Exploring the Antimicrobial Properties of Functionalized calix[4] pyrrole

Bhagav P.Dave[1] and Keyur D.Bhatt[1*]

[1] Department of Chemistry, C U Shah University, Surendranagar – 363030, Gujarat, India
[2] Department of Chemistry, Mehsana Urban Institute of Science, Ganpat University, Kherva – 384012 , Mehsana, Gujarat, India
Corresponding Author: **Keyur D.Bhatt**
Department of Chemistry, Faculty of Science, Ganpat University, Kherva-384012, Mehsana, Gujarat, India
DOI: 10.54882/13202313202317690

Abstract
To determine a chemical's hazardous potential, antimicrobial activity research on several compound kinds is highly helpful. By adding different amounts of certain functionalized calix[4]pyrrole compounds to test organism culture, antibacterial activity has been evaluated. For known bacterial and fungal cultures, synthesised has been evaluated for a specific activity. As test organisms, Staphylococcus aureus, P. aeruginosa, S. pyogenes, and Escherichia coli bacterial subcultures as well as fungal subcultures of Candida albicans, Aspergillus nigricans, and Aspergillus clavatis have been utilised. At various concentrations, all materials were examined against these stains.

Keywords: Calix[4]pyrrole, Antimicrobial Activity, Antifungal, Antibacterial.

Introduction

[1-2] Supramolecular chemistry is a branch of macrocyclic chemistry that has grown in popularity among academics working in a variety of fields. Notably, key turning points in the area were the finding of novel forms of [3]macrocyclic hosts. [4]Crown ethers, [5]cyclodextrins, [6-8]calix[n]renes, and are a few examples of traditional macrocyclic hosts whose supramolecular chemistry has been studied. With cyclodextrins and crown ethers, calixarenes represent the 3rd generation of [9-10]supramolecular substrates.

The antimicrobial activity of calixpyrroles stems from their ability to interact with microbial cells and disrupt essential processes for survival. While the precise mechanisms of action may

vary depending on the specific calixpyrrole derivative and the targeted microorganism, several key modes of action have been proposed. One prominent mechanism is their interference with cell membrane integrity. Calixpyrroles have been shown to interact with the lipid bilayer of microbial cell membranes, causing structural destabilization and permeabilization. This disruption can lead to the leakage of cellular contents and eventual cell death.[11]Cornforth et al. Originally described the biological action of calixarenes in 1955. Their research focused on para-octyl-calix[8]arene, which has polyoxyethylene units on the bottom rim, and it shown that these compounds were effective against [12-14]tuberculosis. In the face of rising antimicrobial resistance and the growing threat of infectious diseases, researchers are continuously seeking novel and potent antimicrobial agents. One such class of compounds that has gained significant attention is calixpyrroles. Calixpyrroles are a unique group of macrocyclic compounds known for their diverse biological activities, including potent antimicrobial properties. In this article, we will explore the introduction of calixpyrrole as an antimicrobial agent, its mechanisms of action, and its potential applications in combating microbial infections.These chemicals are particularly promising since their mode of action differs significantly from those of the other medications now being used to treat tuberculosis and because the resistance to traditional chemotherapeutic treatments is rising. Up until Hwang's patent on the [15-17]anti-viral activity of several calixarene derivatives, no new research has been done on the biological effects of calixarenes for more than 40 years.

In present investigation we have reported the Antibacterial screening of Acetyle benzaldehyde functionalized Calix[4]Pyrrole (ABCP), Glycine modified ABCP (GABCP), Butyric acid functionalised Calix[4]pyrrole (BuACP) and Glycine derived BuCP (GBuACP).

