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ORIGINAL ARTICLE

Synthesis And Anti-Inflammatory Activity of Benzimidazole Coumarins

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ABSTRACT

Objective: Benzimidazoles and coumarins are heterocyclic compounds known for their diverse biological and pharmacological properties, such as antibacterial, antifungal, anti-inflammatory, and cytotoxic effects. Derivatives of benzimidazole coumarins have demonstrated promise as antiviral, antineoplastic, and antifilarial agents. This research focuses on the synthesis of benzimidazole coumarin derivatives and evaluates their potential for anti-inflammatory activity.

Methodology: Coumarin-3-carboxylic acid was synthesized via the Perkin rearrangement of salicylaldehyde and diethylmalonate. This intermediate was then reacted with 4-chloro ortho-phenylenediamine, triethylamine, and ethyl chloroformate to form an amide linkage, resulting in a yellow solid. Subsequent cyclization with polyphosphoric acid produced the benzimidazole-3-coumarin nucleus as a dark yellow solid. Finally, Ullmann coupling with various aryl amines, using L-proline and copper iodide as catalysts, yielded the desired derivatives. These compounds were characterized by melting point analysis, TLC, IR spectroscopy, and NMR spectroscopy. The synthesized derivatives were also evaluated for anti-inflammatory activity using the carrageenan-induced paw edema model in rats.

Findings: Compound P-1 exhibited significant dose-dependent activity ($p<0.001$) at both 100 mg/kg and 200 mg/kg doses, while compound P-2 demonstrated moderate activity. The IR spectra revealed absorption peaks at 3379.89 cm^{-1} (indicative of N-H groups) and 1335.24 cm^{-1} (C-N stretching), confirming the presence of the benzimidazole nucleus. The ^1H NMR spectrum further supported these findings with chemical shifts consistent with the benzimidazole structure. Absorption peaks at 1685.45 cm^{-1} and 1089.81 cm^{-1} confirmed the presence of the pyrone nucleus, while the multiple peaks between δ 6.7 and 8.5 ppm indicated the formation of benzopyrone.

Novelty: Benzimidazole-coumarin hybrids hold promise as new anti-inflammatory agents and warrant further investigation for potential clinical applications.

Keywords: Benzimidazole; Coumarin; AntiInflammatory activity; Antibacterial; Anti Filarial agents

INTRODUCTION

Benzimidazole has been widely studied against both communicable and non-communicable diseases, and many benzimidazole based drugs are in clinical use as antiulcer (omeprazole), anthelmintic (flubendazole), antihistaminic (astmizole) and antihypertensive (telmisartan) agents[1,2]. Coumarins form an elite class of compounds, which exhibit a variety of therapeutic activities including antioxidant, anti-inflammatory, antitumor, antiviral, antituberculosis and antimicrobial[3–6]. Anti-inflammatory activity of coumarin derived compounds has been reviewed extensively and a structure activity relationship (SAR) has been established wherein it is found that an aromatic group when directly fused or linked through amide linkage at the 3-position of coumarin nucleus incurs anti-inflammatory activity[7]. Many such derivatives also possess antioxidant activity through scavenging mechanisms[8,9]. Benzimidazole and coumarin derivatives are recognized for their diverse biological activities, including anti-bacterial, anti-protozoal, anti-microbial, anti-fungal, anti-inflammatory, anti-allergic, anthelmintic, anti-parasitic, cytotoxicity, and DNA topoisomerase-I inhibition. In the present study, benzimidazole derivatives were synthesized and the biological potency of benzimidazole derivatives was evaluated that mimic the biologically active coumarin nucleus, known for pharmacological properties.

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METHODOLOGY

Materials required

Salicylaldehyde, diethyl malonate, piperidine, ethyl chloroformate, triethylamine, 4-chloro orthophenylene diamine, polyphosphoric acid, Copper iodide (CuI), L-Proline and substituted anilines and other solvents were procured from Spectro Chem. The IR spectrum was recorded in Shimadzu FTIR 8400S spectrometer. The NMR spectra of the compounds were carried out in Bruker spectrospin-400 NMR spectrophotometer in Deuterated chloroform (CDCl3).

Synthesis of aryl amine derivatives of benzimidazole coumarins

The aryl amine derivatives of benzimidazole coumarins were synthesized in 4 steps.

