

Research Article

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Growth, characterization, and anti-bacterial activity of L-methionine supplemented with sulphamic acid single crystals

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Abstract: Sulphamic acid (SA) crystals supplemented with L-methionine (LM) were grown at moderate temperatures using a slow evaporation procedure. The powder XRD pattern showed that LM supplemented with SA (LMSA) crystals have an orthorhombic crystal structure. The FTIR studies confirmed the presence of various vibrational modes. Using a UV-Vis spectrometer, the transmittance of LMSA in the UV and visible range was observed, and the band gap of the LMSA was also calculated. The hardness value of LMSA was higher compared to that of pure SA. Photoluminescence emission studies of LMSA pointed out emissions at 491 and 542 nm, which were attributed to the transition from the 5D_4

state to 7F_6 and 7F_5 ground, respectively. LMSA crystals were effective in killing pathogenic bacteria, according to the studies on their anti-bacterial activity.

Keywords: powder XRD, FTIR studies, orthorhombic crystal structure, L-methionine, SA crystals

1 Introduction

The creation of passive and active photonic devices primarily makes use of non-linear optical materials [1]. Optics and electronics are two fields where the utilization of single crystals is evident [2]. Sulphamic acid (H_2NSO_3H , SA) is one of the most generally used inorganic compounds in industry [3]. It is now used in place of traditional Lewis and Bronsted acid catalysts. SA is a white-coloured, odourless, crystalline solid, and is non-corrosive, non-volatile, and non-hygroscopic. It is also a very effective green catalyst in organic synthesis [4] and a potent inorganic acid that assumes a zwitterionic form when combined with water. Because of these benefits, the British Analytical Methods Committee and IUPAC, as well as the Japanese Industrial Standard, suggested SA as a benchmark ingredient in titrimetric testing [5]. In multistage flash evaporation desalination plants, SA identifies good applications for heat exchangers, demisters, cleaning, cooling water systems, etc. [2]. The orthorhombic structure and zwitterionic form of sulphuric acid are present. Various reports have suggested the formation of SA in the shape of single crystals at moderate temperatures [6–10]. Numerous researchers have shown interest in SA crystals, and different combinations of these crystals have been synthesized, and their properties have been reported [11–28]. SA has been utilized in many applications, especially in polymerization, and utilized as a catalyst in many organic reactions. It is employed as a herbicide, cross-linking agent in polymer synthesis, a fire retardant or fireproofing agent, a fabric dyer, an air

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cleaner, a pulp bleaching agent, a scale remover and metal cleaner, a pigment printer, and to make less toxic cigarettes [29]. L-Methionine (LM) is an α -amino acid that possesses a protonated amino group (NH_3^+), S-methyl thioether side chain, and deprotonated carboxylic acid group (COO^-). As a precursor, LM takes part in the biosynthesis of proteins. It is categorized as an aliphatic and non-polar amino acid. The primary characteristic of LM and some of its metal compounds is their catalytic activity, which confers precise antiviral effects. This amino acid is converted into creatine monohydrate, which is a material required for the production of energy and muscle growth [30]. Here, we report the growth of LM supplemented with SA (LMSA) (single crystals), its structural and optical properties, and its anti-bacterial activity.

2 Experimental methods

SA crystals containing LM were produced utilizing the slow evaporation method. LM powder and high-purity SA were employed as the initial components. Both LM and SAs were purchased from Sigma-Aldrich (reagent grade $\geq 98\%$). The saturated aqueous solution of 1,000 ml was prepared using recrystallization SA ($\text{NH}_2\text{SO}_3\text{H}$) and LM SA ($\text{C}_5\text{H}_{11}\text{NO}_2\text{S}$) in a ratio of 3:1 with distilled water at 35°C . At room temperature, the dissolved solution was agitated with a magnetic stirrer for 2–3 h, and a clear solution was obtained. The prepared translucent and saturated solution was then filtered *via* filter paper. Then, the resultant solution was closed using a perforated polythene sheet to crystallize at moderate temperatures with a decontaminate surrounding. Good, colourless, and transparent crystals were obtained after 20 days and were used for characterization.

The stoichiometric equations for the formation of LMSA are as follows:

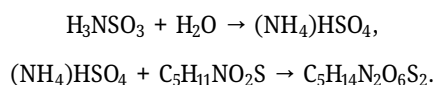


Figure 1 depicts an image of the grown LMSA crystal.

