1. Introduction

Ribose sugar is one of the most potent classes of carbohydrates due to versatile applications of its analogues (Adamo and Pergoli, 2008; Cappellacci et al., 2002; Shecterle et al., 2010). A great deal of D-ribose and D-ribofuranose analogues are used in the synthesis of nucleoside and nucleotide derivatives (Downey et al., 2015; Fujisaka et al., 2019; Komatsu and Araki, 2003; Rahman et al., 2009). Many of these nucleosides possess numerous biological activities and the sugar moiety within these compounds are required for exhibiting these effects (El-Gazzar et al., 2009). D-ribose itself (Fig. 1, structure a) has shown anti-inflammatory effect by reducing serum concentration of renal inflammatory biomarkers in mice (Ueki et al., 2013). It is also used in congestive heart failure as a supplementary therapy and has shown enhancement of diastolic performance in congestive heart failure cases (Omran et al., 2003; Wagner et al., 2009). Adding D-ribose in the treatment regimen of fibromyalgia syndrome caused improvement in clinical outcomes (Gebhart and Jorgenson, 2004; Teitelbaum, 2012). Ribofuranosides of some synthesized pyrimidine (Fig. 1, structure c) and phenothiazine analogues showed better antimicrobial activity than the analogues alone (Kumar et al., 2001). α-D-ribofuranose analogues and its nucleoside derivatives have many reported pharmacological properties including analgesic, anti-inflammatory, antimicrobial and cytotoxic effect (Galmarini et al., 2008; Petrelli et al., 2017; Rahman et al., 2020). Some triazole based D-ribofuranose (Ferreira et al., 2010) and recently isolated α-ribosyl-4-benzyl-4-(hydroxymethyl)-1, 2-ethen-1, 2-dioxolane-D-ribofuranose (1). The synthesized compounds were then subjected to analgesic, anti-inflammatory, antimicrobial and antioxidant assays. Compound 3 demonstrated 79.74% (P < 0.001) writhing inhibition and highest reaction time of 2.55 ± 0.13 min (P < 0.001) after 30 min of oral administration in peripheral and central analgesic assay, respectively, at 50 mg/kg dose. Compound 2 and 6 exhibited significant anti-inflammatory activity at 100 mg/kg dose with paw edema inhibition of 91.15% (P < 0.001) and 95.13% (P < 0.001), respectively, in 4th h. The synthesized analogues did not show notable antioxidant and antibacterial properties. Molecular docking study revealed higher binding affinity of −8.1 kcal/mol and −8.9 kcal/mol of compound 3 towards cyclooxygenase-1 and phospholipase A2, respectively, compared to −7.7 and −7.6 kcal/mol respectively for corresponding native ligands. Compound 2 demonstrated binding affinity of −9.1 kcal/mol towards interleukin-1 receptor-associated kinase-4 compared to −8.7 kcal/mol of the native ligand. The molecular properties related to drug likeness of compounds were found to be within acceptable range. Synthetic D-ribofuranose analogues demonstrated promising analgesic and anti-inflammatory activities and further development may lead to new potent analgesic and anti-inflammatory agents.
in vitro and in vivo studies as well as in silico studies. Therefore, we attempted to synthesize some new derivatives of α-D-ribofuranose in order to investigate their pharmacological properties. In addition, we also examined the biological activity of the previously reported derivatives which had not been accomplished before. Thus, this study is directed to synthesize some known and unknown derivatives of α-D-ribofuranose, to characterize the synthesized molecules by spectroscopic methods and to evaluate their hitherto unknown pharmacological and biological properties. The study also involves an attempt to find the binding affinity of the synthesized compounds against target macromolecules by molecular docking and to see if results are consistent with in vivo results. The molecular properties that affect the drug likeness of compounds, for instance, molecular weight, hydrogen bond acceptors and donors, lipophilicity, molar refractivity were also assessed by in silico approach.

2. Materials and methods

2.1. Chemicals and equipment

The starting compound of this study is a derivative of α-D-ribofuranose known as 3-O-benzyl-4-C-(hydroxymethyl)-1, 2-O-isopropylidene-α-D-ribofuranose which is widely utilized for synthesizing bridged nucleoside analogues (Hari et al., 2002; Rahman et al., 2008; Sharma et al., 2015). Chemicals and solvents applied in the synthetic procedures were procured from either Sigma-Aldrich (USA) or Merck (Germany). Solvents were further purified by drying over CaH₂ and distillation. All glassware was well dried before each reaction. The completion of chemical reactions was detected by thin layer chromatography (TLC) in befitting solvent system. Column chromatography with silica gel 60–120 mesh (LobaChemie, India) was employed to purify crude products. Fourier-transform infrared (FTIR) spectra were recorded from Centre of Advanced Research in Science, University of Dhaka with FTIR spectrophotometer (IRPrestige 21, Shimadzu Corporation, Japan). Proton and carbon nuclear magnetic resonance i.e. 1H NMR and 13C NMR spectra were produced on Bruker (Burker AMX-400) or JNM-ECS-300 (JEOL) spectrometers. Mass spectra were recorded using matrix assisted laser desorption/ionization coupled to time of flight (MALDI-TOF) technique in Spiral TOF JMS-S3000 instrument (JEOL).