Step 1

Synthesis of Coumarin-3-Carboxylic Acid: A mixture of the substituted salicylaldehyde (3.6 mmol), diethyl malonate (1.1 ml, 7.2 mmol), ethanol (10 ml), piperidine (0.2 ml) and glacial acetic acid (3 drops) was refluxed for 3 hours at 75 °C in 250 ml round bottomed flask. After cooling, the ethanol was removed by evaporation at reduced pressure, then a solution of 10% potassium hydroxide (25 ml) was added and mixture was refluxed for one hour at 75 °C. After cooling, 40 ml of water was added and the solution was adjusted to pH 3.0 with 10% Hydrochloric acid (HCl). The precipitate was collected by filtration, the filtrate was recrystallised from methanol, and a white solid for coumarin-3- carboxylic acid was obtained [10].

Step 2

Synthesis of N-(2-Amino-4-Chlorophenyl)-2-Oxo-2H-Chromene-3- Carboxamide: To the ice-cold solution of coumarin-3-carboxylic acid (0.28 gm, 1mmol) in acetone (10 ml), ethyl chloroformate (0.15 ml, 1.2 mmol) and triethylamine (0.164 ml, 1.2 mmol) were added in 250 ml round bottomed flask. The orange suspension turned light yellow and white solid precipitated out in 10 minutes. The triethylamine hydrochloride was filtered off and to the filtrate 4 choro-orthophenylene diamine (0.11 gm, 1mmol) was added and the mixture was stirred at room temperature overnight. The yellow suspension was evaporated to dryness and yellow solid was obtained and was recrystallized from ethanol.

Synthesis of Coumarin-3-Benzimidazole

Polyphosphoric acid (5 gm) was added to the coumarin amide linkage compound, and the mixture was heated at 150-170 °C for 4 hours. After cooling, water was added (reddish-orange colour solution was formed) and then 5N sodium hydroxide (NaOH) solution was used to adjust the pH to 7.0. Yellow oil separated solidified overnight and showed some impure compound. This product was washed with ether and the pure compound obtained was a dark yellow solid [11].

Step 4

Coupling Reaction of Benzimidazole Coumarin and Substituted Aniline Catalysed by Cui and L-Proline: A mixture of benzimidazole-3-coumarin (5 mmol), substituted aniline (7.5 mmol), potassium carbonate (10 mmol), CuI (0.5 mmol) and L-Proline (1 mmol) in 3 mL of DMSO was taken in round bottomed flask and heated at 80 °C -90 °C temperature for 12 hours. The cooled mixture was partitioned with water and ethyl acetate. The organic layer was separated with the help of separating funnel, and the aqueous layer was extracted with ethyl acetate. The combined organic layer was washed with brine, dried over sodium sulphate and concentrated in lyophilizer and then a solid was obtained [12].

Identification and Characterization

The compounds synthesized were identified and characterized using several methods. Melting point determination was employed to assess the purity and identify the compounds. Thin layer chromatography (TLC) was utilized for the separation and analysis of the different components. Infrared spectroscopy (IR) provided insights into the functional groups present within the compounds. Nuclear magnetic resonance spectroscopy (NMR) was used to get detailed information about the molecular structure, while mass spectroscopy (MS) was used to determine the molecular weight and composition.

Melting point determination

The melting points of the synthesized pure compounds were carried out using Thiel's tube method [13].

Thin layer chromatography

Pre-coated silica plates were used and Hexane: Ethyl acetate in the ratio of 2:2 was used as a mobile phase. After the development of the chromatogram, the spots were detected by placing the plate in UV chamber.

Infrared spectroscopy

IR spectroscopy is one of the most important tools for determining the various functional groups and the possible chemical structure. The important advantage of IR over the other technique is that it gives fingerprints (1300-650cm-1) information about the structure (functional group, bonding with each other) of molecules easily. No two compounds have identical fingerprint region. This technique is based upon the molecular vibration of the compound such that each and every bond will vibrate at different frequencies and this vibration frequency corresponds to the IR frequency.

Thus, IR spectra of each and every bond will be formed [14,15]. The IR spectrum was recorded in Shimadzu FTIR 8400S Spectrometer.