The anti-bacterial effect of LMSA crystals was determined with Gram-positive and Gram-negative bacteria using the disk diffusion Kirby–Bauer method. This test was conducted with a Mueller–Hinton agar medium. During this study, the antimicrobial-impregnated discs were positioned on the surface and incubated for 24 h at 35°C . Following incubation, a zone of inhibition corresponding to the degree of antimicrobial activity on the bacteria was observed on the plates. Any zone of inhibition that is observed suggests a level of susceptibility to the antibiotic; the bigger the zone, the

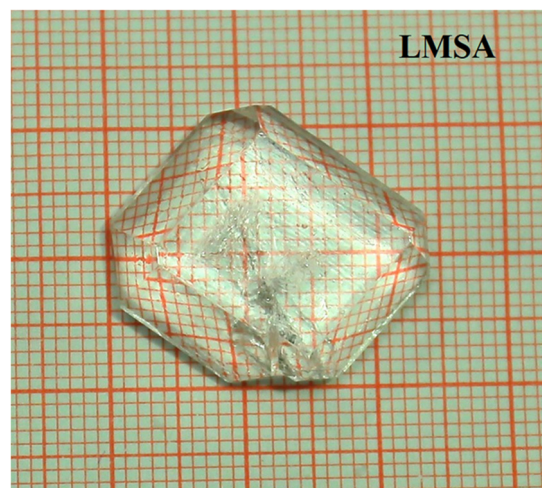


Figure 1: Image of LMSA crystal.

stronger the antibiotic. Conversely, the absence of any zone signifies complete resistance to germs. Afterwards, the diameter of the inhibition zone from the disc centre was measured, and the result was compared with the standard set of values to find the susceptibility of the bacteria in LMSA.

3 Results and discussion

3.1 X-ray diffraction

The XRD patterns of the SA crystal and SALM are shown in Figure 2. The XRD profile's diffraction peaks are compared with the accepted JCPDS data (JCPDS Card No.: 70-0060) of the pure SA crystal. For the pure SA crystal, a high-intensity peak was observed along the (2 2 1) direction. The XRD pattern of LMSA samples shows a high-intensity peak along the (102) direction. In addition, the strength of the strongest peak (211) and other peaks also reduced. The variation in the relative intensities of the different peaks was also discovered, which indicated that LM had been added to the pure SA. The single XRD and powder XRD patterns reveal that the synthesized LMSA possess orthorhombic crystal structures with reduced cell volume (Table 1).

3.2 FT-IR analysis

Figure 3 displays the FT-IR spectrum of the grown crystals (LMSA). The spectrum reveals the NH_3^+ mode of bonding existing at frequencies $3,153$ and $2,872\text{ cm}^{-1}$. Triband overtones and a combination of hydrogen-bonded OH bending modes were responsible for the bands observed at $2,449$ and $2,026\text{ cm}^{-1}$. The bands observed at $1,807$ and $1,451\text{ cm}^{-1}$ were