2.2. Synthesis of 3, 5-di-O-benzyl-4-C-(hydroxymethyl)-1, 2-O-isopropylidene-α-D-ribofuranose (2)

Sodium hydride (0.93 g, 38.60 mmol) was taken into the solution of the starting material 1 (6 g, 19.30 mmol) in dimethyl formamide (80 ml) and benzyl bromide (2.50 ml, 21.20 mmol) at 0 °C. The reaction mixture was then stirred under a N₂ atmosphere to prevent moisture interference for 8 h at room temperature. After the completion of the reaction, the reaction mixture was quenched by adding ice cold water (100 ml) and the resulting mixture was extracted by ethyl acetate (3x100 ml). The extract was washed with water, dried over sodium sulfate and concentrated. Column chromatography (n-hexane: ethyl acetate = 3:1) of the concentrated residue yielded the product 2 as a transparent oil. Yield: 2.78 g, 83.7%; Rₓ = 0.57 (n-hexane: ethyl acetate = 2:1); IR (thin film, cm⁻¹): 3570 (O–H), 2940–2870 (C–H), 1452 (C=O, aromatic), 739 (C–H, aromatic); 1H NMR (400 MHz, CDCl₃, δ/ppm): 1.35 (s, 3H), 1.636 (s, 3H), 3.54 (d, J = 10.4 Hz, 1H), 3.61 (d, J = 10.4 Hz, 1H), 3.83 (d, J = 12 Hz, 1H), 3.94 (d, J = 12 Hz, 1H), 4.28 (d, J = 5.2 Hz, 1H), 4.56 (m, 4H), 4.65 (s, 1H), 4.78 (d, J = 11.6 Hz, 1H), 5.79 (d, J = 3.6 Hz, 1H), 7.34 (m, 10H).

2.3. Synthesis of 4-C-(acetoxymethyl)-3, 5-di-O-benzyl-1, 2-O-isopropylidene-α-D-ribofuranose (3)

Pyridine (1.20 ml, 15 mmol) was added in a dried vessel containing compound 2 (3 g, 7.5 mmol) in acetic anhydride (2.10 ml, 22.50 mmol) under fume-hood and was stirred for 3 h at room temperature. After diluting the reaction with water, the organic phase was extracted by ethyl acetate (3x100 ml) and concentrated using a rotatory evaporator. The concentrate was then co-evaporated with 20 ml toluene to remove excess pyridine. Column chromatography (n-hexane: ethyl acetate = 3:1) of the residue produced purified product 3 as a transparent oil. Yield: 2.78 g, 83.7%; Rₓ = 0.57 (n-hexane: ethyl acetate = 2:1); IR (thin film, cm⁻¹): 3028 (C–H), aromatic); 2870 (C–H), 1738 (C=O, ester), 1449 (C=C, aromatic), 741 (C–H, Aromatic); 1H NMR (400 MHz, CDCl₃, δ/ppm): 1.33 (s, 3H), 1.62 (s, 3H), 2.03 (s, 3H), 3.45 (d, J = 10.4 Hz, 1H), 3.56 (d, J = 10.8 Hz , 1H), 4.26 (d, J = 5.2 Hz, 1H), 4.31 (d, J = 12 Hz, 1H), 4.44 (d, J = 12 Hz, 1H), 4.45 (d, J = 10.4 Hz, 2H), 4.64 (d, J = 12.4, 2H), 4.74 (d, J = 12 Hz, 1H), 5.77 (d, J = 3.6 Hz 1H), 7.30 (m, 9H), 7.24 (d, J = 8 Hz, 1H).

