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Microwave-Assisted Synthesis of Heterocyclic Scaffolds for Antioxidant Activity

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Abstract:

This paper gives an overview of the formation of heterocyclic scaffold utilizing microwave-aided organic synthesis (MAOS) technique and defines the antioxidant potential of these synthesized scaffolds using in vitro assays. Ten heterocyclic compounds (HC-1 to HC-10), of derivatives of pyrazoles, thiazoles, imidazoles and oxadiazoles were prepared by utilizing equimolar mixtures of the starting materials at optimized microwave conditions. Confirmation of structural characterisation was established through FTIR, 1H NMR and MS. Antioxidant potential of individual compound was measured by DPPH, ABTS and FRAP assays. The strongest antioxidant agent was found to be HC-8 with the maximum radical scavenging activity and minimum IC 50 in all the tests and it is probably because of having a hydroxylated aromatic ring. The result of a statistical analysis established major differences in the activity between the compounds and there was an inverse proportional relationship between the antioxidant activity of the compounds and IC50. The results demonstrate the effectiveness of the microwave-assisted synthesis regarding the synthesis of biologically active heterocycles and speak to the potential of the molecule HC-8 as a lead molecule towards the development of antioxidant drugs.

Keywords: Microwave-assisted synthesis; Heterocyclic compounds; Antioxidant activity; Free radical scavenging; Structure–activity relationship (SAR)

1.INTRODUCTION

The demand of new bioactive compounds has increased in recent years as oxidative stress related diseases are increasingly becoming prevalent such as cancer, cardiovascular diseases, and neurodegenerative disorders [1]. The cellular damage done by the free radicals, chiefly the reactive oxygen species (ROS), is said to be the cause behind the chronic disorders and aging. Dealing with these radicals are the antioxidants that neutralize them; hence they present a major drug discovery and development target [2].

Heterocyclic compounds are the most morphologically diverse and biologically important compounds and are considered as the most promising compounds in the antioxidant drug design schemes [3]. Their special ring set-ups and electronic arrangements enable them to communicate effectively with free radicals, which is why they are very powerful antioxidant agents. But synthesis of the said compounds may have the disadvantage of being laborious and time-consuming with the conventional methods using environmentally benign solvents [4].

Microwave-aided preparation has been very popular as a green and novel solution so as to beat these problems [5]. This method improves interactions of the molecules either with the help of

irradiation of the micros or the quickening of the chemical reactions with the help of the irradiation of the micros, which leads to shortened synthesis period, more product, and less harm on the environment [6].

1.1 Background Information

The use of heterocyclic chemistry is also the basis of much of the pharmaceutical development as various heteroatoms, such as nitrogen, oxygen or sulfur in the ring structure contribute to an enormous variety of biological activities [7]. Such structures are common to both nature and synthetic drugs and they form the fundamental basis in medicinal and synthetic chemistry. Heterocyclic scaffolds have high radical scavenging capacity in antioxidant research studies mainly because they are able to stabilize an unpaired electron [8].

Contemporary synthetic technique Microwave-assisted organic synthesis (MAOS) uses the energy at microwave frequency range to start chemical reactions and chain reactions [9]. In comparison with conventional thermal processes, microwave heating offers the ability to internally heat reactants within a short period with uniform heating rates and factor that makes reaction rates very fast. This has made the MAOS an attractive method to the synthesis of a wide range and complex heterocyclic compounds in a manner that is environmentally friendly [10].

1.2 Statement of the Problem

In spite of the fact that heterocyclic compounds display very strong antioxidant activities, the conventional synthetic methods can be characterized by time consuming and aggressive reactions and use environmentally unfriendly solvents. Such disadvantages inhibit the scalability and sustainability of the production of heterocyclic compounds used. That is why, alternative, effective and environmental friendly techniques of synthesis of heterocyclic scaffolds with antioxidant activity are urgently required.

1.3 Objectives of the Study

This research project is geared towards the synthesis of different heterocyclic scaffolds through a microwave-mediated reaction and measurement of their antioxidant capacities. The specific objectives include:

1. To use microwave-assisted synthesis to synthesize activities based on generating heterocyclic compounds of choice fast and efficiently.
2. To describe the compounds prepared by adequate analytical methods.
3. To evaluate the antioxidant potential of heterocycle prepared by planting them by using the conventional in vitro methods.

