^2$. This value was estimated from the π–A curve by extrapolating the condensed region to zero pressure. The liquid condensed region was around 7-10 mNm^{-1} and 19-22 $Å^2$. Longer liquid condensed region may suggest that a more stable condensed film is formed.

The arachidic acid isotherm is shown in Figure 3. In the region of high surface pressure, the isotherm has a nearly constant slope and this part of the isotherm has been extrapolated to zero surface pressure to evaluate the mean molecular area. The limiting mean molecular area is about 21 $Å^2$ while the pressure collapse is higher than 21 mNm^{-1}. The liquid condensed region is about 6-10 mNm^{-1} and 22-25 Å.

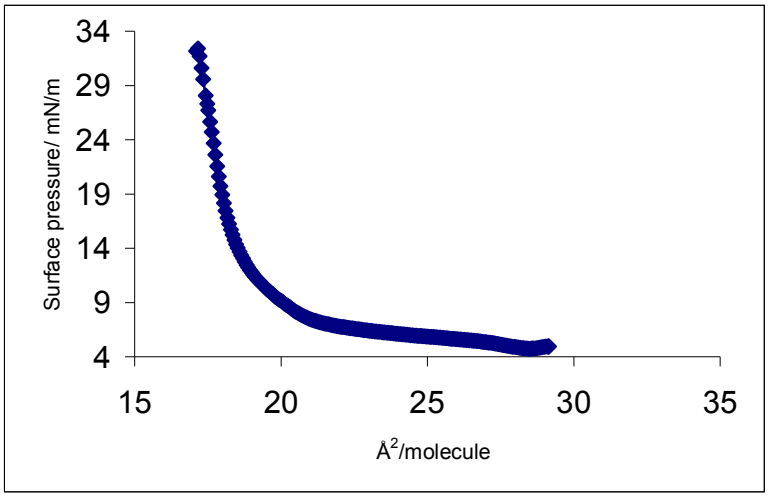

Figure 2. Surface pressure-area isotherm for monolayers of stearic acid

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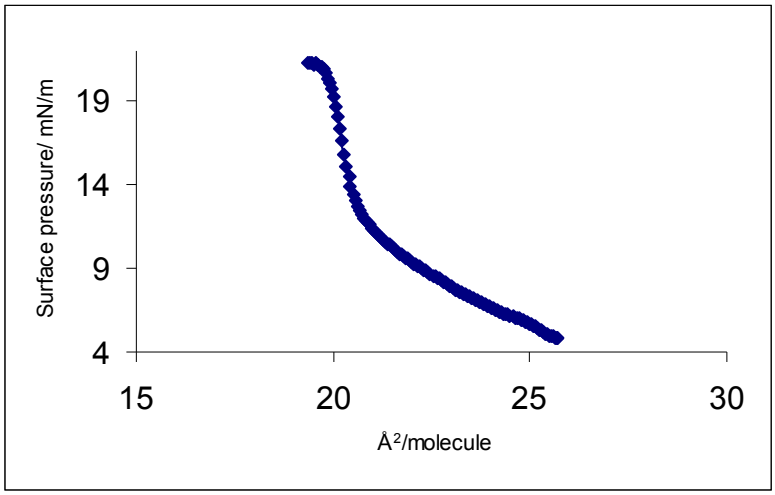

Figure 3. Surface pressure-area isotherm for monolayers of arachidic acid

Atomic force microscopy (AFM)

AFM was used to investigate the smoothness of the surfaces as well as detect possible orientational structures in the LB films. Tapping-mode AFM images can yield information about the surface features of the films.

Figure 4 shows a tapping-mode AFM photograph of chitosan film. The image shows some holes and mountains and it seems to be inhomogeneous. However, the film becomes homogeneous and smoother and the bumps disappear after being dipped in deionised water (Figure 5), which is probably due to a layer of water that forms on the surface. In addition, the surface seems to be robust and not damaged at the tip. However, after transferring a layer of stearic acid or arachidic acid onto the chitosan film at the surface pressure of 9.5 mNm^{-1}, the images show smoother surfaces despite the presence of some bumps as shown in Figure 6. These observations may indicate the existence of interaction between chitosan and the two fatty acids, which is most likely due to the formation of intermolecular hydrogen bonding between the amino and hydroxyl groups in the chitosan and the hydroxyl groups in the fatty acids.