Experimenital

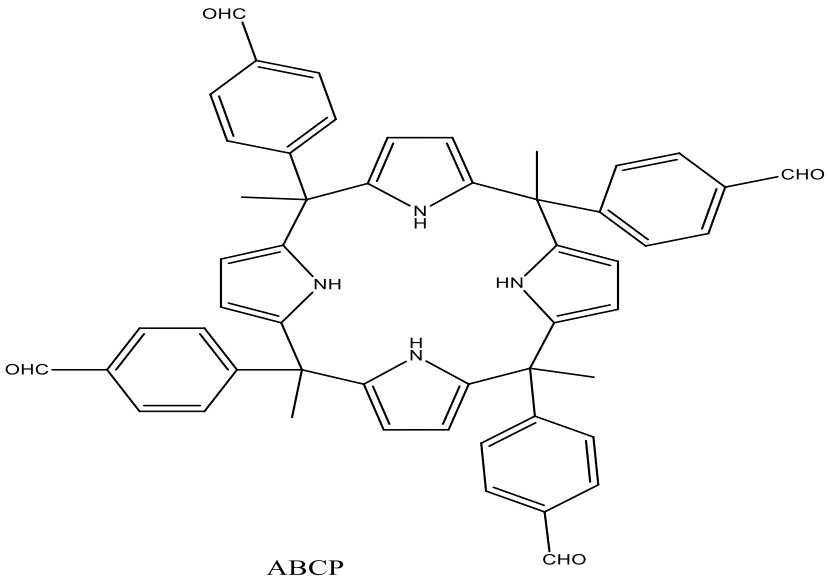
Instruments and Method used

Without additional purification, all of the chemicals were of AR grade and came from Sigma-Aldrich. All of the solutions were made from water that had been deionized and distilled in quartz, then further purified using a Millipore Milli-Q water purifier.

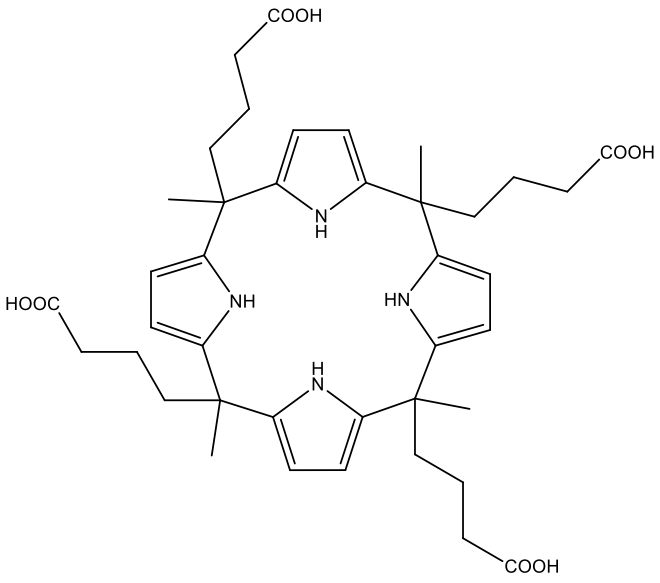
Melting points were measured using an uncorrected VEEGO (Model No: VMP-DS, India) melting point instrument in a single capillary tube. A CEM Discover Microwave Synthesiser was used for the microwave synthesis task. The Julabo F-25 bath was used for low-temperature processes. On a Brok Tensor 27 infrared spectrophotometer, FT-IR spectra of KBr pellets were captured and expressed in cm'1. Elementar Vario Micro Cube underwent an elemental analysis.
Up until recently, traditional culturing techniques were frequently used to investigate bacteria and fungus. Although still considered the gold standard, these methods are outmoded because to their lengthy nature, poor sensitivity, and inability to recognise particular cellular morphological states, such as the viable but non-culturable (VBNC) state, which causes false negative results. Traditional methods of microbial detection are being replaced by novel, rapid, and efficient diagnostics.

Synthesis of Benzaldehyde derivative of Calix[4]Pyrrole (ABCP) and Butyric acid derivative of calix[4]Pyrrole (BuACP) by Microwave irradiation method

The combination of 2.0 ml Pyrrole (0.03 mole), 0.2 ml CH3SO3H, and 20 ml methanol is then microwave-irradiated for 8 to 10 minutes. The pyrrole reaction mixture was then microwave irradiated for 8 to 10 minutes with every one and a half minute break. The solution of 4-formylacetophenone benzaldehyde (3 gm, 0.045 mole) produced in methanol (20 ml) was added. As soon as the reaction was finished, a dark brownish black colored mixture came out. The mixture was subsequently decanted in 100 ml of cold water to produce a precipitate of residue that was brown and black in colour. Filtered, dried, and then dissolved in a 30 ml solution of diethyl ether, the residue was. Once more, filtration was done to get rid of any black contaminants. After evaporation, the residual was double crystallised to form pure moiety for the further study.