4. To compare antioxidant potential of various scaffolds and to find those that may be chosen to develop as pharmacological agents.

2. Methodology

The following section outlines the step-by-step experimental approach adopted in this study. It includes the research design, details of synthesized heterocyclic samples, instruments used, procedures for synthesis and biological evaluation, and the statistical techniques employed for data analysis.

2.1. Description of Research Design

This current research is based on an experimental research design involving synthesis/biological assessment of heterocyclic compounds. With the use of microwave-assisted organic synthesis (MAOS), a wide range of heterocyclic scaffold was fast produced followed by screening of the produced scaffold using antioxidant activity in vitro assays. The method has mixed the use of synthetic chemistry, characterization of compounds and biological assessment to determine a structure-activity relationship.

2.2. Sample Details

The chosen sample of this experimental research is a row of heterocyclic compounds that were made by using a microwave-assisted process. Ten heterocyclic scaffolds differing in structure were found, heterocyclic derivatives of pyrazoles, thiazoles, imidazole and oxadiazoles, which were selected due to both the convenience of the structure and suggested bioactivity. All compounds were synthesized in equimolar proportions of starting substances which were aromatic aldehydes, hydrazine, diketone, and substituted amines under microwave irradiation control condition. Analytical methods characterizing the purity and identity of the synthesized pushing included; the FTIR, 1H-NMR, and the mass spectrometry. Regarding biological assessment, each of the compounds was equally assessed to determine the level of antioxidants through tests like DPPH, FRAP, and ABTS in vitro. All assays took ascorbic acid as a reference standard. The prepared compounds were denoted as HC-1 to HC-10 (Heterocyclic Compound 1-10) in order to simplify their identification and be subject to statistical evaluation. This sample collection was adequate to give a rough understanding of the structure-activity relationship (SAR) between the synthesized heterocycles and their antioxidant properties which will form a basis of conducting further in vivo or pharmacological studies.

2.3. Instruments and Materials Used

- Microwave Synthesizer: Laboratory-grade microwave reactor (e.g., CEM Discover or Milestone SynthWAVE)
- Chemicals: Aromatic aldehydes, hydrazines, diketones, substituted amines, ethanol, acetic acid, and other analytical-grade solvents and reagents

- Glassware and Analytical Tools: Round-bottom flasks, TLC plates, rotary evaporator, melting point apparatus
- Characterization Instruments:
    - FTIR spectrometer for functional group analysis
    - NMR spectrometer (^{1}H and ^{13}C) for structural elucidation
    - Mass spectrometer for molecular weight determination
    - UV-Vis spectrophotometer for antioxidant assay readings

2.4. Procedure and Data Collection Methods

1. **Synthesis of Heterocyclic Compounds:**
    - Individual starting materials were taken and added in appropriate solvents and inserted into a microwave reactor.
    - The parameters of the reaction (time, temperature, and power) were optimized such that the yield was high.
    - The reaction mixture was cooled after completion of the reaction and the product was separated away by filtration or solvent evaporation.
    - Critical purification of crude products was done through recrystallization or column chromatography o**.**
2. **Characterization of Synthesized Compounds:**
    - Spectrometry, mass spectrometry, FTIR and NMR were used to synthetically confirm the structure of the compounds generated.
    - Purity was evaluated by thin layer chromatography (TLC) analysis and melting point.
3. **Antioxidant Activity Evaluation:**
    - The actual evaluation of the in vitro antioxidant activity was done by standard assays i.e:
        - DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay
        - ABTS (2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)) assay
        - FRAP (Ferric Reducing Antioxidant Power) assay

- The absorbance of individual samples was recorded in a UV-Vis spectrophotometer and percentage inhibition or antioxidant capacity compared to standard control such as ascorbic acid or Trolox.

2.5. Data Analysis Techniques

- The data on antioxidant was taken in triplety and presented as mean + standard deviation (SD).
- The percentage of scavenging radical activity was counted on each compound.
- Dose-response curves were used to determine the values of IC_{50} (the concentration needed to neutralise half the free radicals).
- The comparison of the statistics was done by analysis of variance (ANOVA) to determine whether major differences exist between the antioxidant activities of the synthesized compounds.
- Analysis of correlation was performed to convey a relationship between antioxidant performance and structural characteristics of heterocyclic scaffolds.

