Supplementary Material

Synthesis and antimicrobial activity of some new thienopyrimidine derivatives

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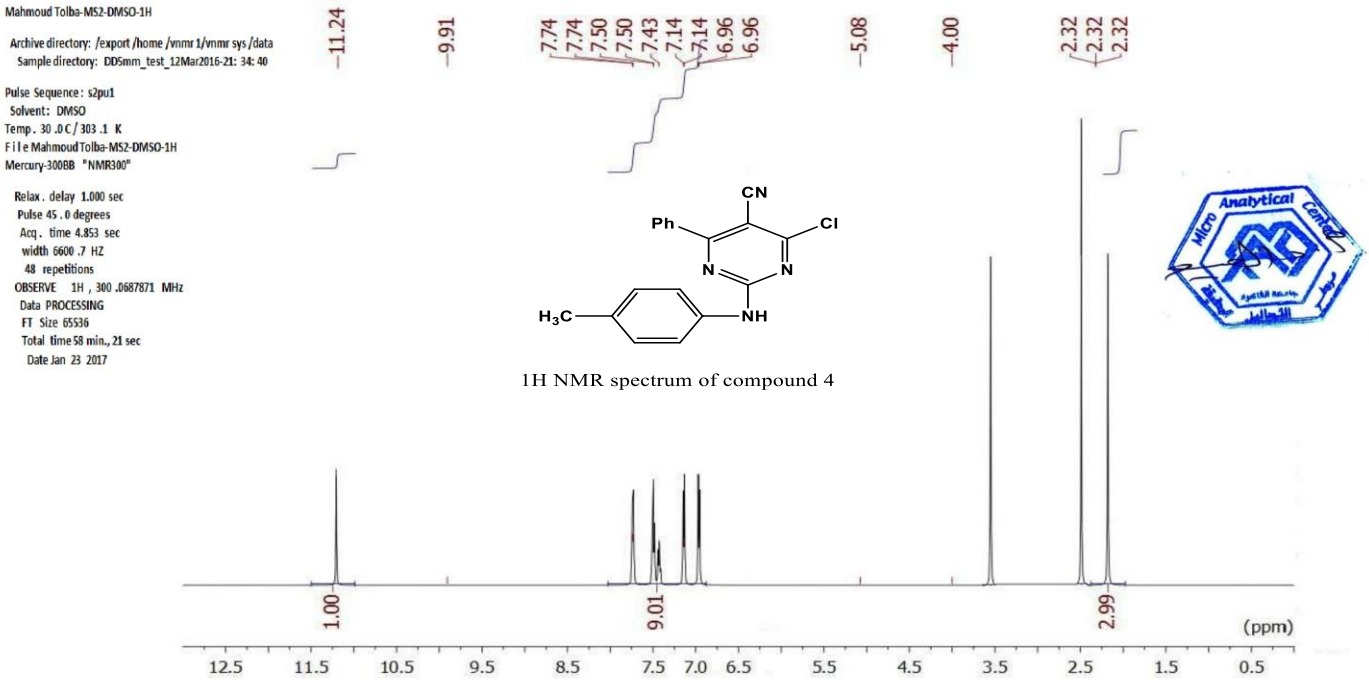
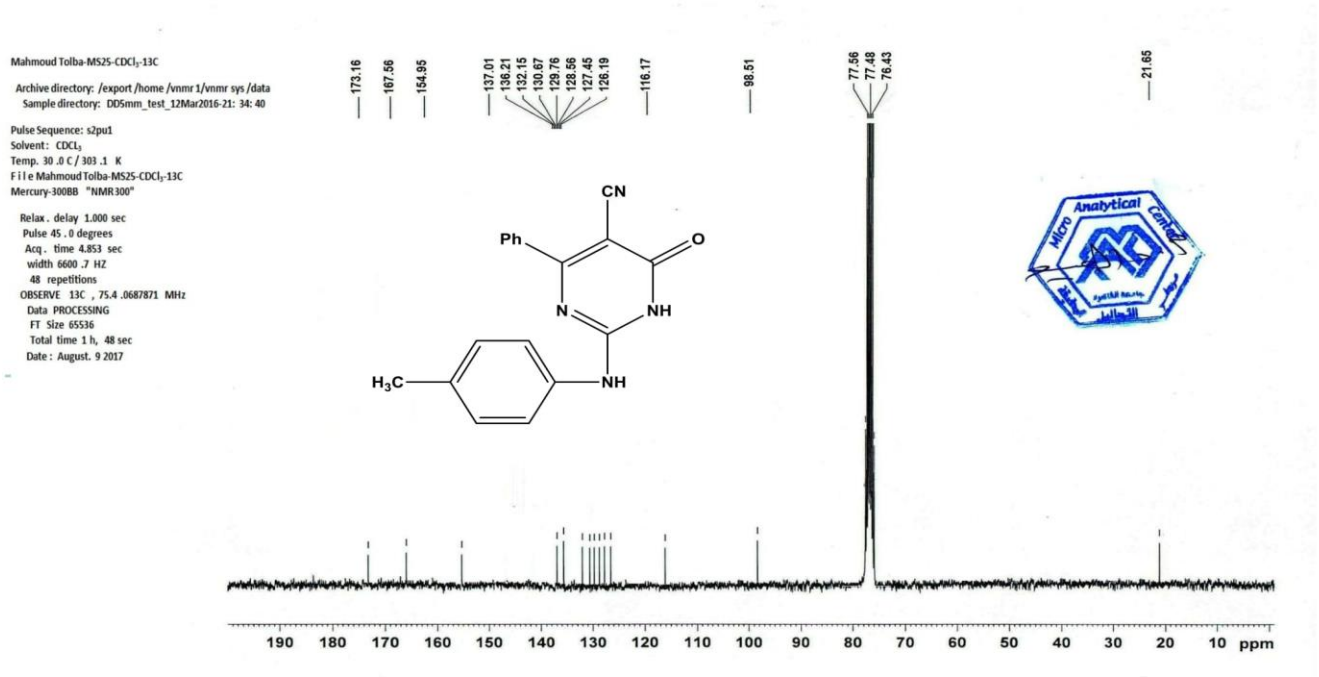
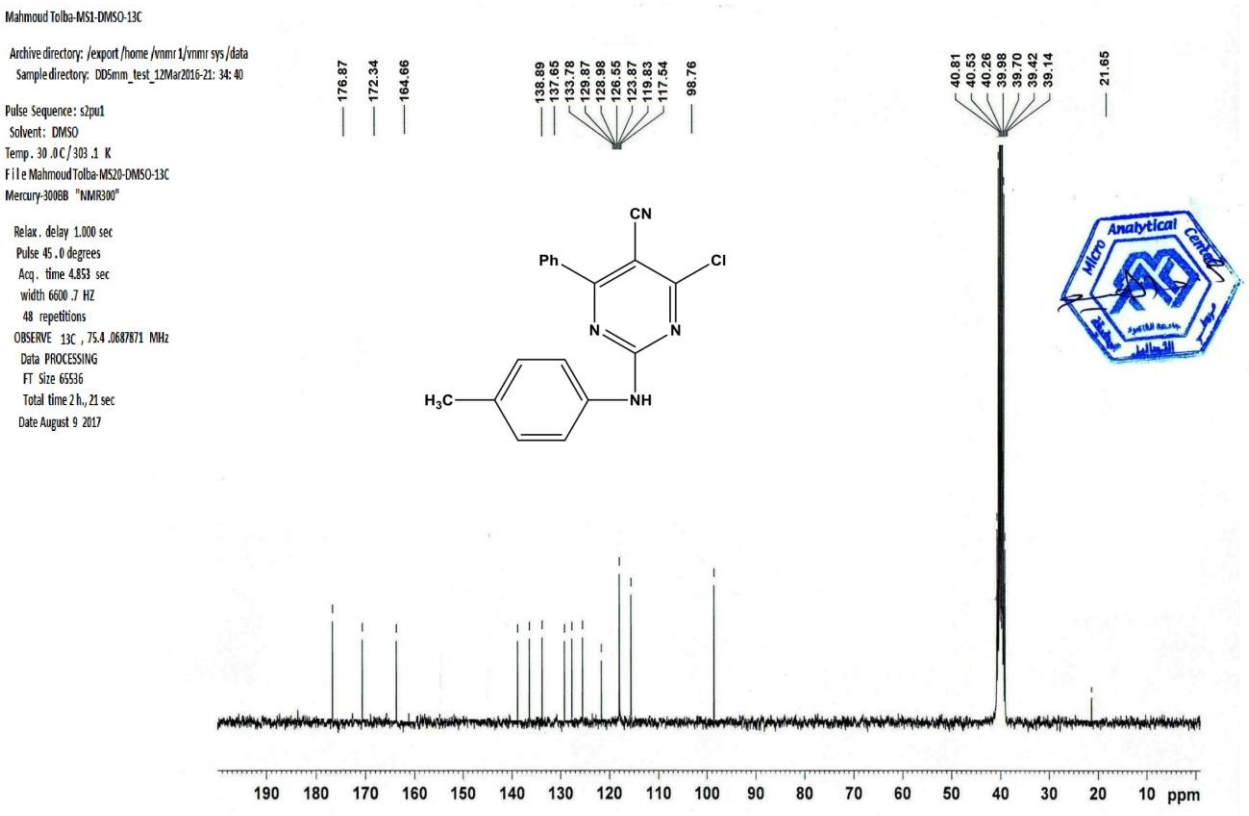