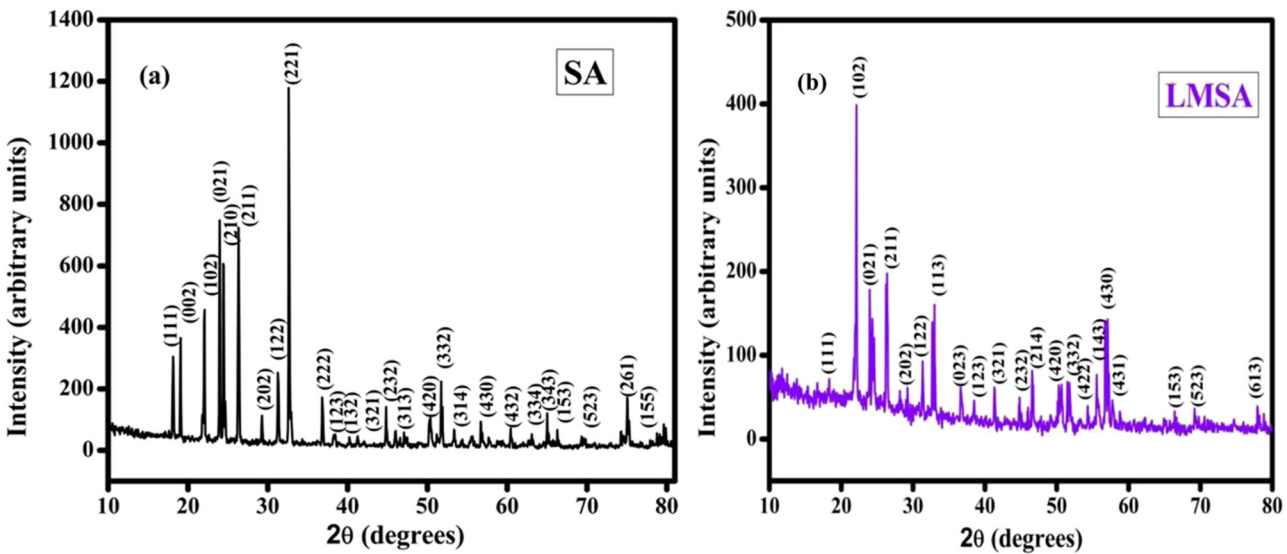


Figure 2: X-ray diffraction of SA and LMSA crystal.

attributed to the NH_3^+ mode of vibration deforming, while the mode at $1,066\text{ cm}^{-1}$ was attributed to symmetric SO_3^- stretching, and the vibration bands found at $1,285\text{ cm}^{-1}$ were due to degenerated SO_3^- stretching. The generation of zwitterions in SA and LMSA crystals was verified by the presence of the rocking mode vibration of NH_3^+ at $1,002\text{ cm}^{-1}$ [26].

The N–S stretching vibration at 695 cm^{-1} was observed, while the band at 536 cm^{-1} was due to the degraded SO_3^- deformation [6]. All of the IR bands that appeared in the grown crystals were analogues of bands that had been theoretically computed and were in excellent accordance with previous studies [26,31]. The observed wave numbers and the corresponding assignments of SA and LMSA are listed in Table 2. However, nearly all of the modes were apparent in LM-supplemented crystals. However, there was a small variation in the intensities, indicating that the sublattice geometry may have slightly changed.

3.3 UV-Vis studies

Considering that the formed crystals are used in a variety of optoelectronic device applications, it is crucial to understand

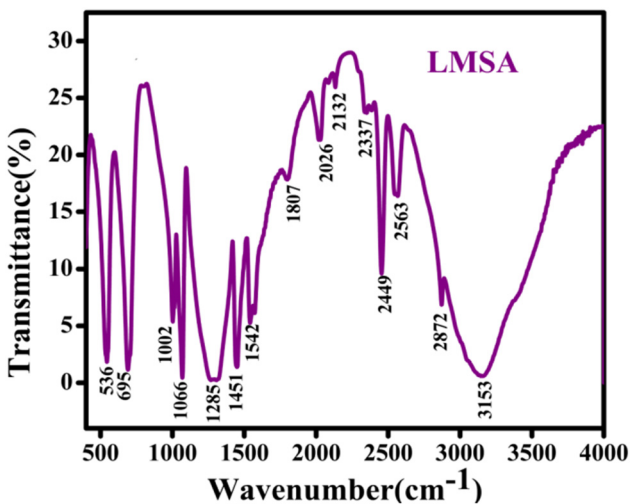


Figure 3: FTIR spectrum of LMSA.

the optical transparency of these materials. Figure 4 shows the transmittance spectrum of LMSA and indicates that the prepared crystals are 98% transparent. Because of their increased crystalline nature, LMSA crystals are more transparent. According to Senthil Pandian *et al.* [32], the scattering from point and line defects is likely the cause of the decrease

Table 1: Lattice parameters and structures of SA and LMSA

Sample	a (Å)	b (Å)	c (Å)	$\alpha = \beta = \gamma$	Volume (Å ³)	Structure
SA	8.1266	8.0928	9.2298	90°	607.0166	Orthorhombic
LMSA	8.1311	8.0695	9.2362	90°	606.0270	Orthorhombic

