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INTRODUCTION

Fruit juice consumption is beneficial for the maintenance of good health and prevention of diseases. The positive health benefits of juices have been ascribed in part to vitamin C (ascorbic acid), the major vitamin found in fruits and vegetable [1,2]. Citrus fruits are also known to contain bioactive compounds including antioxidative, anti-inflammatory, antitumor, and antimicrobial activities [3,4]. *Citrus spp.* is believed to possess bioactivities such as antioxidant [5,6]. They are important source of many bioactive compounds. *Citrus aurantifolia* (L.) Swingle. Family: Rutaceae, commonly known as lime. It is a medicinal plant and very popular in Nakhon Si Thammarat province, Thailand. Lime juice is also commonly used in the food, drug, and cosmetic industries because of their medicinal properties [7]. It may be an alternative medicine for antibacterial materials and use as natural preservatives. However, the antibiotic resistance is highly desirable to develop additional and more effective antibacterial agents. Nanoparticles appear to exhibit high potential to solve the problem of bacterial multidrug resistance. So Lime juice in nanoparticle form may exhibit a better of antibacterial activity against Gram-negative and Gram-positive bacteria than lime juice. Therefore, we want to prepare Lime juice - graphene quantum dots (LJ-GQDs) which is in nanoparticle form and apply to against various bacteria.

EXPERIMENTAL SECTION



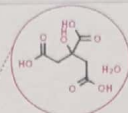
Lime Tree



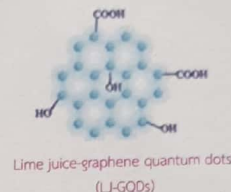
Lime fruit



Lime Juice



Pyrolysis (200°C)
NaOH (0.25 mol/L)



Lime juice-graphene quantum dots (LJ-GQDs)

Antibacterial Activity



Gram-positive bacteria (*Staphylococcus aureus*)

Gram-negative bacteria (*Escherichia coli*)

1. Preparation of plant extracts

Lime (*Citrus aurantifolia*) was crushed and filtered. First part was stored for the study of antibacterial activity and the another part was continuously heated to 200°C in a paraffin oil bath. Then add 0.25 M NaOH to stop the reaction and heat continuously until the solution is dark brown and viscous. The final product was collected at 4°C until use.

2. Antibacterial activities of plant extracts (Lime juice and Lime juice-GQDs)

The antibacterial activities of plant extracts was evaluated against *S. aureus* and *E. coli* by agar well diffusion method.

3. Determination of minimum inhibitory concentration (MIC) using micro-broth dilution test

The different concentrations of plant extracts were investigated by agar well diffusion method and micro-broth dilution method. The lowest concentration of the extracts that did not exhibit any visible growth was determined as MIC value of extracts against tested bacteria.

4. Minimum bactericidal concentration (MBC) and MIC index determination

The turbidity tubes in MIC test were cultured again on MHA and incubated for 24 h at 37°C. The concentration of the extracts with no growth was regarded as MBC.