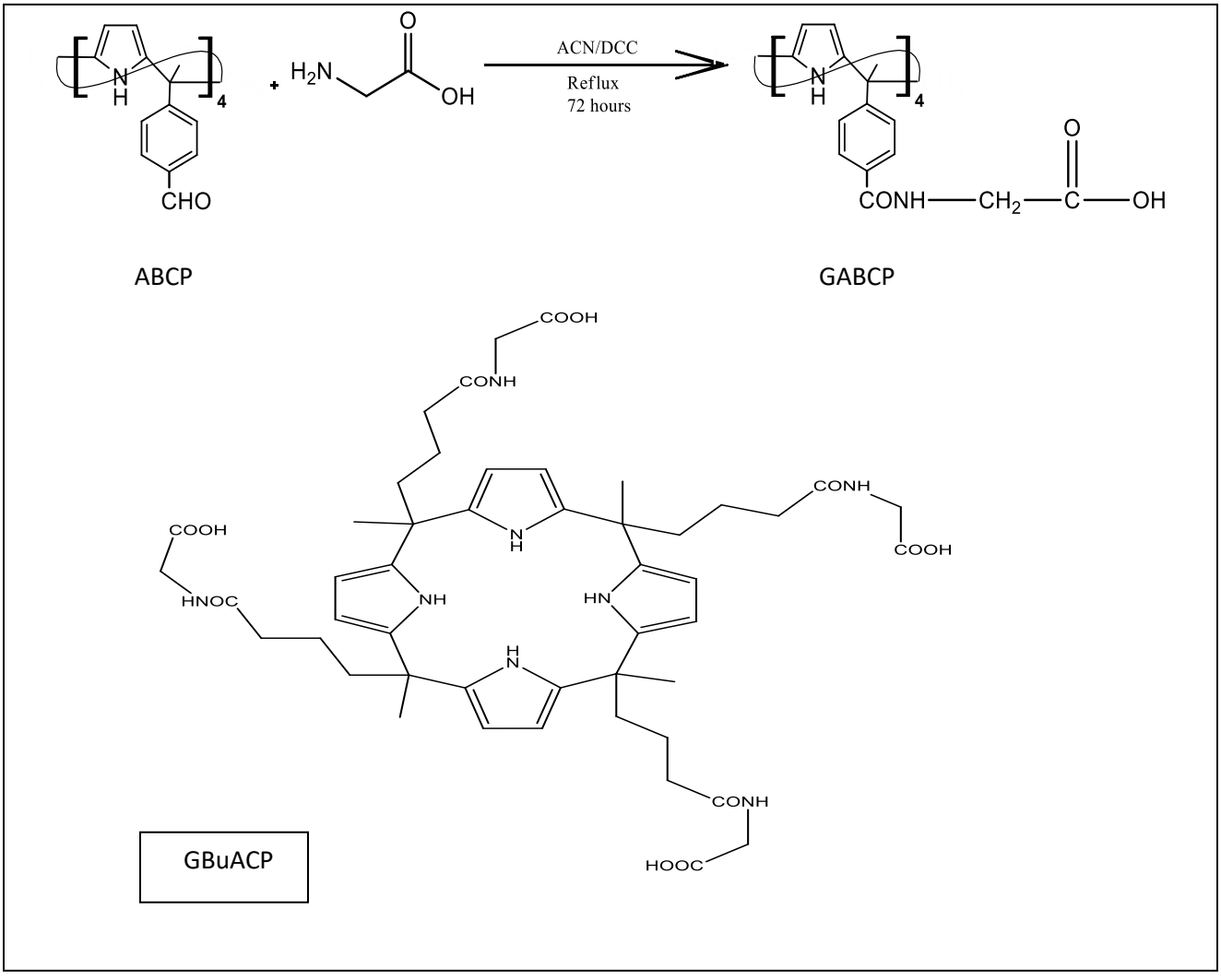
ABCP



BACP

Synthesis of Glycine Derivatives GABCP and GBuACP

The reaction mixture is prepared by dissolving 2 g of ABCP that is 0.0012 mole in 100 ml of acetonitrile, followed by the addition of 0.66 g of glycine that is 0.008 mole to the flask containing the ABCP, and 2 ml of N,N'-Dicyclohexylcarbodiimide - DCC as a acid amine coupling agent. After being refluxed for nearly 72 hours, the resulting mixture is then cooled to room temperature and poured into 100 cc of cold water to produce the residue. Filtered and dried the leftover material. The leftovers were once more dissolved in diethyl ether. After evoparation, the leftover material was recrystallized from methyle alcohol to produce the pure product.(Figure 2).



Antimicrobial activities

Evaluation of antibacterial activity utilising the general procedures listed below:

The new moiety's antibacterial and antifungal properties were evaluated in vivo. Staphylococcus aureus and streptococcus pyogenes were gram-positive bacteria, while Escherichia coli and pseudomonas aeruginosa were gram-negative.