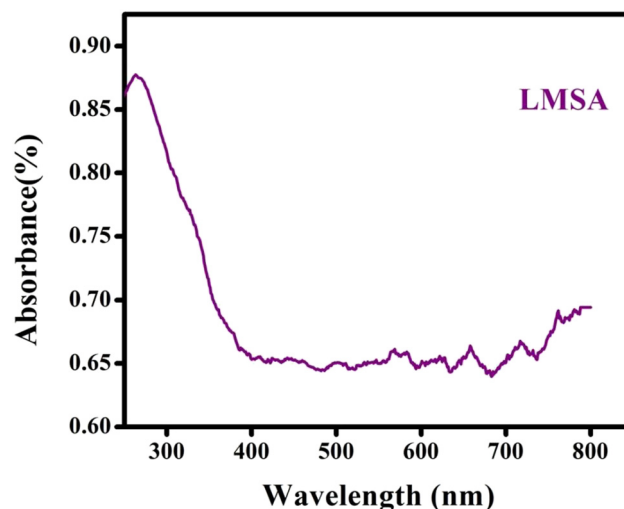
Table 2: Vibrational assignment of LMSA

Wave number (cm ⁻¹)	Assignment
LMSA	
3,153	Degenerated NH ₃ ⁺ stretching
2,872	Symmetric NH ₃ ⁺ stretching
2,449	S-H stretching
2,026	N-H stretching
1,807	Symmetric NH ₃ ⁺ deformation
1,542	Degenerated NH ₃ ⁺ deformation
1,451	Symmetric NH ₃ ⁺ deformation
1,285	Degenerated SO ₃ stretching
1,066	Symmetric SO ₃ stretching
1,002	Rocking mode NH ₃ ⁺
695	N-S stretching
536	Degenerated SO ₃ deformation

in transmittance in SA crystals. As shown in Figure 5, LMSA shows a lower UV cutoff wavelength of 263 nm, which indicates that LMSA has a higher percentage of absorption than that of pure SA [33]. To determine the band gap energy value, a Tauc plot analysis was conducted, as shown in Figure 6, and the optical energy band gap of LMSA was calculated to be 3.5 eV.

3.4 Vickers hardness test

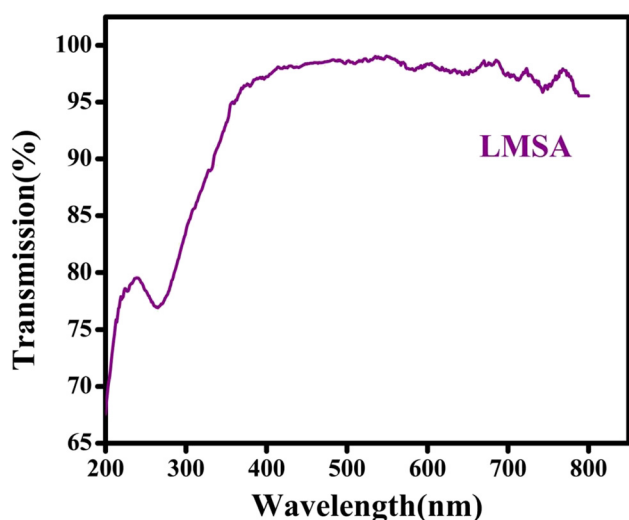
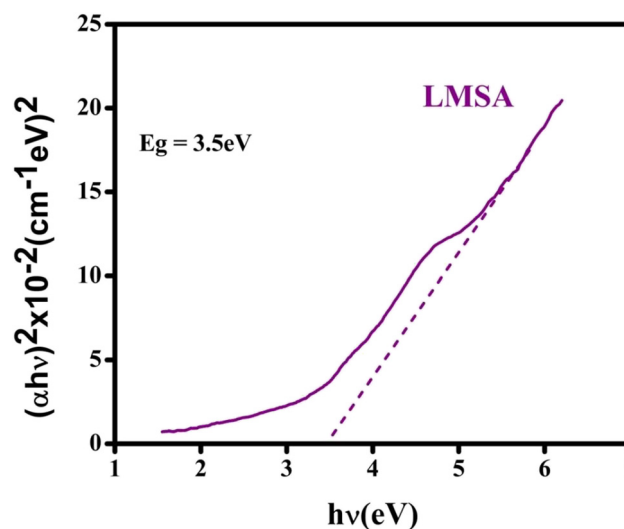
The effectiveness of crystals depends mostly on their optical properties and sound mechanical behaviour [25,26]. At room temperature, the Vickers microhardness measurements were conducted on LMSA crystals with loads ranging

**Figure 5:** UV absorption of LMSA crystal.

from 25 to 100 g. For different utilized loads p (in g), the indentation diagonal lengths d (in m) were measured. The Vickers hardness number was derived by the following relationship:

$$H_v = 1.8544P/d^2 \text{ kg mm}^{-2}.$$

The curve between the load and hardness number is shown in Figure 7. Up to 100 g, with an increase in the load, the hardness value increased; however, no break was spotted in the plot. The LMSA crystals showed an enhanced hardness in the presence of LM. Arumugam *et al.* [33] and Ramesh Babu *et al.* [12] also noted the same behaviour. Figure 8 depicts the outcomes of measuring the work hardening coefficient from the plot of $\log p$ vs $\log d$. The straight