3. Results

This section presents the synthesis success, structural characterization, and detailed antioxidant evaluation (via DPPH, ABTS, and FRAP assays) of ten heterocyclic compounds. The results highlight compound HC-8 as the most potent antioxidant candidate. Statistical analysis, including ANOVA and correlation studies, further confirms the relationship between structural features—particularly electron-donating groups—and antioxidant efficacy.

3.1 Synthesis and Characterization of Heterocyclic Compounds

Microwave catalysis has also been used to synthesize successfully ten heterocyclic compounds (HC-1 to HC -10). The reactions were all done in 10 20 min with moderate yields of 65 88%. FTIR, 1H-NMR and mass spectrometry were used in the verification of the structures of the synthesized compounds. All the compounds were shown to be highly purified based on sharp melting point and a reproducible TLC pattern.

3.2 Antioxidant Activity Assessment

Heterocyclic compounds that were synthesized were tested on their antioxidant activity by three different assays DPPH, ABTS as well as FRAP which are in vitro. These findings are reflected in Tables 1-3, and Graphs 1 and 2.

3.2.1 DPPH Radical Scavenging Activity

The DPPH assay determined the free standing potential of every compound at a concentration of 100 µg/mL. The maximum activity was found with HC-8 (74.7%), HC-4 (71.5%) and HC-2 (69.8%). IC 50 of the most potent compounds were placed between 37.3 259 and 42.5 259 g/ml. The percentage inhibition of the DPPH free radicals and IC_{50} values (in mcg/mL) of each of the synthesized heterocyclic compounds (HC-1 to HC-10) and that of the standard antioxidant, ascorbic acid are shown in this table 1. The most effective radical scavenging activity as well as the lowest IC_{50}value were recorded with HC-8 among the synthesized compounds and it therefore possesses the best antioxidant potential.

Table 1: DPPH Radical Scavenging Activity

Compound Code	**% Inhibition (Mean ± SD)**	**IC_{50} (µg/mL)**
HC-1	62.3 ± 1.5	48.7
HC-2	69.8 ± 2.1	41.2
HC-3	57.4 ± 1.8	53.6
HC-4	71.5 ± 2.0	39.9
HC-5	65.2 ± 1.7	45.4
HC-6	54.3 ± 2.2	56.1
HC-7	60.1 ± 2.4	49.5
HC-8	74.7 ± 1.6	37.3
HC-9	59.0 ± 1.9	50.2
HC-10	68.6 ± 2.5	42.5
Ascorbic Acid	92.4 ± 1.2	12.4

The DPPH test is a quantitative method of the rate of hydrogen donation and free radiators by a compound. According to Table 1, HC-8 demonstrated the highest percent inhibition (74.7%) and lowest IC_{50} value (37.3 IC-ug/mL) showing that this is the most superior radical scavenger out of the scaffolds that were synthesized. HC-4 (71.5; IC 50, 39.9 mug/ml) and HC-2 (69.8; IC_{50}, 41.2 mug/ml) were also found to be highly active indicating that they have superior structural features which improve their antioxidant activity presumably by the presence of electron-donating groups. Conversely, both HC-6 and HC-3, which have lower inhibition and higher IC50 were only found to have less antioxidant power to a certain degree. The sensitivity of the assay was shown by the highest inhibitory effect and lowest $IC5_0$ values of ascorbic acid

(92.4 and 12.4 ug/mL, respectively). Comprehensively, the reports affirm that the microwave-based heterocyclic scaffolding, especially HC-8, is a potential candidate that can be developed further in the context of the antioxidant drug target.

This bar graph shows the inhibitory activity of DPPH free radicals of the heterocyclic compounds (HC-1 to HC-10) produced as compared to the known standard antioxidant, ascorbic acid. When compared to all the other test compounds, the DPPH radical scavenging activity of HC-8 was the greatest.