Nuclear magnetic resonance spectroscopy

The NMR spectra of the compounds were carried out in Bruker spectrospin-400 NMR spectrophotometer and chloroform was used as solvent. NMR allows comprehensive analysis for precise structural characterization of benzimidazole-coumarin derivatives, confirming their molecular frameworks and interactions. The interaction between matter and electromagnetic forces can be observed by subjecting a substance simultaneously to two magnetic forces, one stationary and the other varying at some radio frequency. At a particular combination of fields, energy is absorbed by the sample and absorption can be observed as a change in signal developed by a radio frequency detector and amplifier. This energy of absorption can be related to the magnetic dipolar nature of the spinning nuclei [16,17].

Mass spectroscopy

The technique involved the bombardment of electrons and converted to highly energetic positively charged ions (molecular ions), which can break up into smaller ions (fragment ions) and sorting them in gas phase, into a spectrum according to their mass/charge ratio [18]. The Mass spectra of the compounds were carried out in Electron spray mass spectrometer.

RESULTS

Physicochemical profile

Compound P-1

The compound was found to have the molecular formula $C_{22}H_1N_3O_2$, with a molecular weight of 355.38 g/mol. Its chemical name was 3-[5-(phenylamino)-2,3-dihydro-1H-benzimidazol-2-yl]-2H-chromene-2-one. The molecular composition consisted of carbon (74.78%), hydrogen (4.28%), nitrogen (11.89%), and oxygen (9.06%). Its melting point was 256-257 °C, and it appeared as a brown solid. The compound was soluble in chloroform and had an Rf value of 0.65. The percentage yield was recorded at 56%. The IR spectra (KBr, λ cm^{-1}) showed characteristic peaks for C=C at 1621.82 cm^{-1}, C-C at 1155.38 cm^{-1}, C-O-C at 1090.75 cm^{-1}, C=O at 1732.85 cm^{-1}, C-N at 1307.78 cm^{-1}, N-H at 3466.84 cm^{-1}, and aromatic C-H at 3143.24 cm^{-1}. The ^{1}H NMR spectrum ($CDCl_3$ δ ppm) included signals at δ 4.01 (s, 1 H, NH), δ 7.82 (s, 1H, Ar-H), δ 8.77 (s, 1H, Ar-H), δ 8.05 (d, 2H, Ar-H), and δ 7.05-7.44 (m, 10H, Ar-H). The mass spectral data displayed significant peaks at m/z M+ 355 (2%), 341 (3%), 211 (10%), 106 (90%), and 83 (85%).

Compound P-2

The compound was characterized by a molecular formula of $C_{22}H_1ClN_3O_2$ and a molecular weight of 389.83 g/mol. Its chemical name was 3-{5-[(2-chlorophenyl)amino]-2,3-dihydro-1H-benzimidazol-2-yl}-2H-chromene-2-one. The molecular composition consisted of carbon (67.78%), hydrogen (4.14%), nitrogen (10.78%), oxygen (8.21%), and chlorine (9.09%). It had a melting point of 260-261 °C and presented as a brown solid. The compound was soluble in chloroform and had an Rf value of 0.58, with a percentage yield of 54%. The IR spectra (KBr, λ cm^{-1}) revealed characteristic absorption bands at 1600.33 cm^{-1} for C=C, 1148.92 cm^{-1} for C-C, 1089.81 cm^{-1} for C-O-C, 1685.45 cm^{-1} for C=O, 1335.24 cm^{-1} for C-N, 3379.89 cm^{-1} for N-H, 3059.43 cm^{-1} for aromatic C-H, and 825.50 cm^{-1} for C-Cl. The ^{1}H NMR spectrum ($CDCl_3$, δ ppm) included signals at δ 4.3 (s, 1H, NH), δ 8.93 (s, 1H, Ar NH), δ 7.8 (s, 1H, Ar H), δ 8.53 (s, 1H, Ar H), and δ 6.75-7.69 (m, 10H, Ar H).