When the monolayers are transferred onto a solid substrate, molecules are fixed and their distribution is not changed. Figure 7 shows that the distribution of the fatty acids is quite uniform and the substrate (chitosan) surface seems to be covered by the film.

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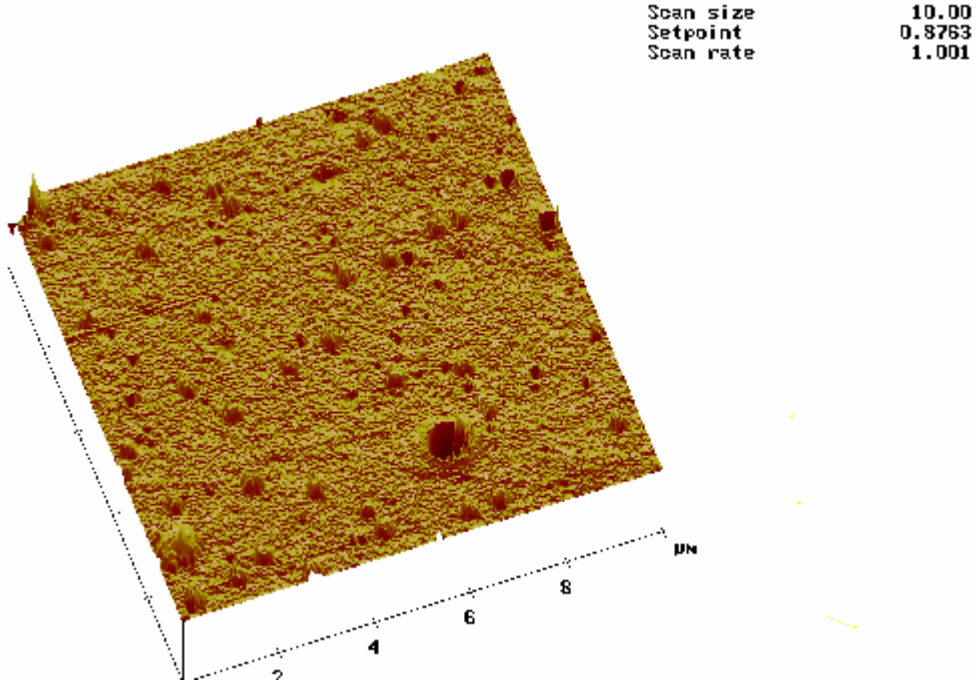