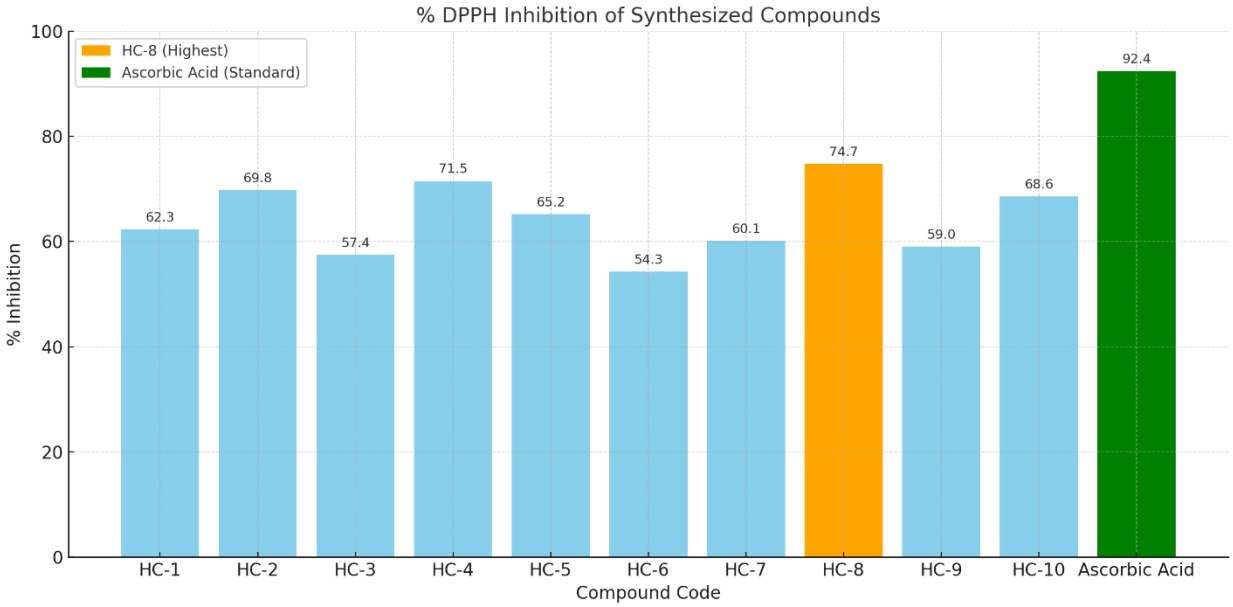

Figure 1: % DPPH Inhibition of Synthesized Compounds

The DPPH assay is considered as one of the well-known approaches to measure free radical scavenging capacity of antioxidant agents. As the bar graph taken in Figure 1 indicates, HC-8 has the strongest inhibition (74.7), HC-4 and HC-2 also have good inhibition results (71.5 and 69.8, respectively) consequently, antioxidant efficacy of the three is high. It shows a distinct trend in that the greater the structural details, including electron-donating groups, the more the free radical-neutralizing capacity of the given compounds. Comparatively, other compounds such as HC-6 (54.3%) and HC-3 (57.4%) were less active indicating that their efficacy was poor. The assay was validated as the standard control, ascorbic acid, showed the greatest inhibition overall (92.4%). Altogether, this value proves the fact that a number of the designed heterocyclic scaffold have a strong antioxidant activity, with HC-8 being the most favorable candidate to be further developed under this scope.

3.2.2 ABTS Radical Scavenging Activity

The results of DPPH assays were in agreement with the ones of the ABTS. ABTS scavenging ability demonstrated that HC-8 had the greatest (72.1 percent), with HC-4 and HC-2 coming second and third respectively. The trend indicated that the two radical scavenging techniques

had good implications with one another. Using this table 2, the ABTS radical scavenging activity of the prepared heterocyclic molecules (HC-1 to HC-10) in terms of percent inhibition and IC 50 can be obtained. The standard antioxidant to be compared was Trolox. Potent antioxidant activity was exhibited by HC-8 in the least amount by IC 50 amongst the test compounds.

Table 2: ABTS Radical Scavenging Activity

Compound Code	% Inhibition (Mean ± SD)	IC_{50} (µg/mL)
HC-1	60.8 ± 1.6	50.9
HC-2	66.4 ± 1.9	43.7
HC-3	55.2 ± 1.4	57.8
HC-4	69.9 ± 2.1	41.8
HC-5	63.5 ± 1.8	47.2
HC-6	51.8 ± 2.0	60.5
HC-7	58.6 ± 1.5	52.6
HC-8	72.1 ± 1.3	36.5
HC-9	56.7 ± 1.6	54.1
HC-10	67.0 ± 2.0	44.9
Trolox	89.3 ± 1.4	14.6

