Cyclopentylamine contains nitrogen that bears an unshared pair of electrons. The tendency of nitrogen to share this pair of electrons underlies the entire chemical behavior of amines. Cyclopentylamine reacts with phthalimide derivatives to yield a substituted amide and the amide formed in this case is a carboxamide, which have been known to possess some interesting biological activities. Non-steroidal anti-inflammatory drugs (NSAIDs) belong to a variety of chemical classes with no common features except the absence of a steroidal structure. Their primary effect is pain relief but also have antipyretic and anti-inflammatory activities. Their prostaglandic effects are explained by sensitization of nociceptive nerve endings to the stimulating effect (algogenic) of kinins (bradykinin), serotonin and histamine. In addition, production of prostanoid in the brain has a thermoregulatory effect (1). It has been found that phthalimides and their derivatives has their usefulness as inhibitors of tumor necrosis factor-alpha production (2, 3). A good number of new carboxamides and their derivatives have been synthesized and screened for various pharmacological properties including anti-inflammatory and analgesic activities.

New antiinflammatory agents, 4-hydroxy-2-(1H)-oxo-1-phenyl-1-8-naphthyridine-3-carboxamides, were synthesized and found to have a pronounced effect on antiinflammatory activity (4). Synthesis of new aryl (alkyl) carboxamides derived from 2-amino-4,6-dimethylpyridine and benzoxazolinoine pharmacophore and designed as antiinflammatory and analgesic agents was accomplished (5). Novel anti-inflammatory agents of 5-oxo-5H-1,3,4-thiadiazole[3,2-a]pyrimidine-6-carboxamides were synthesized and screened to have a pronounced anti-inflammatory effect (6). Our recent studies have shown that some phthalimides could be opened with sodium borohydride, isopropylamine and benzylamine to obtain novel carboxamides with interesting anti-inflammatory and analgesic activities (7-9). The biological relevance of carboxamides and their derivatives encouraged us to investigate the products of the nucleophilic reaction of cyclopentylamine with phthalimide derivatives. The products obtained were evaluated for antinociceptive activity using the mouse writhing assay.

MATERIAL AND METHODS

4-Phthalimidobutyric acid and N-prop-2-ynylphthalimide 1a and 4-phthalimidobutyric acid 1b was carried out in dimethylformamide at room temperature to afford benzamido-N-prop-2-ynyl-N-cyclopentyl-carboxamide (3) and benzamido-N,N-bis (cyclopentyl)-carboxamide (4), respectively. IR, NMR, and microanalyses were used to unequivocally characterize the compounds obtained. The new compounds were evaluated pharmacologically for their in vivo antinociceptive activity using acetic acid-induced writhing assay and compound 3 had significant activity (70%) comparable to indomethacin (71%) but better than acetylsaliclyc acid (59%), however, compound 4 had lower activity (29%). The effects were dose-dependent.

Keywords: cyclopentylamine, phthalimide derivatives, mice, antinociception, mouse writhing test

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were recorded on a Buck Scientific IR M500 instrument. NMR spectra were recorded on a Varian Gemini 200 apparatus. Chemical shifts are reported in ppm relative to tetramethylsilane. Microanalyses were performed on a Perkin-Elmer CHN 240 apparatus. Analyses indicated by the symbols of the elements agreed favorably with the calculated values.

Anal. (C16 H18N2 O2): C, H, N.

CHEMISTRY

Benzamido-N-prop-2-ynyl-N-cyclopentyl-carboxamide (3)

To a stirred solution of N-prop-2-ynylphthalimide (1.00 g, 5.40 mmol) in 10 mL of dimethylformamide was added dropwise cyclopentylamine (0.53 ml, 5.40 mmol) for 24 h at room temperature. The reaction mixture was poured into 5 mL of 2M HCl and 30 mL of cold water and stirred for 5 min and then extracted with ethyl acetate. The organic phase was combined, washed with brine and dried over Na2SO4. The solvent was evaporated under reduced pressure and the residue was purified by recrystallization from methanol to give benzamido-N-prop-2-ynyl-N-cyclopentyl-carboxamide. (1.2 g, 82%), mp: 155-157°C, IR (KBr): 3250 (NH), 2950 (C-H), 1656 (C=O), 1560 (C=C), 1350, 720 (1,2-disubstitution) cm⁻¹; 'H-NMR (250 MHz, DMSO-d6) δ (ppm): 1.49 (m, 2H, -CH2-cyclopentyl), 1.61-1.65 (m, 2H, -CH2-C-cyclopentyl), 1.81-1.83 (m, 4H, -CH2-cyclopentyl), 3.11 (t, 1H, J = 2.90 Hz, =C-H), 3.97-3.99 (d, 2H, J = 2.98 Hz, -CH2-), 4.10-4.13 (quint, 1H, J = 5.73 Hz, -CH-cyclopentyl), 7.44-7.52 (m, 4H, Ar-H), 8.12-8.15 (d, 1H, J = 7.04 Hz, -NH), 8.63-8.68 (t, 1H, J = 5.18 Hz, -NH).