Aspergillus niger, Candida albicans, and A. clavatus all showed antifungal activity. The antibacterial and antifungal activity was evaluated using the micro broth dilution method. The Surat Tuberculosis Center's Microcare laboratory provided standard strains for activities screening.

Standard medications like Ciprofloxacene, ampicilline, ,norfloxacene gentamycine, , and choloramphenicol were used to measureantibacterial activity, and gresofulvin and nystatin were used to measure antifungal action. Sabouraud The dextrose Broth was employed for fungal growth, and Mueller Hinton Broth was utilised as a nutritional media for bacteria.

Inoculum sizes were increased to 110 CFU/ml of experimental strains to assess turbidity. For both primary and secondary screening, successive dilutions were made. Both the conventional medications and the target substances were mixed together in DMSO-water at a concentration of 2.5 mg /ml.

For the primary screening, multiple concentrations of drugs, including 62.5,125, 250 and 500 microgram/mililiter, were used. Results for the initial standard was not collected because of the high DMSO content (12%).

The primary screening-active compounds were then employed for secondary screening for bacteria.

For secondary screening, the active drug was diluted at different concentrations, including 6.25 gm/ ml, 12.5 gm/ml, 25 gm/ml, 50 gm /ml, and 100 gm/ ml. The maximum dilution inhibition (which must be at least 99 percent) was used as the minimum inhibitory concentration (MIC) after an overnight incubation in an environment that was humid at 37 degree Celsius.(Jadhav et al. 2017).

Results and Discussion

The MIC value shows that the synthesized novel compounds exhibited modest to fine inhibition. An excellent antibacterial activity was shown by synthesized compound GABCP and GBuACP. **(Table 1, Figure 1)**. Compound GABCP and GBuACP showed excellent antifungal activity **(Table 2, Figure 2)**. Rest of all compound did not show better activity.

Table 1 : Antimicrobial Activity

Antimicrobial Activity Table					
Minimal Inhibition Concentration					
Sr.no	**Chemical Code**	**E.COLI MTCC 443**	**P.AERUGINOSA MTCC 441**	**S.AUREUS MTCC 96**	**S.PYOGENUS MTCC 442**
1	ABCP	250	500	250	125
2	GABCP	50	100	100	50
3	BuACP	250	250	125	125
4	GBuACP	125	100	50	100

Antifungal Activity Table				
Minimal Inhibition Concentration				
Sr.no	Chemical code	C.ALBICANS MTCC 227	A.NIGER MTCC 282	A.CLAVATUS MTCC 1323
1	ABCP	500	>1000	>1000
2	GABCP	62.5	100	100
3	BuACP	1000	>1000	500
4	GBuACP	250	500	250