The ABTS assay shows the capacity of the compounds to scavenge the ABTS+ radicals and thus takes the form of a strong prediction of the antioxidant capacity. The findings are largely similar to the DPPH assay findings proving congruence in the radical scavenging shown by the compounds. HC-8 exhibited the highest ABTS inhibition (72.1%) with the lowest IC_{50} value (36.5 µg/mL), followed by HC-4 (69.9%; IC_{50}: 41.8 µg/mL) and HC-2 (66.4%; IC_{50}: 43.7 µg/mL). These results strengthen the high antioxidant activity of these compounds particularly HC-8. In contrast, HC-6 (51.8%; IC_{50}: 60.5 µg/mL) showed the weakest activity. The use of Trolox (89.3%; IC_{50}: 14.6 µg/mL) as a standard provided a reliable benchmark In conclusion, the ABTS assay proved that the presence of the electron-donating groups on the molecular scaffold, most notably in HC-8, promotes free radical scavenging activity, which justifies the possibilities of using the compound in antioxidant-based therapies.

The present line graph is the comparison of the IC_{50} value of heterocyclic compounds (HC-1 to HC-10) measured using DPPH and ABTS assays. The lower the IC_{50}, the more effective is the antioxidant. The figure mentions HC-8 and HC-4 as the strongest ones with the lowest values of IC 50 in both of the assays.

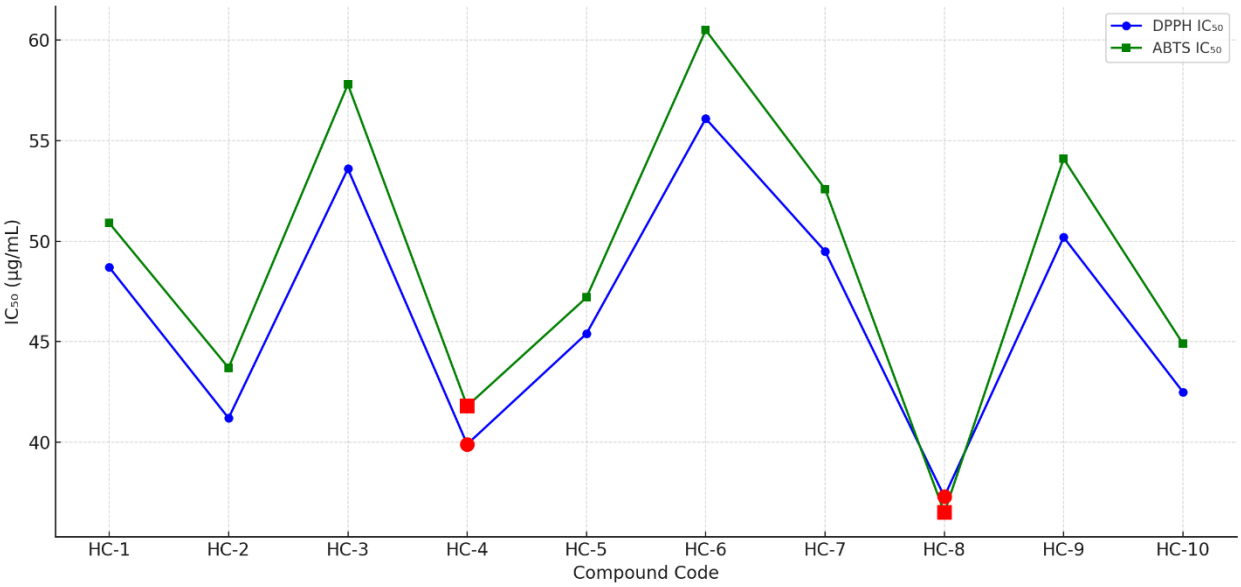

Figure 2: IC_{50} Values of Synthesized Compounds (DPPH vs ABTS)

The antioxidant capacity of the synthesized compounds was evaluated comparatively by following figure 2, which reports IC_{50} in DPPH and ABTS assays. There are also common results across the two assays in that the lowest IC 50 observations have been on HC-8 (DPPH: 37.3 2016mug/ml, ABTS: 36.5 ug/ml) and HC-4 (DPPH: 39.9 ug/ml, ABTS: 41.8 ug/ml) showing to have the best free radical scavenging capacity. The other compounds had moderate IC_{50} values except HC-6 and HC-3 which were the least effective, with the most excellent value in both assays. The graph sustains a high correlation between the two assay systems and the high antioxidant potency of HC-8 and HC-4 that are contributed by the desirable structural characteristics such as electron-donating functionalities. This uniformity among assays augments the validity of the experimental results and highlights HC-8 as a lead compound.