**Figure 4:** UV transmission spectrum of LMSA crystal.**Figure 6:** Tauc's plot of LMSA.

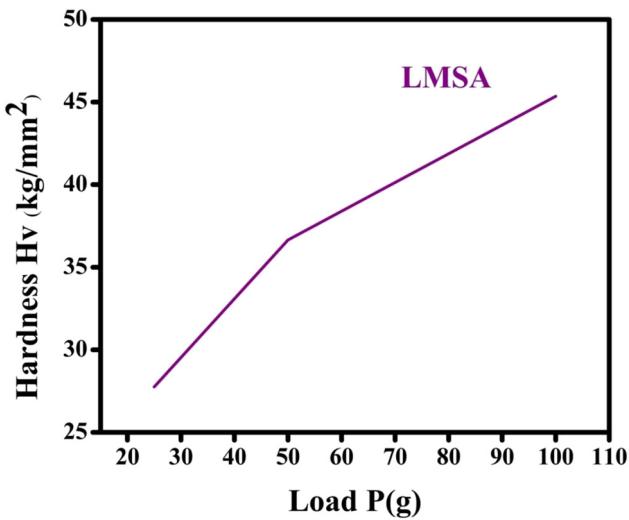


Figure 7: Load P and hardness number of LMSA.

line produced by the least-squares fitting was in fine agreement with Meyer’s law. The value of n was obtained from the slope. In line with Senthilkumar *et al.* [33], n needs to be above 1.6 for soft materials and between 1 and 1.6 for hard materials. Since n is above 1.6 for the LMSA crystal, it is considered a soft material.

3.5 Photoluminescence studies

Figure 9 displays the LMSA crystal’s emission spectrum, which revealed emissions in the visible and ultraviolet zones. In the visible area, emissions of green, blue, and violet are observed. A strong blue emission, observed at 491 nm, is the strongest among all the detected emissions.

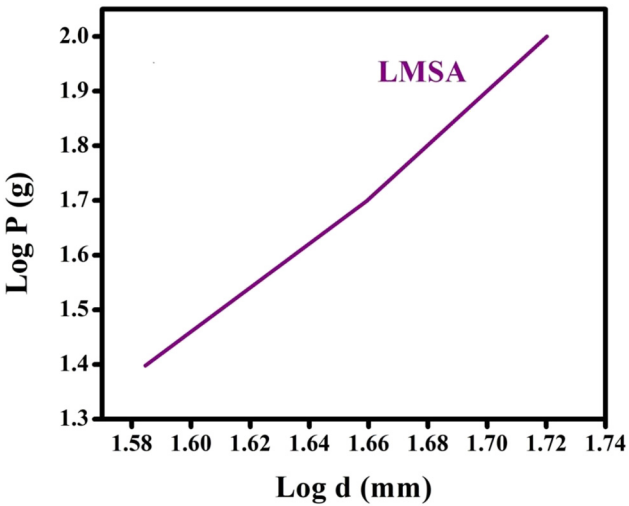


Figure 8: Log P and Log D of LMSA.

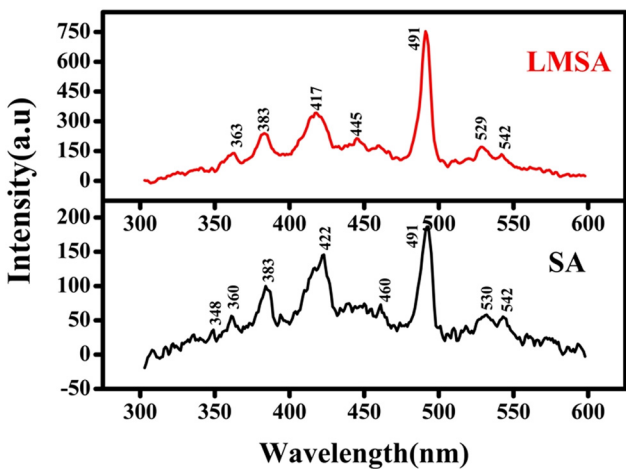


Figure 9: Photo luminescence spectrum of SA and LMSA.