2.4. Synthesis of 3, 5-di-O-benzyl-4-C-(hydroxymethyl)-O-methyl - D-ribofuranose (4)

Water (6.00 ml) and methanol (12.00 ml) and 28% HCl (31.00 ml) were added sequentially in a vessel containing 3 (1.5 g, 3.39 mmol) and was stirred at room temperature for 19 h. The organic phase extraction was carried out with ethyl acetate (3 x 100 ml), washed with brine and dried over anhydrous sodium sulfate. The concentrated residue was subjected to column chromatography (n-hexane: ethyl acetate = 3:1) to acquire the purified product 4 as a transparent oil. Yield: 1.01 g, 79.56%; Rₓ = 0.18 (n-hexane: ethyl acetate = 2:1); IR (thin film, cm⁻¹): 3390–3317 (O–H), 3031 (C–H, aromatic); 2930–2863 (C–H), 1454 (C=C, aromatic), 1044 (C=O), 1101 (C=O), 739 (C–H, aromatic); 1H NMR (300 MHz, CDCl₃, δ/ppm): 3.28 (s, 3H), 3.39 (d, J = 9.6 Hz, 1H), 7.30 (m, 9H), 7.24 (d, J = 8 Hz, 1H), 4.26 (d, J = 5.2 Hz, 1H), 4.31 (d, J = 12 Hz, 1H), 4.44 (d, J = 12 Hz, 1H), 4.45 (d, J = 10.4 Hz, 2H), 4.64 (d, J = 12.4, 2H), 4.74 (d, J = 12 Hz, 1H), 5.77 (d, J = 3.6 Hz 1H), 7.30 (m, 9H), 7.24 (d, J = 8 Hz, 1H).
2.5. Synthesis of 4-C-(acetoxymethyl)-3, 5-di-O-benzyl-O-methyl - D-ribofuranose (5)

Pyridine (0.40 ml, 5 mmol) was added in a dried vessel containing compound 4 (0.90 g, 2.40 mmol). Acetic anhydride (0.23 ml, 2.40 mmol) was added to the mixture under fume-hood and was stirred for 3 h at room temperature. After quenching the reaction with water, the organic phase was extracted by ethyl acetate (3 × 100 ml), washed with brine and concentrated using a rotatory evaporator. The concentrate was then evaporated with 20 ml toluene to eliminate excess pyridine. Column chromatography (n-hexane: ethyl acetate = 3:1) of the residue produced purified product 5 as a transparent oil. Yield: 0.85 g, 85.04%; Rf = 0.29 (n-hexane: ethyl acetate = 2:1); IR (thin film, cm⁻¹): 3318 (O=H), 3032 (C-H, aromatic), 2923–2864 (C-H), 1741 (C=O, ester), 1454 (C=C, aromatic), 739 (C-H, aromatic); ¹³C NMR (400 MHz, CDCl₃, δ/ppm): 2.10 (s, 3H), 3.33 (s, 3H), 3.37–3.41 (m, 2H), 3.39 (d, J = 9.2 Hz, 1H), 3.46 (br s, 1H), 3.64 (d, J = 5.0 Hz, 1H), 4.23 (d, J = 5 Hz, 1H), 4.31 (d, J = 11.9 Hz, 1H), 4.50 (d, J = 11.9 Hz, 1H), 4.58 (d, J = 3.2 Hz, 1H), 4.67 (d, J = 4.2 Hz, 1H), 4.91 (s, 1H), 7.31–7.41 (m, 10H), ²⁷Al NMR (100 MHz, CDCl₃, δ/ppm): 20.99, 54.98, 64.67, 72.92, 73.43, 73.59, 73.85, 81.35, 83.75, 107.85, 127.68 (2C), 127.73(2C), 127.78, 128.03, 128.43 (2C), 128.51(2C), 128.59 (2C), 137.56, 137.98. HRMS (MALDI-TOF) m/z: [M+Na]+ calculated for C₂₃H₂₅O₆Na 397.1622; found 397.1627.

2.6. Synthesis of 3-O-benzyl-4-C-(hydroxymethyl)-D-ribofuranose (6)

Compound 1 (0.50 g, 1.61 mmol) in 50% (w/v) acetic acid (1.94 ml, 16.11 mmol) was kept stirring at room temperature for 2 h. The organic phase was then extracted using ethyl acetate (3 × 100 ml), washed with water and dried over anhydrous sodium sulfate. The organic phase was then concentrated using a rotatory evaporator. Finally, the product 5 was purified as a transparent oil by column chromatography (n-hexane: ethyl acetate = 1:99). Yield: 0.293 g, 67.7%; Rf = 0.16 (n-hexane: ethyl acetate = 1:99); IR (thin film, cm⁻¹): 3391–3318 (O=H), 2930–2878 (C-H), 1456 (C-C, aromatic ring), 745 (C-H, aromatic); ¹³C NMR (400 MHz, DMSO d₆, δ/ppm): 3.09 (m, 2H), 3.45 (m, 2H), 3.62 (m, 1H), 3.84 (d, J = 11.6 Hz, 1H), 4.42 (m, 2H), 4.65 (m, 2H), 4.85 (m, 2H), 6.29 (m, 1H), 7.21 (m, 5H), ²⁷Al NMR (75 MHz, CDCl₃, δ/ppm): 62.67, 65.01, 68.40, 71.10, 73.21, 80.05, 95.09, 127.28, 127.53 (2C), 128.16 (2C), 139.09. HRMS (MALDI-TOF) m/z: [M+Na]+ calculated for C₁₃H₁₉O₄Na 249.1001; found 249.0991.