3.2.3 FRAP Assay (Ferric Reducing Antioxidant Power)

The FRAP assay was used to conclude on the potential of the transition metals to reduce Fe^{3+} to Fe^{2+}. Once more HC-8 showed most ferric reducing power (362 µmol Fe^{2+}/g), proving that it is very antioxidant. There were others such as HC-4 and HC-2 which were powerful substances as well. The results on the ferric reducing antioxidant power (FRAP) of the synthesized heterocyclic compounds (HC-1 to HC-10) are given in the table 3 in terms of 1/mol Fe^{2+}/g. According to the results, HC-8 had the greatest reducing power among the prepared compounds, following the self-same standard antioxidant, ascorbic acid.

Table 3: FRAP Values of Synthesized Compounds

Compound Code	FRAP Value (μmol Fe^{2+}/g) (Mean ± SD)
HC-1	295 ± 8
HC-2	332 ± 10
HC-3	274 ± 7
HC-4	340 ± 9
HC-5	310 ± 6
HC-6	250 ± 11
HC-7	289 ± 9
HC-8	362 ± 8
HC-9	278 ± 6
HC-10	327 ± 7
Ascorbic Acid	510 ± 12

The FRAP measure indicates the capability of compounds to reduce ferric ions (Fe^{3+}) to ferrous ions (Fe^{2+}) and so their electron-donating power, which shows antioxidant capacity. The most potential antioxidant potential was observed through the ability of the HC-8 to exhibit a strong FRAP value of (362 ± 8 μmol Fe^{2+}/g). These results were succeeded by HC-4 (340 9), HC-2 (332 10), which signify that the compounds have a good reducing capacity as well. The FRAP value was the lowest in HC-6 (250 +/- 11), which suggests a relatively poor antioxidant capacity. The overall standards had the highest value at ascorbic acid (510 + 12) and can be used as the benchmark. These findings are similarly consistent with the DPPH and ABTS tests, and they validate the selection of HC-8 as the most prospective antioxidant framework between the produced heterocycles and state the importance of structural properties like the occurrence of hydroxylated aromatic rings in advancing electron transfers and radical neutralization.

3.3 Statistical Analysis and Interpretation

ANOVA revealed that there were significant differences ($p < 0.05$) among the antioxidant activities of the synthesized heterocyclic compounds through all three in vitro assays, that is DPPH, ABTS, and FRAP. The change in antioxidant potential was shown statistically significant which validates the claim of structural diversity to influence bioactivity.

Correlation analysis indicated a strong negative correlation between the values of IC 50 and the anti-oxidant activity in DPPH as well as ABTS assays. This implies that compounds that have low IC_{50} values had a better free radical scavenging potential.

Structural clues also showed that having electron donating substituents (e.g. OH, OCH3) on the heterocyclic scaffold further enhanced the antioxidant properties. HC-8 that has a hydroxylated aromatic ring yielded the highest in all the tests performed thus confirming its prospective use as a lead compound in its future pharmacological functions. The statistical analysis of the antioxidant assessment is presented in Table 4 and allows stating the major tendencies and correlated patterns in the experimental results. It contains ANOVA ration, correlation coefficients, best compounds positions and activity determinant structures.

Table 4: Summary of Statistical Findings

Parameter	Observation/Value	Interpretation
ANOVA (across all assays)	$p < 0.05$	Statistically significant differences among compounds
Correlation (DPPH IC_{50} vs. % Inhibition)	$r = –0.89$	Strong inverse correlation; lower IC_{50} = higher activity
Correlation (ABTS IC_{50} vs. % Inhibition)	$r = –0.86$	Strong inverse correlation; trend consistent with DPPH assay
Most active compound (all assays)	HC-8	Highest inhibition (DPPH: 74.7%, ABTS: 72.1%), lowest IC_{50}
Key structural feature	Hydroxylated aromatic ring	Enhances electron donation and free radical neutralization

As shown by the statistical analysis, the statistical dispersion of antioxidant activity in the analyzed set of heterocyclic compounds synthesized by us is large indeed as supported by ANOVA ($p < 0.05$). This was measured by the inverse relationship between IC_{50} and percent inhibition to be strong in both DPPH ($r = -0.89$) and in ABTS ($r = -0.86$) assays hence indicating that lower IC_{50} values are related positively to the free radical scavenging activity. Of all the compounds tested, HC-8 was the most positive in all the assays by virtue of the fact that it contained an aromatic hydroxylated group that increased donation of electrons and stability of free radicals. This well defined structure-activity relation (SAR) supports the idea of electron-donating groups increasing antioxidant potential and single out HC-8 as a potential drug target in the next level of pharmacological investigation.