13C-NMR (63 MHz, DMSO-d6) δ (ppm): 23.51 (C-), 28.36 (C=C), 32.02 (-CH3), 50.72 (-CH2), 72.97 (-CH2), 80.86 (-CH3), 127.52, 127.73, 129.04, 129.46, 135.10, 136.61 (Ar-C), 167.56 (C=O), 167.68 (C=O).
Nucleophilic reaction of cyclopentylamine on phthalimide...

To a solution of 4-phthalimidobutyric acid (1 g, 4.29 mmol) in 15 mL of dimethylformamide was added dropwise cyclopentylamine (1.27 mL, 12.86 mmol) for 50 h, at room temperature. The reaction mixture was poured into 5 mL of 2M HCl and 30 mL of cold water and stirred for 5 min and then extracted with dichloromethane. The organic phase was combined, washed with brine and dried over Na₂SO₄. The solvent was evaporated under reduced pressure, and the residue was purified by recrystallization from methanol and further purified by column chromatography (CH₂Cl₂ : EtOAc 5:1, v/v) to afford white needlelike crystals of benzamido-N,N-bis(cyclopentyl)-carboxamide (4).

(0.54 g, 42%) m. p: 255-256 °C. IR (KBr): 3300 (NH), 2990 (C-H), 1630 (C=O), 1611 (C=C) cm⁻¹; ¹H-NMR (250 MHz, DMSO-d₆) δ (ppm): 1.50-1.53 (m, 2H, -CH₂-cyclopentyl), 1.54-1.57 (m, 2H, -CH₂-cyclopentyl), 1.65-1.81 (m, 4H, -CH₂-cyclopentyl ), 4.08- 4.16 (quintet, 1H, J = 7.50 Hz, -CH-cyclopentyl), 7.42-7.49 (m, 2H, Ar-H), 8.12-8.14 (d, 1H, J = 5.00Hz, NH); ¹³C-NMR (63 MHz, DMSO-d₆) δ (ppm): 23.50 (-CH₂-), 32.06 (-CH₃-), 50.72 (-CH₂-), 127.68, 129.01, 136.02, (Ar-C), 167.67 (C=O). Anal. (C₁₈H₂₄N₂O₂): C, H, N.

Pharmacological evaluation

Swiss mice (20-25 g) of either sex kept at the Laboratory animal home of the Faculty of Pharmacy, University of Benin, Benin City, Nigeria were used. The animals were maintained under standard environmental conditions and had free access to standard diet and water (test compounds were administered orally by gavage). Screening of analgesic activity was performed after p.o. administration of the test compounds at different dose levels. 1h after drug administration 0.2 mL of 0.6% acetic acid solution was given to mice intraperitoneally. Stretching movements consisting of arching of the back, elongation of body and extension of hind limbs were counted for 20 min after acetic acid injection. Indomethacin (10 mg/kg) and acetylsalicylic acid (100 mg/kg) were administered orally as reference drugs, while 10% Tween 80 was used as negative control. The analgesic activity (12) was expressed in terms of % inhibition:

\[ \% \text{ Analgesic activity} = \left( \frac{n - n'}{n} \right) \times 100 \]

(where n = mean number of writhes of control group, n’ = mean number of writhes of test group).

RESULTS AND DISCUSSION

Chemistry

The synthesis of benzamido-N-prop-2-ynyl-N-cyclopentyl-carboxamide (3) and benzamido-N, N-bis(cyclopentyl)-carboxamide (4) were accomplished in dimethylformamide at room temperature by the reaction of cyclopentylamine (2) with N-prop-2-ynylphthalimide 1a and 4-phthalimidobutyric acid 1b as shown in Scheme 1. The reaction of 1a with equimolar cyclopentylamine afforded 3 (82% yield) but when excess of cyclopentylamine was used with 1b it gave 4 (42% yield) that was disubstituted. However, when the ratio of 4-phthalimidobutyric acid 1b : cyclopentylamine (2) was 1:1 or 1:2 no product was obtained. As can be seen in Scheme 1, our desired product was compound 5 but this was not obtained.