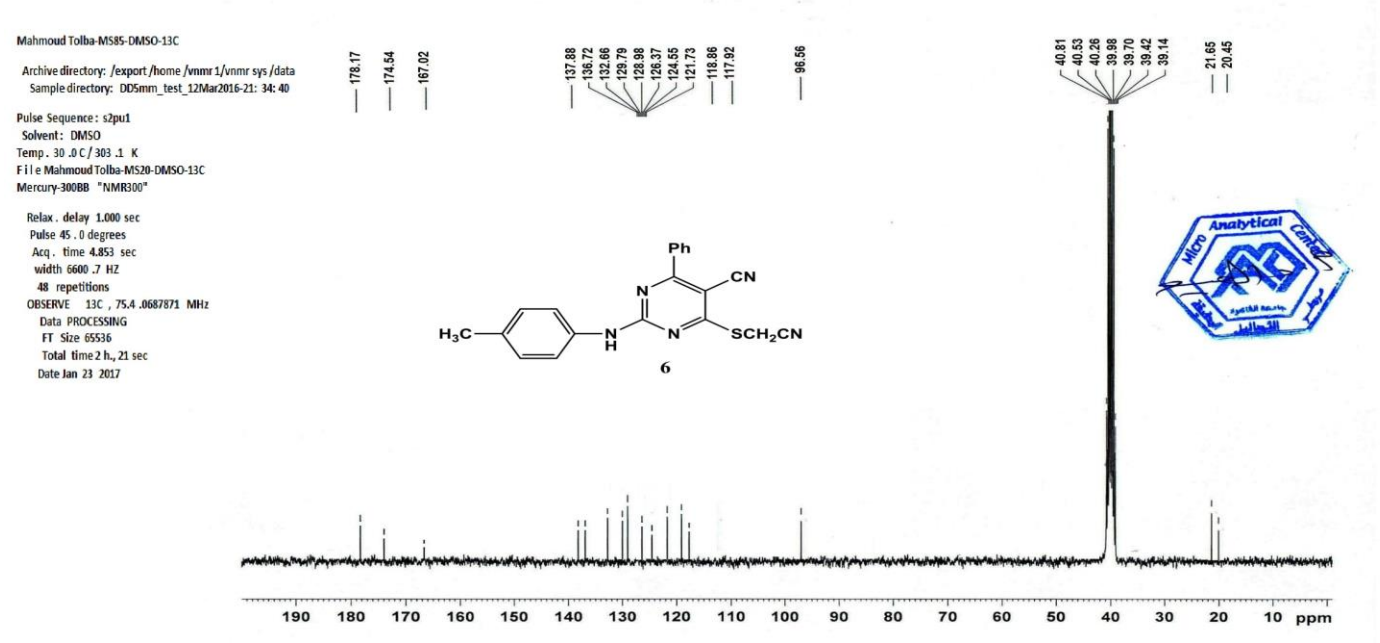
1H NMR spectrum of compound 4



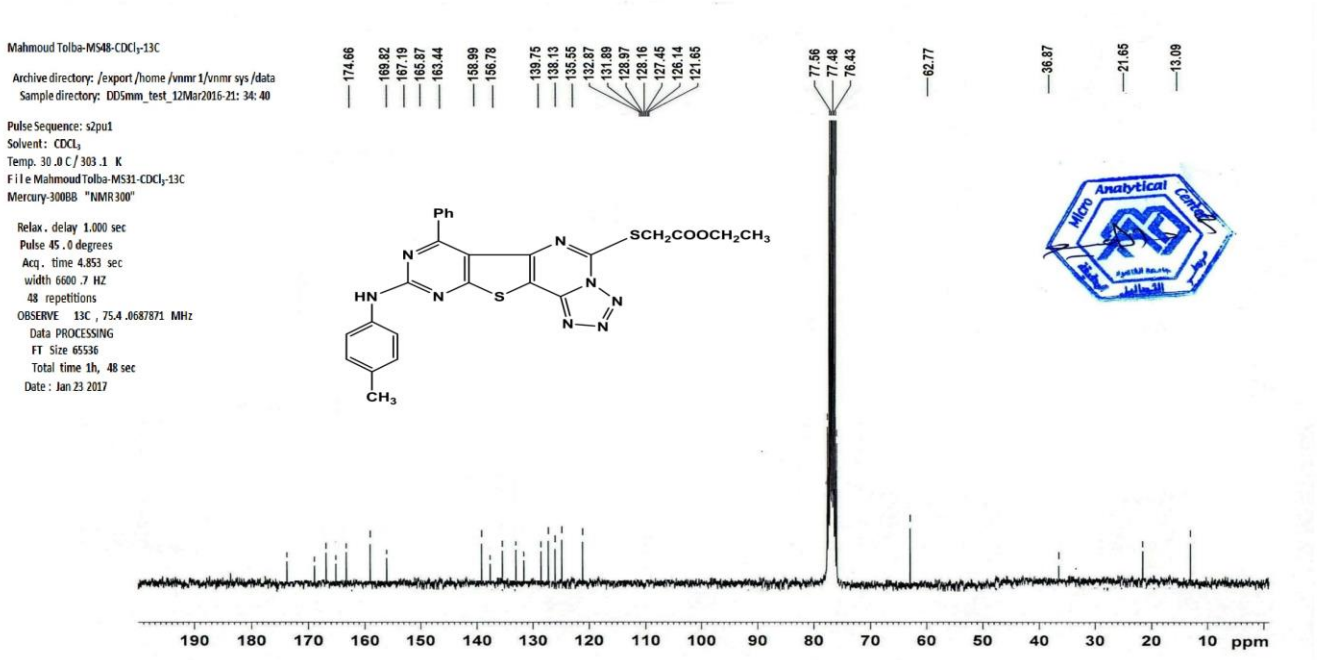
13C NMR of compound 3



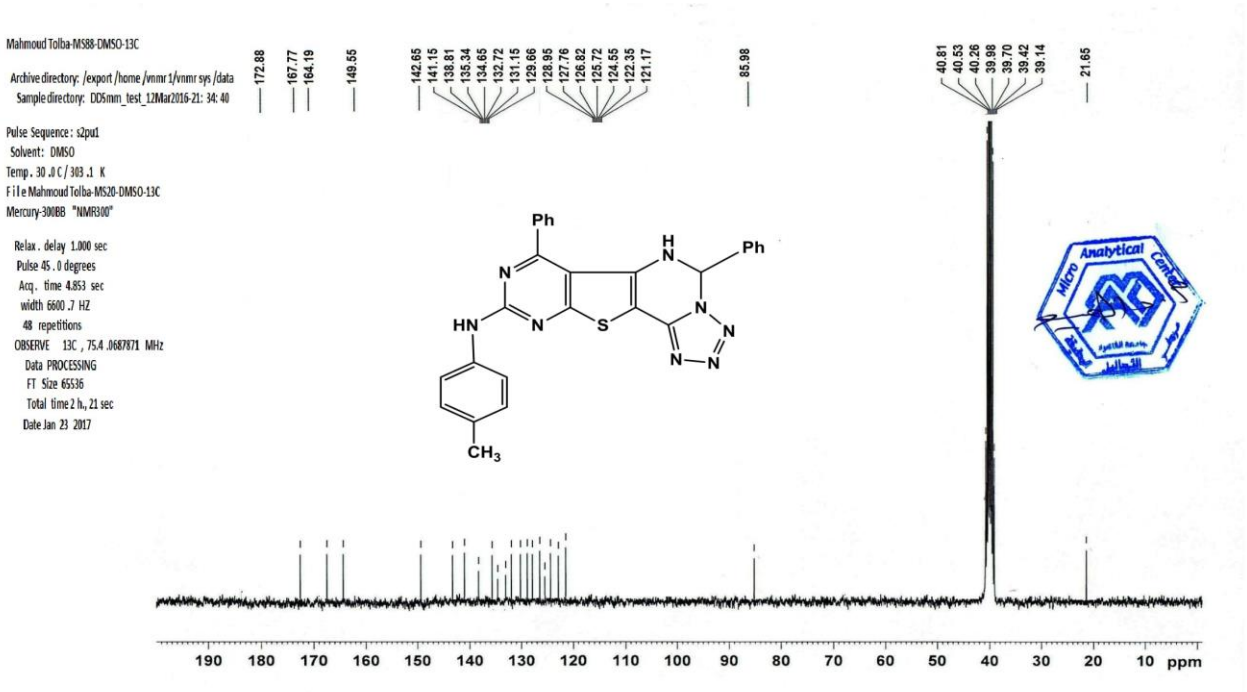
13C NMR of compound 4



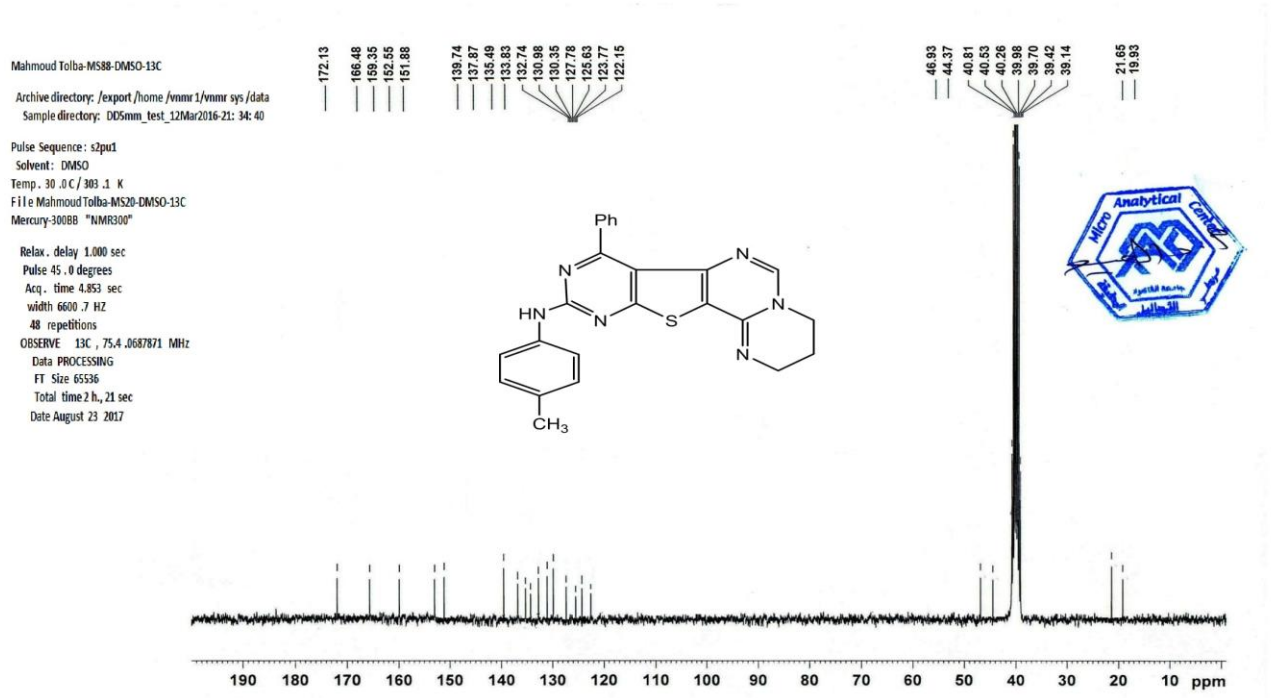
13C NMR of compound 6



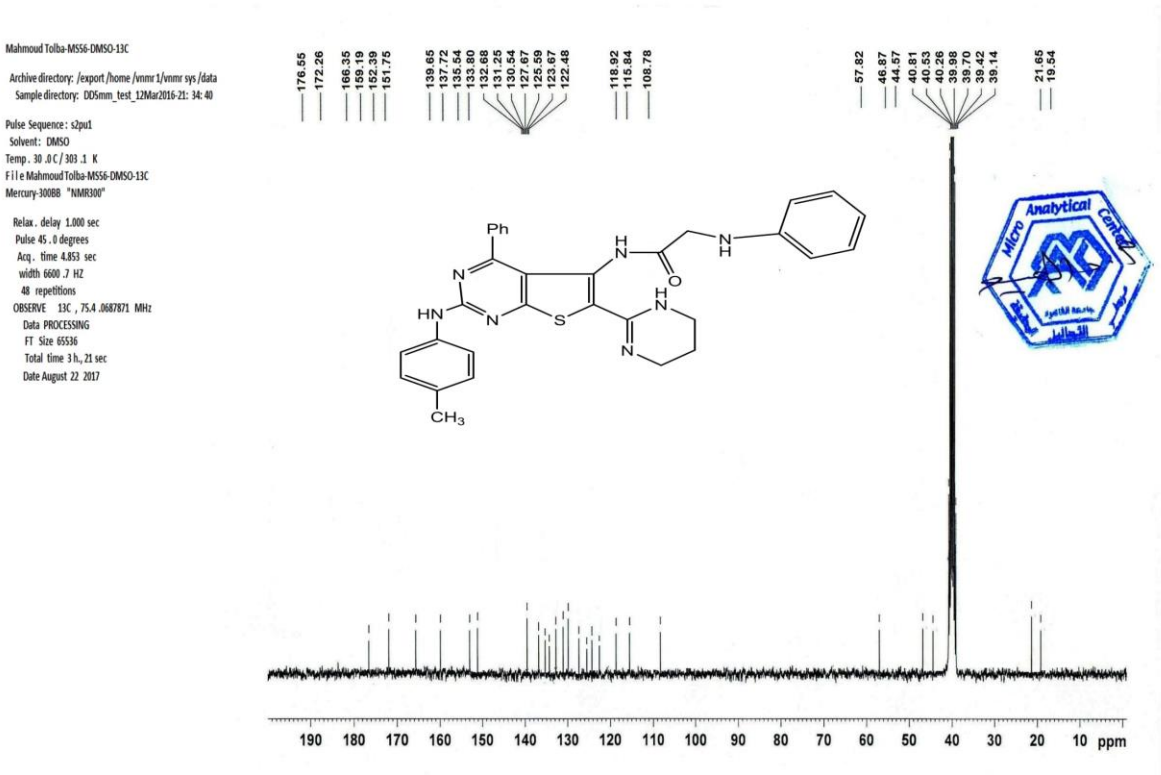
13C NMR of compound 12 a



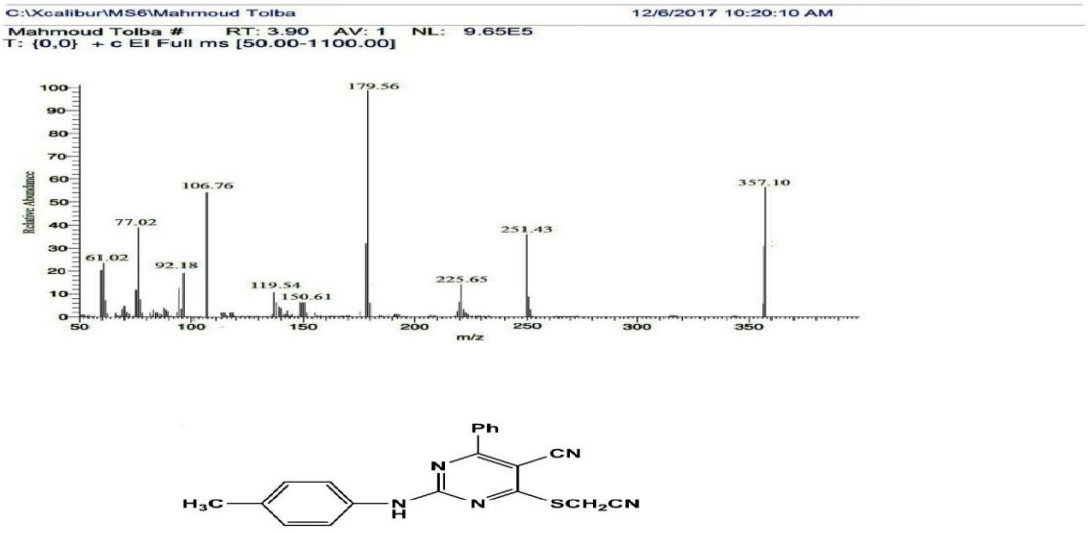
13C NMR of compound 10



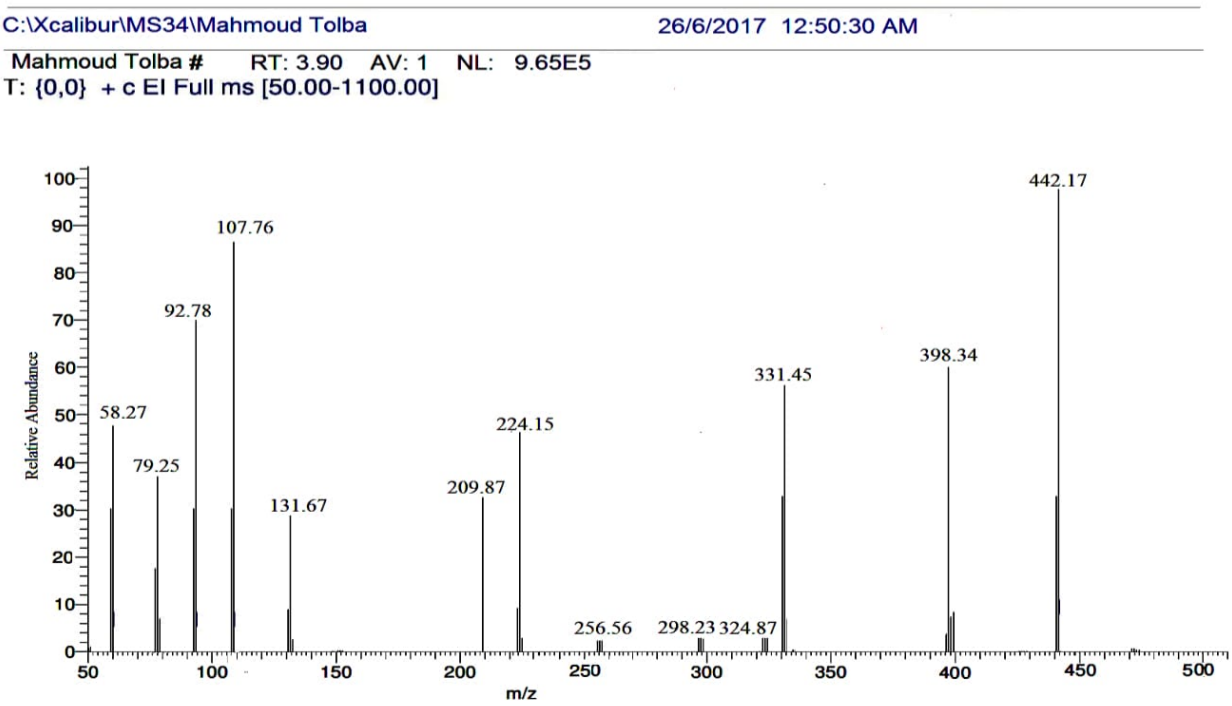
13C NMR of compound 14