Compound P-3

The compound was defined by its molecular formula $C_{22}H_1FN_3O_2$, with a molecular weight of 373.37 g/mol. Its chemical name was 3-{5-[(4-fluorophenyl)amino]-2,3-dihydro-1H-benzimidazol-2-yl}-2H-chromene-2-one. The molecular composition included carbon (70.77%), hydrogen (4.32%), nitrogen (11.25%), oxygen (8.75%), and fluorine (5.09%). The compound had a melting point of 346-347°C and appeared as a reddish-brown solid. It was soluble in chloroform and had an Rf value of 0.41, with a percentage yield of 62%. The IR spectra (KBr, λ cm^{-1}) revealed characteristic absorption bands at 1611.41 cm^{-1} for C=C, 1166.85 cm^{-1} for C-C, 1135.99 cm^{-1} for C-O-C, 1720 cm^{-1} for C=O, 1293.18 cm^{-1} for C-N, 3467.77 cm^{-1} for N-H, 3007.78 cm^{-1} for aromatic C-H, and 1311.50 cm^{-1} for C-F. The ^{1}H NMR spectrum ($CDCl_3$, δ ppm) included signals at δ 8.69 (s, 1H, Ar H), δ 8.09 (d, 1H, Ar H), δ 7.83 (s, 1H, Ar H), δ 7.03-7.62 (m, 9H, Ar H), δ 8.51 (s, 1H, Ar H), and δ 4.01 (s, 1H, NH). The mass spectral data showed significant peaks at m/z M+ 373 (2%), 341 (4%), 211 (10%), and 106 (90%).

Compound P-4

The compound was characterized by its molecular formula $C_2H_1ClN_3O_3$, with a molecular weight of 419.86 g/mol. Its chemical name was 3-{5-[(4-chlorophenyl)amino]-2,3-dihydro-1H-benzimidazol-2-yl}-6-methoxy-2H-chromene-2-one. The molecular composition consisted of carbon (65.79%), hydrogen (4.32%), nitrogen (10.01%), oxygen (11.43%), and chlorine (8.44%). The compound had a melting point of 283-284°C and was a dark brown solid. It was soluble in chloroform and had an Rf value of 0.42, with a percentage yield of 56%. The IR spectra (KBr, λ cm^{-1}) showed characteristic absorption bands at 1609.59 cm^{-1} for C=C, 1193.86 cm^{-1} for C-C, 1102.01 cm^{-1} for C-O-C, 1740 cm^{-1} for C=O, 1275.93 cm^{-1} for C-N, 3374.37 cm^{-1}

for N-H, 3143.24 cm^{-1} for aromatic C-H, 2939.34 cm^{-1} for aliphatic C-H, 733.35 cm^{-1} for C-Cl, and 3060.07 cm^{-1} for aromatic C-H. The ^1H NMR spectrum ($CDCl_3$, δ ppm) included signals at δ 7.584-7.052 (m, 9H, Ar H), δ 7.821 (s, 1H, Ar H), δ 8.022 (d, 1H, Ar H), δ 8.773 (s, 1H, Ar H), δ 4.006 (s, 3H, OCH_3), and δ 3.104 (s, 1H, NH). The mass spectral data displayed significant peaks at m/z M+ 419 (4%), 389 (2%), 355 (5%), 341 (9%), 211 (20%), 106 (110%), and 83 (85%).

Compound P-5

The compound was defined by its molecular formula $C_{24}H_{19}N_3O_3$, with a molecular weight of 399.44 g/mol. It was chemically named 3-{5-[(4-methylphenyl)amino]-2,3-dihydro-1H-benzimidazol-2-yl}-6-methoxy-2H-chromene-2-one. The molecular composition included carbon (72.16%), hydrogen (5.30%), nitrogen (10.52%), and oxygen (12.02%). The compound had a melting point of 270-273°C and appeared as a pale brown solid. It was soluble in chloroform and had an Rf value of 0.65, with a percentage yield of 63%. The IR spectra (KBr, λ cm^{-1}) revealed characteristic absorption bands at 1620.44 cm^{-1} for C=C, 1199.21 cm^{-1} for C-C, 1095.83 cm^{-1} for C-O-C, 1713 cm^{-1} for C=O, 1274.35 cm^{-1} for C-N, 3361.37 cm^{-1} for N-H, 3026.32 cm^{-1} for aromatic C-H, and 2921.58 cm^{-1} for aliphatic C-H. The ^1H NMR spectrum ($CDCl_3$, δ ppm) included signals at δ 8.63 (s, 1H, NH), δ 8.77 (s, 1H, Ar H), δ 6.59-7.40 (m, 10H, Ar H), δ 3.93 (s, 3H, OCH_3), δ 3.24 (s, 1H, NH), and δ 2.24 (s, 3H, CH_3).