Table 1: Effect of the test compounds on acetic acid-induced writhing in mice.

<table>
<thead>
<tr>
<th>Compounds</th>
<th>Doses in mg/kg (p.o.)</th>
<th>Numbers of writhing (per 20 min)</th>
<th>% Inhibition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control 10% Tween 80</td>
<td>0.2ml</td>
<td>53.33 ± 3.21</td>
<td>-</td>
</tr>
<tr>
<td>Acetylsalicylic acid</td>
<td>100</td>
<td>21.75 ± 2.72</td>
<td>59.22</td>
</tr>
<tr>
<td>Indomethacin</td>
<td>20</td>
<td>15.60 ± 0.87</td>
<td>70.75</td>
</tr>
<tr>
<td></td>
<td>40</td>
<td>28.50 ± 3.50</td>
<td>46.56</td>
</tr>
<tr>
<td></td>
<td>20</td>
<td>16.25 ± 3.17</td>
<td>69.53</td>
</tr>
<tr>
<td></td>
<td>40</td>
<td>49.50 ± 5.75</td>
<td>7.18</td>
</tr>
<tr>
<td></td>
<td>20</td>
<td>38.00 ± 2.55</td>
<td>28.75</td>
</tr>
<tr>
<td></td>
<td>40</td>
<td></td>
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</tr>
</tbody>
</table>

Values are mean ± S.E.M. *p < 0.05, †p < 0.01, ‡p < 0.001, significantly different from control, paired t-test (n = 5), p.o. = peroral

Antinociceptive activity

Acetic acid induced writhing test (10, 11) was performed by an i.p. injection of 0.6% acetic acid solution in a volume of 0.2 mL/mouse. In each group five Swiss mice of both sexes (pregnant females excluded) were kept and given a dose of a test compound by gavage. Screening of analgesic activity was performed after p.o. administration of the test compounds at different dose levels. 1h after drug administration 0.2 mL of 0.6% acetic acid solution was given to mice intraperitoneally. Stretching movements consisting of arching of the back, elongation of body and extension of hind limbs were counted for 20 min after acetic acid injection. Indomethacin (10 mg/kg) and acetylsalicylic acid (100 mg/kg) were administered orally as reference drugs, while 10% Tween 80 was used as negative control. The analgesic activity (12) was expressed in terms of % inhibition:

\[ \% \text{ Analgesic activity} = \left( \frac{n - n'}{n} \right) \times 100 \]

(where n = mean number of writhes of control group, n’ = mean number of writhes of test group).
The infra-red (IR) data clearly reveal the presence of -NH between 3250-3300 cm⁻¹ for compounds 3 and 4, which was completely absent in the corresponding phthalimide 1a and 1b. The proton NMR clearly shows -NH at the downfield region; 3 as a triplet at about 8.63-8.68 ppm (coupling constant $J = 5.18$ Hz) and also as a doublet at about 8.12-8.15 ppm (coupling constant $J = 7.04$ Hz) and 4 as a doublet at 8.12-8.14 ppm (coupling constant $J = 5.00$ Hz) which confirmed the disubstitution in 4. IR, NMR and microanalyses were used to unequivocally characterize the compounds obtained.

**Pharmacology**

The *in vivo* analgesic activity was determined using mouse writhing assay, which is a test useful for evaluating mild analgesic NSAIDs, and the results obtained are summarized in Table 1. At a dose of 40 mg/kg, the most active compound was 3 with 70% inhibition (p < 0.001) comparable to indomethacin with 71% inhibition (p < 0.001), the activity was greater than acetylsalicylic acid with 59% inhibition. Compound 4 was the least active at the dose used, however, the activity was dose-dependent. This assay suggests that the analgesic effect of compounds 3 and 4 may be peripherally mediated (13).

**Statistical analysis**

All data were expressed as the mean ± SEM; the Student’s $t$-test was applied to determine the significance of the difference between the control group and mice treated with the test compounds.

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**REFERENCES**


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