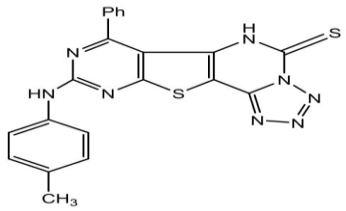


13C NMR of compound 18



Mass spectrum of compound 6





Compound 11

Antimicrobial screening

The antibacterial activity of all synthesized compounds was tested *in vitro* against *Staphylococcus aureus* and *Bacillus cereus* as Gram-positive bacteria and *Escherichia coli* and *Pseudomonas aeruginosa* as Gram-negative bacteria. The inhibition zone (mm) was compared with a series of antibiotics according to the sensitivity of each type of bacteria to the most effective antibiotic for it as a standard. The compounds were also screened for their antifungal activities against four antifungal species, *Candida albicans*, *Geotrichum candidum*, *Aspergillus flavus*, *Trichophyton rubrum*. The inhibition zone (mm) was compared with Clotrimazole as a standard. These strains are common contaminants of the environment in Egypt and all microbial strains were kindly provided by the Biotechology Laboratory, Assiut Sugar Technology Research, Assiut University . To prepare inocula for the bioassay, bacterial strains were individually cultured for 48 h in 100 mL conical flasks containing 30 mL nutrient broth medium. Fungi were grown for 7 d in 100 mL conicals containing 30 mL Sabouraud's dextrose broth. Bioassay was done in 10 cm sterile plastic Petri plates in which microbial suspension (1 mL/ plate) and 15 mL appropriate agar medium (15 mL/plate) were poured. Nutrient agar and Sabouraud's dextrose agar were respectively used for bacteria and fungi. After solidification of the media, 5 mm diameter cavities were cut in the solidified agar (4 cavities/plate) using a sterile cork borer. Chemical compounds dissolved in DMSO at 2% weight per volume (w/v) (=20 mg/mL) were pipetted in the cavities (20 μL/cavity). Cultures were then incubated at 28 ^{o}C for 48 h in case of bacteria and up to 7 d in case of fungi. Results were determined as the diameter (in mm) of the inhibition zone around cavities. To determine the MICs, chemical compounds giving positive results were diluted with DMSO to prepare a series of descending concentrations down to 0.02 mg/mL. Diluted chemicals were similarly assayed as mentioned before and the least concentration (below which no activity) was recorded as the MIC. The screening tests were carried out in triplicate and the results were expressed as a mean of three determinations.