Figure 4. AFM image (10 x 10 μm) of chitosan film

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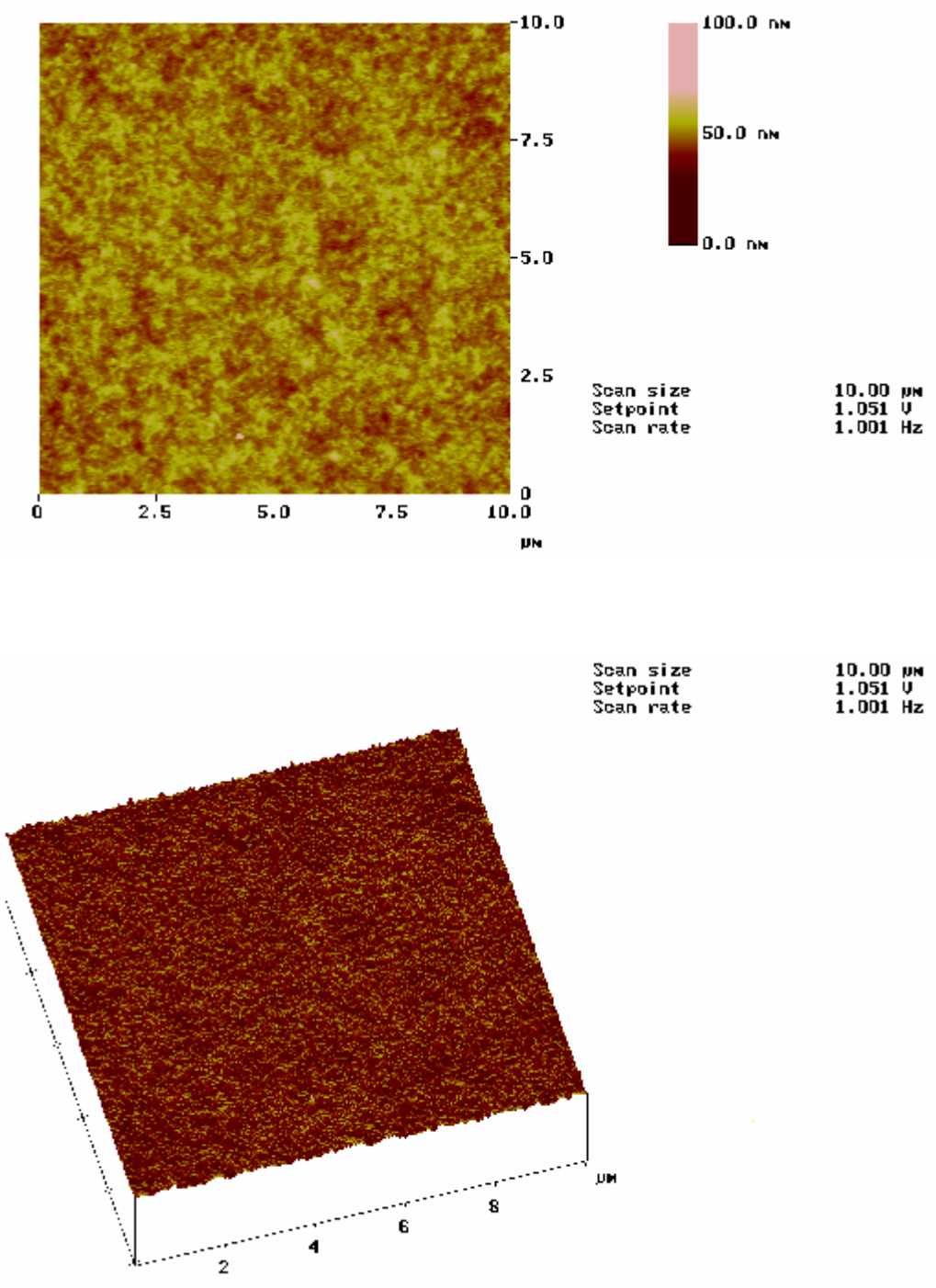

Figure 5. Topographic AFM images (10 x 10 µm) of chitosan film after being dipped in deionised water