Compound P-6

The compound was identified by its molecular formula $C_{23}H_1FN_3O_3$, with a molecular weight of 403.40 g/mol. It was chemically named 3-{5-[(4-fluorophenyl)amino]-2,3-dihydro-1H-benzimidazol-2-yl}-6-methoxy-2H-chromene-2-one. The molecular composition included carbon (68.48%), hydrogen (4.50%), nitrogen (10.42%), oxygen (11.90%), and fluorine (4.71%). The compound had a melting point of 337-338°C and appeared as a brown solid. It was soluble in chloroform and had an Rf value of 0.49, with a percentage yield of 63%. The IR spectra (KBr, λ cm^{-1}) exhibited characteristic absorption bands at 1608.83 cm^{-1} for C=C, 1192.97 cm^{-1} for C-C, 1099.94 cm^{-1} for C-O-C, 1742.91 cm^{-1} for C=O, 1277.79 cm^{-1} for C-N, 3455.29 cm^{-1} for N-H, 3090.30 cm^{-1} for aromatic C-H, 2936.47 cm^{-1} for aliphatic C-H, and 1367.47 cm^{-1} for C-F. The ^1H NMR spectrum ($CDCl_3$, δ ppm) included signals at δ 4.0 (s, 3H, OCH_3), δ 3.2 (s, 1H, NH), δ 7.8 (s, 1H, Ar H), δ 8.7 (s, 1H, Ar NH), δ 8.1 (d, 1H, Ar H), and δ 7.0-7.7 (m, 9H, Ar H).

The results obtained for the synthesized benzimidazole coumarins were summarized in Tables 1 and 2.

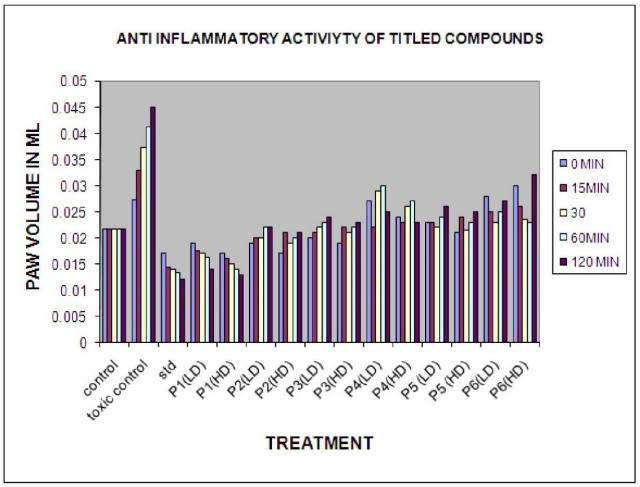

Fig. 1: Anti-inflammatory activity for synthesized benzimidazole coumarins in carrageenan induced paw edema

Animal study

DISCUSSION

Different derivatives of benzimidazole coumarin were obtained in four steps. In the first step, salicylaldehyde and diethylmalonate underwent a Perkin rearrangement reaction to yield coumarin-3-carboxylic acid. This compound was then treated with 4-chloro-1,2-phenylenediamine (4-chloro OPD) to form an amide linkage in the presence of triethylamine (TEA) and ethyl chloroformate. The resulting yellow solid was subjected to cyclization in the presence of polyphosphoric acid (PPA), producing a dark yellow solid of benzimidazole coumarin. This benzimidazole coumarin was further subjected to an Ullmann coupling reaction with aryl amines in the presence of L-proline and copper iodide catalyst.

The structures of the synthesized compounds were established from spectral data. The IR spectra exhibited an absorption at 3379.89 cm^{-1}, indicating the presence of N-H, supported by the C-N stretching at 1335.24 cm^{-1}, which confirmed the benzimidazole nucleus. This was further supported by the chemical shift seen in the ^1H NMR spectrum at δ 4.3. Absorptions at 1685.45 cm^{-1} and 1089.81 cm^{-1} confirmed the presence of the pyrone nucleus. The formation of the benzopyrone structure was established from the multiple peaks observed at δ 6.7 to 8.5 ppm.

All synthesized derivatives were screened for anti-inflammatory activity using the carrageenan-induced paw edema method in rats. Significant dose-dependent activity ($p<0.001$) was observed with compound P-1 at doses of 100 mg/kg and 200 mg/kg, while moderate activity was seen with compound P-2. The first phase of inflammation was attributed to the release of histamine and serotonin, and the second phase was due to the release of bradykinin, proteases, prostaglandins, and lysosomes.