According to Arumugam *et al.* [34], the peaks in the ultra-violet area that are noticed below 400 nm were caused by the electronic transition $\pi-\pi^+$. The peaks observed at wavelengths below 500 nm resembled blue emission [35]. These emissions were caused by the transition between the energy levels 5D_4 to 7F_6 . The transition from 5D_4 to 7F_5 energy levels caused the peak at 529 nm to be associated with green emission [24]. A close inspection reveals an improvement in the blue emission intensity. This crystal can be used in blue-emission optoelectronic devices in the future due to its strong blue-emission properties [36]. Sun *et al.* [41], Wang *et al.* [42], and Zhang *et al.* [43] highlight the physical importance of low molecular weight polysaccharides from *Dendrobium officinale*, the surface-functionalized design of blood-contacting biomaterials, and the behaviour of environmental pH on the interaction characteristics of WP-EGCG non-covalent nano-complexes. Cui *et al.* [44], Zhao *et al.* [45], and Xu *et al.* [46] scrutinized the characterization and synthesis of Eu (III) complexes of modified cellulose and found an efficient reduction of beta-lactoglobulin allergenicity and that royal jelly acid suppresses hepatocellular carcinoma tumourigenicity, respectively. Selective enrichment of partial denitrifying bacteria, partial

Table 3: Anti-bacterial effect of LMSA crystals

Name of bacteria	Inhibition zone of LMSA (mm)	Inhibition zone of the standard antibiotic Amikacin (mm)
<i>S. aureus</i>	35	22
<i>B. klebsiella</i>	32	20
<i>E. coli</i>	35	22
<i>Serratia</i>	32	19

Table 4: Inhibition zone of LMSA compared with other reported materials

Materials	Bacteria name	Zone of inhibition (mm)	Ref.
MSA	<i>E. coli</i>	35	Present work
	<i>S. aureus</i>	35	
Piper betle	<i>E. coli</i>	22.5	[37]
	<i>S. aureus</i>	15.3	
LTSB (L-threoninum sodium bromide)	<i>E. coli</i>	15	[38]
	<i>S. aureus</i>	12	
f6-(1,3-Benzodioxol-5-ylmethyl)-5-ethyl-2-([2-(morpholin-4-yl)ethyl]sulfanyl)pyrimidin-4(3H)-one	<i>S. aureus</i>	20	[39]
Ferulic acid crystals	<i>E. coli</i>	20	[40]

nitrification-anammox, and sulphur autotrophic denitrification in one reactor and cinnamic hydroxamic acid collector, as well as the examination of flotation attributes and the appliance of scheelite, has been recently discussed by Ahmad *et al.* [47], Zhao *et al.* [48], and Yao *et al.* [49], respectively.

3.6 Anti-bacterial activity

Using the disk diffusion Kirby–Bauer method, the anti-bacterial effect of LMSA was investigated against *Staphylococcus aureus* and *Bacillus*, which were Gram-positive, and *Klebsiella*, *Escherichia coli*, and *Serratia*, which were Gram-negative. The Mueller–Hinton agar medium was used in the disk diffusion test.

The recognized normal antibiotic half-life of *E. coli* is 22 mm; however, in LMSA, it was lengthened. The other bacteria, such as *S. aureus*, *Bacillus*, *Klebsiella*, and *Serratia*, also showed a lengthened inhibition zone compared to the inhibition zone of the standard antibiotic. Table 3 shows the anti-bacterial effect of the LMSA crystal. The anti-bacterial effect of LMSA is compared with other reported materials and is given in Table 4. From this, we deduced that the produced crystals were more potent than conventional and recently developed antibiotic materials.

4 Conclusion

At room temperature, slow evaporation was used to grow SA crystals and crystals containing LM. The structure of the grown crystals and lattice constants were validated by powder XRD. When LM was added to SA, a modest change in the crystal's lattice properties and volume was observed. When LM was added to SA, there was no discernible phase shift. The vibrational modes of the LMSA crystals were analysed using FTIR. The infrared bands observed in the

grown crystals were all theoretically computed bands that matched well with those in previous research. The optical studies verified that LMSA crystals become more transparent due to their increasing crystalline nature. The band gap value of the LMSA crystal was calculated to be 3.5 eV. The addition of LM enhanced the mechanical strength of the crystal. The greatest peak in the photoluminescence experiments that resulted from the 5D_4 to 7F_6 transition reveals blue emission. Thus, the grown crystals open the door to future use of these crystals in blue emission optoelectrical systems. The enriched anti-bacterial effect of the LMSA confirms that this crystal will be a promising material for eliminating harmful microorganisms.

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