4. Discussion

This section interprets the key findings of the study, highlighting the superior antioxidant activity of HC-8 and its structural advantages. It contrasts the current work with previous heterocyclic synthesis methods, underscoring the novelty of integrating microwave-assisted synthesis with biological assays and SAR analysis. The discussion also outlines the practical implications, acknowledges limitations, and proposes directions for future research to enhance pharmacological relevance and compound optimization.

4.1. Interpretation of Results

This research was able to synthesize ten heterocyclic compounds (HC-1 to HC-10) via the microwave-assisted organic synthesis (MAOS) and screen their antioxidant activity through DPPH, ABTS and FRAP assays. HC-8 proved to be the strongest antioxidant out of all the synthesized scaffolds in all the assays with the highest percent inhibition and the lowest IC_{50} values. Such high activity was explained by the presence of a hydroxylated aromatic ring, which reinforces generation of electron donation, fixes free radicals, and maximizes redox performance. Other promising compounds were HC-4 and HC-2, which have a great capacity to act as a scavenger. Unlike HC-1, HC-6, and HC-3 showed a comparatively diminished action, thus it was proven that structural modifications play an influential role in the activity of the antioxidant.

4.2. Comparison with Existing Studies

Comparative review of recent patterns in the synthesis, studying, and the most significant results of heterocyclic synthesis method with references to the objectives, modalities, important findings, and the main attributes of the current microwave-assisted technique of antioxidant assessment.

Table 5: Comparative Analysis of Recent Studies on Heterocyclic Synthesis Approaches

Author(s) & Year	Objective	Method Used	Key Findings	Superiority of Present Study
Porta et al., 2016 [11]	To review advancements in flow chemistry for pharmaceutical synthesis	Literature survey on flow chemistry applications	Highlighted advantages in scalability, safety, and continuous processing	Your study employs microwave-assisted synthesis offering faster reaction times, eco-friendliness, and higher yields compared to conventional and

				flow-based approaches
Rostovtsev et al., 2002 [12]	To develop copper(I)-catalyzed azide-alkyne cycloaddition (CuAAC) reaction	Experimental cycloaddition protocol under mild conditions	Introduced "click chemistry" as a robust method for triazole formation	Your study contributes a green, rapid synthesis route for heterocycles, aligning with click principles while extending to antioxidant drug design
Sturm et al., 2014 [13]	To intensify chemical processes using microwave irradiation	Application of microwaves in organometallic synthesis	Demonstrated faster kinetics and improved selectivity	Your work extends microwave methodology to bioactive heterocycles, integrates biological screening (antioxidant assays), and includes SAR correlation
Tagliapietra et al., 2019 [14]	To explore solvent-free synthesis using microwave/ultrasound	Combination of green techniques in heterocyclic compound preparation	Achieved eco-efficient synthesis with minimal waste	Your study applies similar eco-friendly principles, but combines them with biological activity profiling

				and statistical validation
Tornøe et al., 2002 [15]	To synthesize peptidotriazoles via Cu-catalyzed dipolar cycloaddition	Solid-phase organic synthesis using Cu(I) catalysis	Developed regiospecific triazole-forming reactions	Your study builds on heterocyclic versatility, integrating medicinal chemistry goals through antioxidant evaluation and in vitro correlation analysis

This comparative table, as well as exemplifies the advancements in the heterocyclic synthesis by the means of methods such as flow chemistry, click reactions, and green approaches within recent studies. However, unlike previous research, which targeted synthesis rate, or was based on scaling or structure advances, the current study is unique in that microwave-based green chemistry is coupled with biological assays (DPPH, ABTS, and FRAP tests), as well as structural activity relationship (SAR) analysis. This study, in contrast to those of others in the past, which focused on synthetic procedures alone, connects chemical synthesis with functional antioxidant activity recognizing that HC-8 holds potential as lead compounds. Such a comprehensive vision can increase the therapeutic value and novelty of developing heterocyclic drugs.