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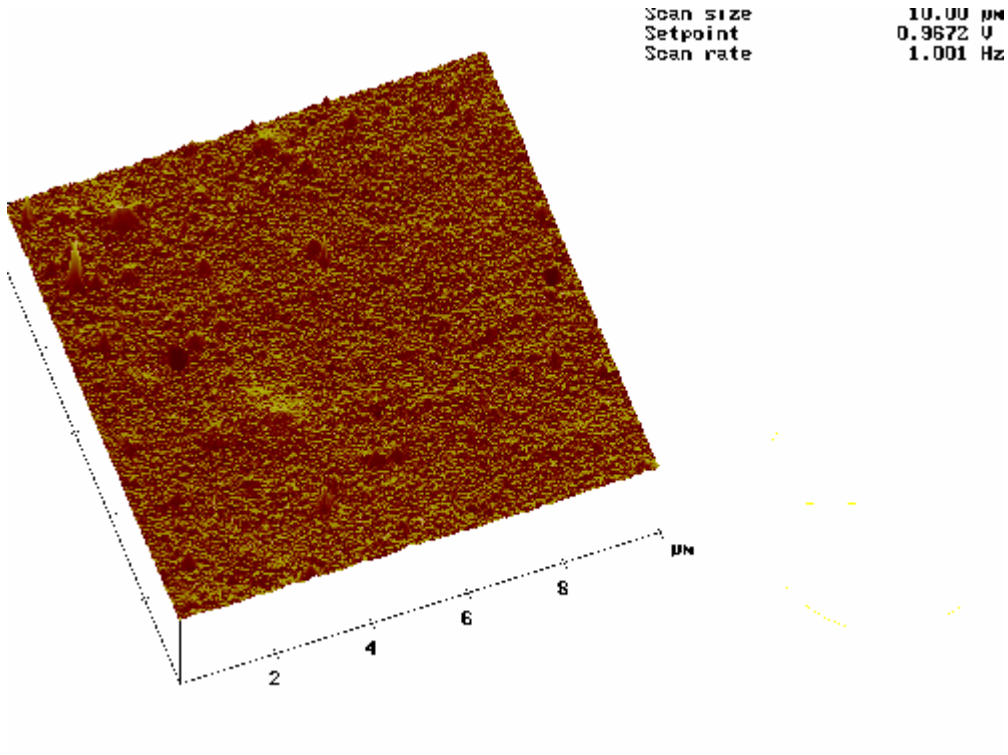
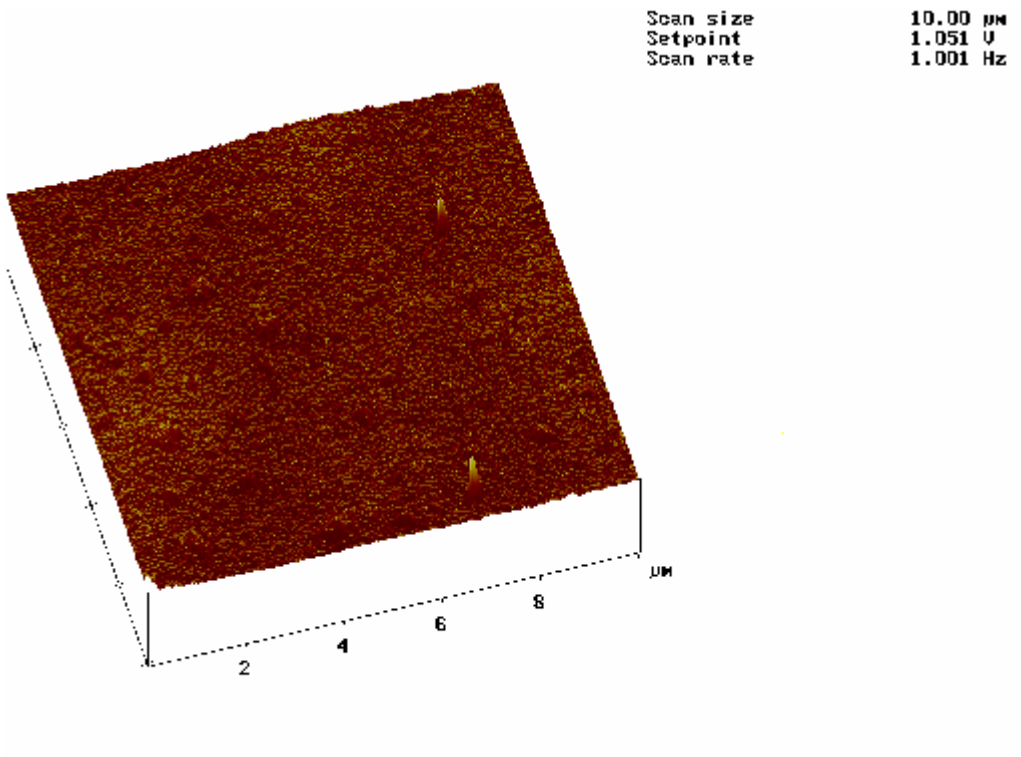


(A)



(B)

Figure 6. AFM photographs (10 x 10 µm) of a transferred monolayer of stearic acid at 9.5 mN m^{-1}(A) and arachidic acid at 9.5 mN m^{-1}(B) onto chitosan film