CHARACTERIZATION

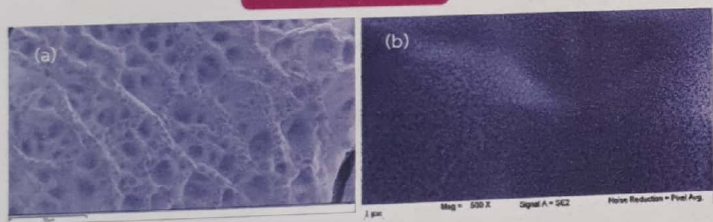


Fig. 1. SEM images of (a) Lime juice and (b) Lime juice-graphene quantum dots

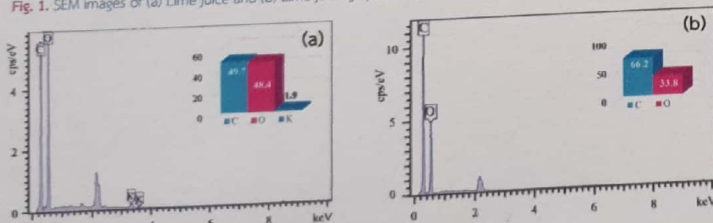


Fig. 2. EDX spectrum of (a) Lime juice and (b) Lime juice-graphene quantum dots

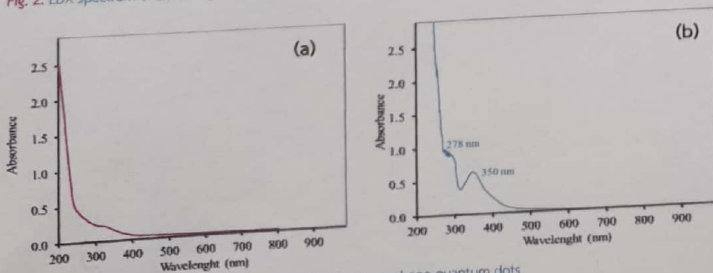


Fig. 3. UV-Vis spectrum of (a) Lime juice and (b) Lime juice-graphene quantum dots

The morphology of all materials were characterized by SEM technique. Fig. 1 shows the SEM images of Lime juice (Fig. 1(a)) and Lime juice-graphene quantum dots (LJ-GQDs) (Fig. 1(b)). It can be seen that the morphology of Lime juice is in sheet form while LJ-GQDs is in spherical shape which particle sizes is in nanometer. To determine the major elements of Lime juice and LJ-GQDs. The plant extracts were analyzed by EDX technique. From Fig. 2(a), the EDX spectrum of Lime juice shows various elements including C, O and K while LJ-GQDs shows only strong peaks of C and O in the prepared sample (Fig. 2(b)), indicating that LJ-GQDs was successfully synthesized. And also UV-Vis spectrum (Fig. 3(b)) confirmed the formation of LJ-GQDs with absorption peaks at 278 nm and 350 nm, corresponding to $\pi-\pi^*$ and $n-\pi^*$.

RESULTS AND DISCUSSION

ANTIBACTERIAL ACTIVITY

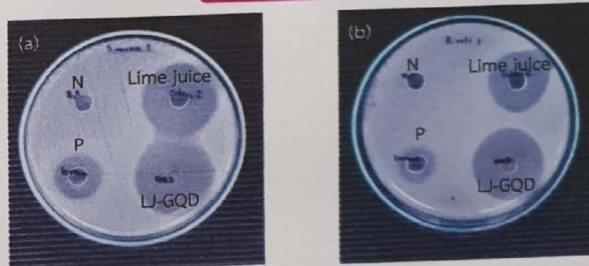


Fig. 4. Inhibition zone of Lime juice and LJ-GQD (a) *S. aureus* (b) *E. coli*
N = distilled water, P = citric acid

Fig. 4 shows the zone of inhibition produced by samples LJ-GQDs against *E. coli* and *S. aureus* bacterial strains. Their inhibition zones were compared with Lime juice, distill water (N) and citric acid (P). From the results, it was found that the synthesized LJ-GQDs demonstrated excellent antibacterial activity for *E. coli* bacteria with their MIC values were 12.5 mg/L and 25 mg/L for *E. coli* bacteria and *S. aureus* bacteria, respectively. While the Lime juice cannot kill the both types of bacteria but inhibits the growth of bacteria at 50 mg/mL which showed in Table 1.

Table 1. Minimum inhibitory and bacteriostatic concentrations of Lime juice and Lime-GQDs against *S. aureus* and *E. coli* expressed in mg/mL

Type of material	<i>S. aureus</i>		<i>E. coli</i>	
	MIC	MBC	MIC	MBC
Lime Juice	50	-	50	-
Lime juice-GQDs	12.5	12.5	12.5	25

CONCLUSION

In our work, we have reported the antimicrobial activity of graphene quantum dots (GQDs) derived from Lime juice as resource. Different characterization techniques have been shown that LJ-GQDs were synthesized successfully. The prepared LJ-GQDs exhibits antibacterial activity against both Gram-positive (*S. aureus*) and Gram-negative (*E. coli*), with the MIC at 12.5 mg/L. Indicating that the synthesized LJ-GQDs demonstrated excellent antibacterial activity for both *E. coli* bacteria and *S. aureus* bacteria.

REFERENCES

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