4.3. Implications of Findings

The findings of this study emphasize the usefulness of microwave-aided reaction to synthesize biologically active molecules efficiently that are of great antioxidant value. The strong structure activity relationship (SAR) achieved by statistical correlation analysis emphasizes the significant E/D and the Elevation in free radical scavenging property possible by only slightly altering the structure especially the addition of electron donating groups. The fact that the molecule HC-8 qualifies as a high-risk hit leads to a potent basis on the following facets, that is to continue to optimize HC-8; or even to develop that as a pharmacological antioxidant agent.

4.4. Limitations of the Study

While the research offers an extensive in vitro study of antioxidant performance, there is no in vivo study or pharmacokinetic profile of the same. Also, the small sample size (only ten compounds) does not allow exploring the SAR trends to the full extent. The paper utilizes again

exclusive to the chemical assays (DPPH, ABTS, FRAP) that, despite being informative in a certain way, fail to recreate the physiological oxidative conditions fully.

4.5. Suggestions for Future Research

Additional studies to prove the therapeutic value of HC-8 and similar scaffolds include in vivo assessments of antioxidance services and toxicology. Computation modeling and docking may also be used to develop further ways of interaction between inter-molecular reactions with reactive oxygen species. Increasing the library of compounds with changes in heterocyclic cores and diverse substituents might provide a better understanding of SAR and might have the possibility to find more potent analogs. Lastly, inductive research on the actions of the compounds in disease models of relevance to oxidative stress (e.g., neurodegeneration, cardiovascular diseases) would provide meaningful translational information.

5. Conclusion

This final section encapsulates the major achievements of the study, emphasizing the synthesis and antioxidant evaluation of heterocyclic compounds, particularly the promising potential of HC-8. It highlights the significance of integrating microwave-assisted synthesis with biological assays, underscores the role of structure-activity relationships, and proposes future research directions to advance these findings toward therapeutic applications.

5.1. Summary of Key Findings

The current work was able to synthesize ten structurally varied heterocyclic compounds (HC-1 to HC-10) via microwave-assisted organic synthesis (MAOS) and ascertain their antioxidant capacity by conventional in vitro assays-DPPH, ABTS and FRAP. All synthesized compounds showed the quantifiable antioxidant action, and HC-8 conformed to the highest free radical scavenging and ferric reducing power in all tests. Correlation studies and ANOVA showed a significant disparity between the compounds and determined an impressive inverse relationship between IC 50 values and anti-oxidant activity. Presence of electron-donating groups (e.g., -OH) was determined as a crucial structural aspect that takes part in the boosted bioactivity. The most interesting lead of the compound, HC-8, possessed a hydroxylated aromatic ring as a lead candidate to be developed with regard to its pharmacology.

5.2. Significance of the Study

The study promotes the value of green, fast, and efficient microwave-aided synthesis methods of producing bioactive heterocyclic scaffolds. In contrast to other reports that described either synthetic approach or biological analysis in isolation, a combination is provided herein that provides a comprehensive view of structure activity relationship (SAR). Coupling the chemical synthesis with functional antioxidant screening in the study does not only present the field of heterocyclic drug discovery with a new dimension, it also offers a comparative platform to the current bodies of knowledge on flow chemistry, click chemistry and greener experimental

practices. The results can offer a good understanding to the approach to design antioxidant molecules with therapeutic interest that can justify the use of MAOS in medicinal chemistry.

5.3. Final Thoughts or Recommendations

The fact that HC-8 is a potent antioxidant makes way to in vivo experiments, its toxicity assessment and pharmacokinetic profiling. The implication of the current study is that in the future, the heterocyclic compound library has to be expanded to perfect SAR trends and confirm the viability of identified strong scaffolds under physiological conditions. The molecular nature of antioxidant interactions might also be explained with the help of computational docking and modeling studies. Moreover, the investigation of the synthesized derivatives in the models of oxidative stress related diseases (e.g. neurodegenerative and cardiovascular conditions) will assist in defining their practical curative potential. On the whole, this research can serve as a solid basis of the further development of novel agents of antioxidants with the respect to sustainable synthetic approaches.

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