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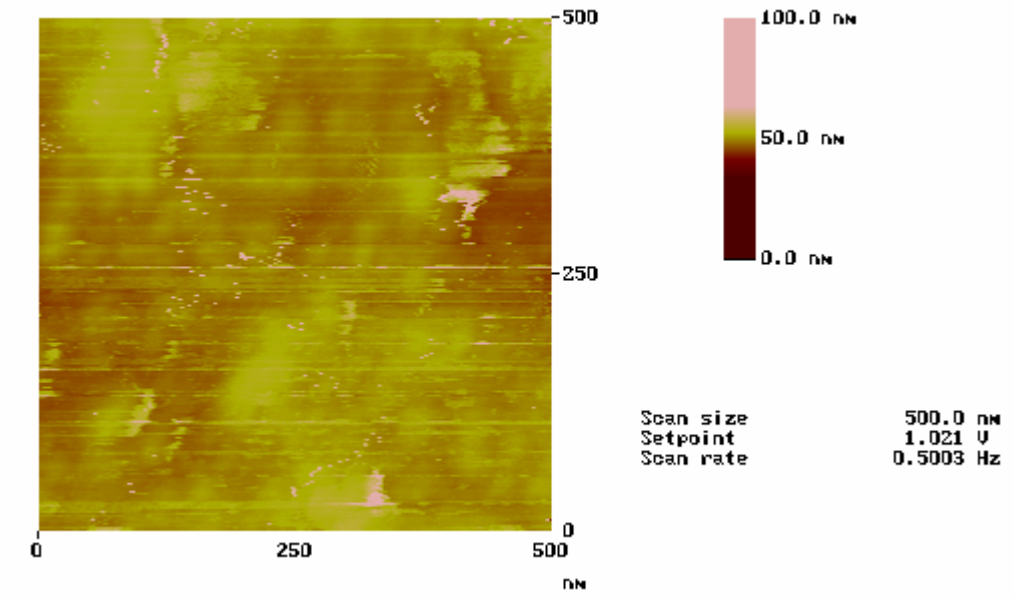
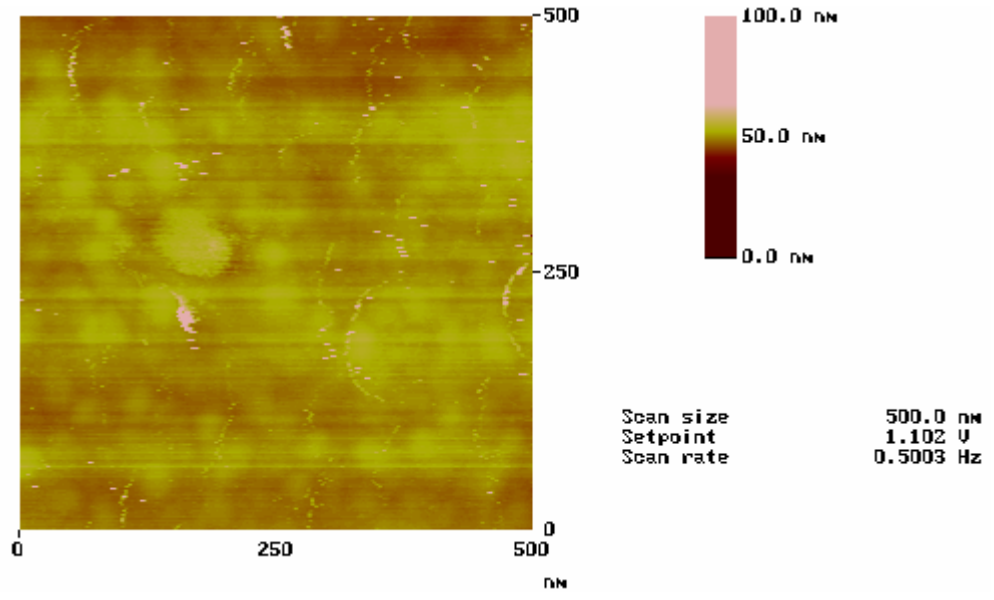


(A)



(B)

Figure 7. AFM photograph (0.5 x 0.5 μm) of LB film of a transferred monolayer of stearic acid at 9.5 mNm^{-1}(A) and arachidic acid at 9.5 mNm^{-1}(B) onto chitosan film

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Conclusions

This study has shown that the LB measurements of stearic acid and arachidic acid give distinct isotherm plots. AFM photographs show holes and mountains on the surface of chitosan film. However, improvement in the surface morphology of chitosan film can be achieved after transferring a monolayer of fatty acids onto the film, i.e. smoother surfaces are obtained after coating chitosan film with a fatty acid monolayer, which makes this film suitable for certain applications.

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