Table 1: Physicochemical properties of the synthesized benzimidazole coumarins

Compound Code	Molecular formula	Mol weight	Melting point (°C)	Yield (%)	Rf Value	Anti-inflammatory activity
P 1	$C_{22}H_{17}N_30_2$	355.38	256-570C	56%	0.73	Significant Active
P 2	$C_{22}H_{16}ClN_3O_2$	389.83	260-2610C	54%	0.58	In active
P 3	$C_{22}H_{16}FN_3O_2$	373.37	346-3470C	62%	0.41	In active
P 4	$C_{28}H_{18}ClN_3O_3$	419.86	283-2840C	56%	0.42	In active
P 5	$C_{24}H_{19}N_3O_3$	399.44	270-2730C	63%	0.65	In active
P 6	$C_{23}H_{18}FN_3O_3$	403.40	337-3380C	63%	0.49	In active

Table 2: Spectral data of synthesized benzimidazole coumarins

Compound code	Mol formula	IR-KBr cm^{-1} spectral data	^{1}HNMR- $CDCl_3$ δ (ppm) spectral data
P 1	$C_{22}H_{17}N_30_2$	C=C(1621.82cm^1), C-C (1155.38cm^{-1}), C-O-C (1090.75cm^{-1}), C=O (1732.85cm^{-1}), C-N (1307.78cm^{-1}), N-H (3466.84cm^{-1}), C-Har (3143.24 cm^{-1}).	δ (4.01, S,1H, NH), δ (7.82, S,1H, ArH), δ (8.77, S,1H,ArH), δ (8.05, d,2H, ArH), δ (7.05-7.44,m,10H,Ar H).
P 2	$C_{22}H_{16}ClN_3O_2$	C=C(1600.33cm^{-1}), C-C (1148.92cm^{-1}), C-O-C (1089.81cm^{-1}), C=O (1685.45cm^{-1}), C-N (1335.24cm^{-1}), N-H (3379.89cm^{-1}), C-Har (3059.43cm^{-1}), C-Cl (825.50 cm^{-1}).	δ (4.3, S,1H, NH), δ (8.93, S,1H, ArNH), δ (7.8, S, 1H, ArH), δ (8.53, S,1H, ArH), δ 6.75-(7.69,m,10H, Ar H).
P 3	$C_{22}H_{16}FN_3O_2$	C=C(1611.41cm^{-1}), C-C (1166.85cm^{-1}), C-O-C (1135.99 cm^{-1}) C=O (1720 cm^{-1}), C-N (1293.18 cm^{-1}), N-H (3467.77cm^{-1}), C-Har (3007.78cm^{-1}), C-F (1311.50 cm^{-1})	δ (8.69, S,1H, Ar H) δ (8.09, d,1H, ArH), δ (7.83, S,1H, ArH), δ (7.03-7.62, m,9H,ArH), δ (8.51, S, 1H, Ar H), δ (4.01, S,1H, NH)
P 4	$C_{28}H_{18}ClN_3O_3$	C=C(1609.59cm^{-1}), C-C (1193.86cm^{-1}), C-O-C (1102.01 cm^{-1}), C=O (1740 cm^{-1}), C-N (1275.93cm^{-1}), N-H (3374.37cm^{-1}), C-H Ar (3143.24cm^{-1}), C-HAlip (2939.34cm^{-1}), C-Cl (733.35cm^{-1}), C-HAr (3060.07 cm^{-1})	δ (7.584-7.052,m,9H,ArH), δ (7.821, S,1H, Ar H), δ (8.022, d,1H, Ar H), δ (8.773, S,1H, Ar H), δ (4.006, S,3H, OCH_3), δ (3.104, S,1H, NH).
P 5	$C_{24}H_{19}N_3O_3$	C=C(1620.44cm^{-1}), C-C (1199.21cm^{-1}), C-O-C (1095.83cm^{-1}), C=O (1713 cm^{-1}), C-N (1274.35 cm^{-1}), N-H (3361.37cm^{-1}), C-HAr (3026.32cm^{-1}), C-HAlip (2921.58 cm^{-1}).	δ (8.63, S,1H, NH), δ (8.77, S,1H, Ar H), δ (6.59-7.40, m,10H, ArH), δ (3.93, S,3H, OCH_3), δ (3.24, S,1H, NH), δ (2.24, S, 3H, CH_3).
P 6	$C_{23}H_{18}FN_3O_3$	C=C(1608.83cm^{-1}), C-C(1192.97cm^{-1}), C-O- C(1099.94cm^{-1}), C=O(1742.91cm^{-1}), C-N (1277.79cm^{-1}), NH (3455.29cm^{-1}), C-HAr(3090.30cm^{-1}), C-H Alip(2936.47cm^{-1}), C-F (1367.47 cm^{-1}).	δ (4.0, S,3H, OCH_3), δ (3.2, S1H, NH), δ (7.8, S1H, Ar H), δ (8.7, S,1H, Ar NH), δ (8.1, d,1H, Ar H), δ (7.0-7.7, m,9 H,Ar H).