Table 2 : Antifungal Activity

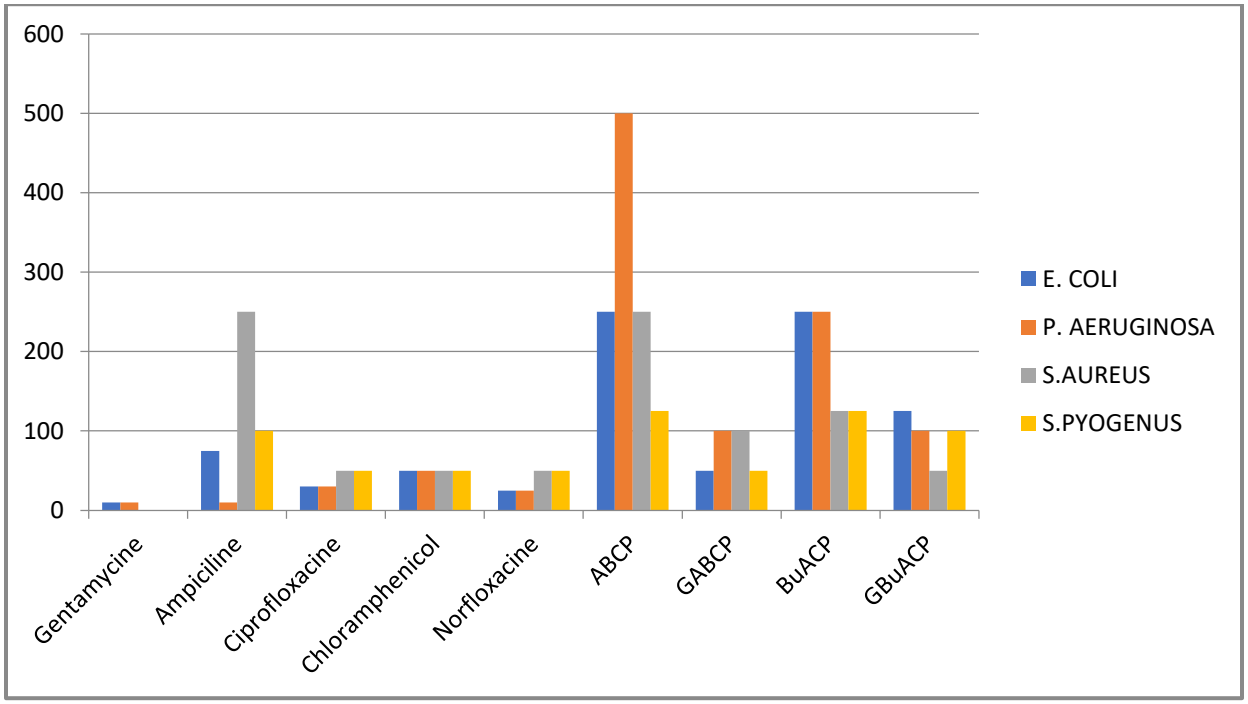


Figure 1 : Antimicrobial Activity Comparison Chart

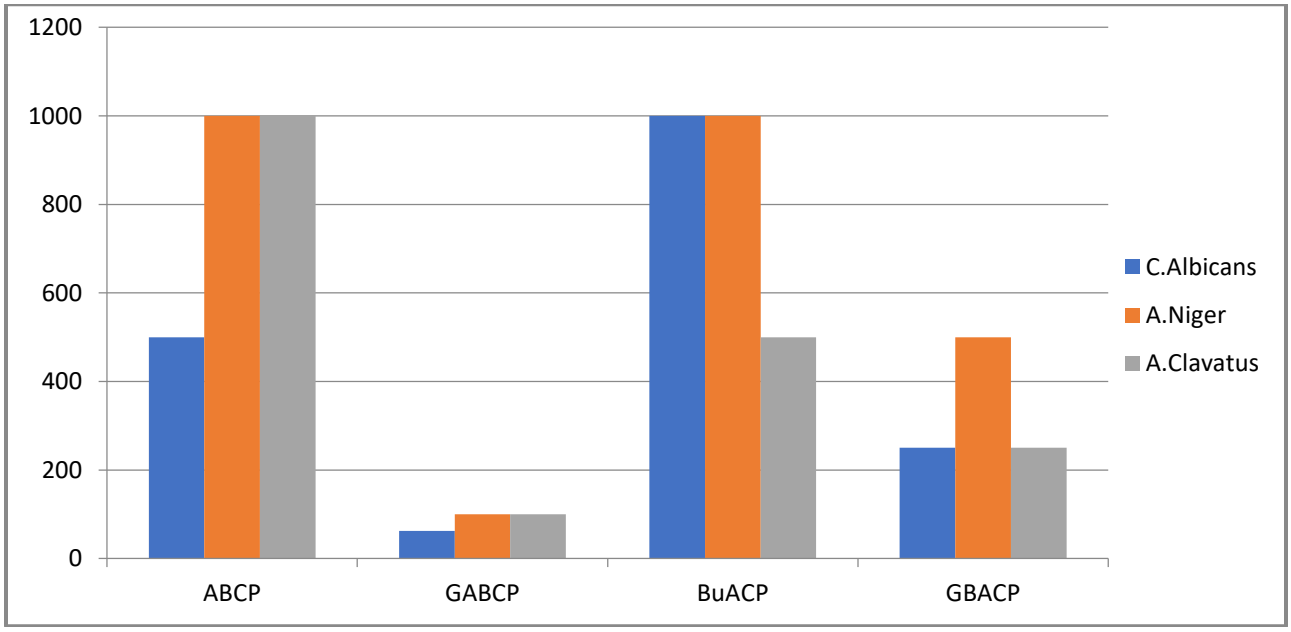


Figure 2: Antifungal Activity Comparison Chart

Conclusion

Two moieties ABCP and GABCP were synthesized and two novel moieties BuACP and GBuACP were synthesized and tested for the Antimicrobial activities and found that GABCP and GBuACP tested good active for Antibacterial infection and Antifungal infections.

Acknowledgement

The Author aknowledge C. U. Shah university, Surendranagar, for carried out the Research work for Antimicrobial study and Ganpat University, Mehsana, for the Analysis of the Synthesized compounds.

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