Therefore, it could be concluded that the inhibitory effect of compound P-1 on carrageenan-induced paw edema was likely due to the inhibition of cyclooxygenase, leading to a reduction in prostaglandin synthesis[19]. The study revealed that the unsubstituted aniline derivative of benzimidazole coumarin favoured the anti-inflammatory response, whereas derivatives with halogen substitutions on the aniline ring did not produce an anti-inflammatory effect.

CONCLUSION

The new derivatives were successfully synthesized in four steps. The structures, confirmed by IR, NMR, and mass spectroscopy, showed that compound P1 exhibited a significant dose-dependent anti-inflammatory effect. The study concluded that unsubstituted aniline derivatives of benzimidazole coumarin were more effective, while halogen-substituted derivatives were not.

Table 3: Anti-inflammatory activity for synthesized benzimidazole coumarins in carrageenan induced paw edema

Treatment	Paw volume in ml at different time intervals				
	0 Min	15 Min	30 Min	60 Min	120 Min
Control	0.0216±0.001 8	0.0216±0.001 6	0.0216±0.0016	0.0216±0.0016	0.0216±0.0016
Toxic control	0.0273±0.001 4**	0.033±0.0020* *	0.0373±0.003**	0.0413±0.002** *	0.045±0 .002***
Standard	0.017±0.0035 *aaa	0.0143±0.006[a]aa	0.014±0.0040* aaa	0.0133±0.0017 *aaa	0.012±0.0011**aaa
P1-100 mg/kg	0.019±0.000 5aaa	0.0176±0.00 8aaa	0.017±0.005[aaa]	0.01633±0.00 08aaa	0.014±0.0008* aaa
P1-200 mg/kg	0.017±0.0011* aaa	0.016±0.0005[a] aa	0.015±0.0008[aa] a	0.014±0.0005* aaa	0.013±0.0003* aaa
P2-100 mg/kg	0.019±0.0005[a] aa	0.020±0.0005[a]	0.020±0.0005[a]	0.022±0.0005[a]	0.022±0.0005[aa]
P2-200 mg/kg	0.017±0.0011* aaa	0.021±0.0011[a]	0.019±0.0011[aa]	0.020±0.0011[aa]	0.021±0.0011[aa]
P3-100 mg/kg	0.020±0.000 5aaa	0.021±0.005[aa]	0.022±0.0005[aa]	0.023±0.0005[a]	0.024±0.0005[aa]
P3-200 mg/kg	0.019±0.0011[a] aa	0.022±0.0011[a] a	0.021±0.0011[aa] a	0.022±0.0011[aa]	0.023±0.0011[aa]
P4-100 mg/kg	0.027±0.0005	0.022±0.005[aa]	0.029±0.0005* * a	0.030±0.0005	0.025±0.0005* aaa
P4-200 mg/kg	0.024±0.0011	0.023±0.0011[a] a	0.026±0.0011* aa	0.027±0.0011	0.023±0.0011[aa] a
P5-100 mg/kg	0.021±0.0011[a] aa	0.024±0.0011[a] a	0.0215±0.0011[a] aa	0.023±0.0011[aa] a	0.025±0.0011[aa]
P5-200 mg/kg	0.033±0.0001 2aaa	0.034±0.0001 2aa	0.035±0.00012[a] aa	0.036±0.00012[a] aa	0.037±0.00012 aaa
P6-100 mg/kg	0.028±0.0005	0.025±0.0035[a]	0.023±0.0005[aa]	0.025±0.0005[a] aa	0.027±0.0005[aa]
P6-200 mg/kg	0.030±0.0011	0.026±0.0011[a]	0.0235±0.0011[a] a	0.023±0.0011[aa] a	0.0011±0.026[aa]

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