2.7. Animals

Swiss albino mice (Mus musculus) of around 25–30 g of either sex having age range of around 4–5 weeks, collected from Jahangirnagar University were used for investigation. Animals were used in experiments in lowest possible numbers maintaining the ethical standards and guidelines devised by Swiss Academy of Medical Sciences. Ethics committee approval number: Ref: DU/Pharm ETA: 01_09/2019.
2.11. Antioxidant activity

The antioxidant property of the test samples was evaluated by percent inhibition of DPPH (2, 2-diphenyl-1-picrylhydrazyl) free radical where ascorbic acid was used as standard (Ahmed et al., 2019). Concentration of the compound that provides 50% inhibition or reduction of DPPH, also known as IC₅₀ was calculated from the equation of the percentage inhibition versus concentration of the sample logarithmic curve.

2.12. Statistical analysis

Calculations were performed using XLSTAT software for Microsoft Excel 2013. Values were shown as the mean ± standard error of mean (SEM). One-way analysis of variance (ANOVA) and Dunnett’s test were carried out where P < 0.05 was regarded as statistically significant.

2.13. Molecular docking simulation

The structure of proteins phospholipase A₂ (PLA₂) (PDB code: 4UY1), cyclooxygenase-1 (COX-1) (PDB code: 1E0G), cyclooxygenase-2 (COX-2) (PDB code: 5IKT), NF-κB-inducing kinase (NIK) (PDB code: 4DIV), interleukin-1 receptor associated kinase-4 (IRAK-4) (PDB code: 5KX7) was collected from Research Collaboratory for Structural Bioinformatics Protein Data Bank (RCSB PDB) (Berman et al., 2002). Energy minimization of ligands was performed in Open Babel (version 2.4.0) (O’Boyle et al., 2011). Validation of docking procedure was done by re-docking the native ligands in the binding pockets of respective proteins. The target protein–ligand docking was executed using AutoDock Vina (version 1.1.2) (Trott and Olson, 2009). Docking simulation was done using grid with its centers positioned at the active site of respective proteins and the dimensions of the grid were 25.00, 25.00, 25.00 Å. The interactions of ligands with target macromolecules were analyzed by Discovery Studio Client 2019.

2.14. Lipinski’s rule of five prediction

Molecular weight, hydrogen bond donors and acceptors, lipophilicity, molar refractivity of the synthesized compounds were calculated using SwissADME (Daina et al., 2017).

3. Results

3.1. Synthesis of α-D-ribofuranose derivatives

Ribofuranose derivatives were synthesized from the starting material 3-O-benzyl-4-C-(hydroxymethyl)-1, 2-O-isopropylidene-α-D-ribofuranose (1) (Scheme 1). Compound 1 was subjected to benzylation at room temperature which yielded di-benzyl derivative 2 along with an undesired tri-benzyl derivative similar to that reported in our previous study (Rahman et al., 2020). Compound 2 was separated from the undesired product and reacted with acetic anhydride to produce acetylated derivative 3 in high yield. Acidic hydrolysis of compound 3 with 28% HCl followed by methylation produced compound 4 in good yield with additional removal of acetyl group. Compound 4 was acetylated using reaction conditions similar to previous acetylation. Compound 6 was also synthesized by hydrolytic cleavage of isopropylidene ring of the starting material 1. The structures of the synthesized compounds were established by FTIR, 1H NMR, 13C NMR and mass spectra as presented in Sections 2.2, 2.3, 2.4, 2.5 and 2.6. Compound 2 and 3 were known compounds which were confirmed by comparing the spectral data with the reported data (Koshkin et al., 1998).

3.2. Analgesic activity of synthesized compounds

3.2.1. Acetic acid induced writhing method

The peripheral analgesic property of synthesized compounds was investigated by acetic acid induced writhing method. The effects of the synthesized compounds to subside the pain caused by acetic acid are presented in Table 1. In this method compound 3 showed potent analgesic activity with inhibition of writhing of 54.74% and 79.74% at doses of 25 mg/kg and 50 mg/kg, respectively, whereas diclofenac as positive control at dose of 25 mg/kg inhibited writhing by 83.19%. At 50 mg/kg dose compound 2, 4 and 5 showed writhing inhibition of 63.36%, 59.91% and 54.45%, respectively. Compound 6 at both doses showed mild analgesic activity compared to other test compounds.

3.2.2. Tail immersion method

The changes in sensitivity of test animals due to the analgesic activity of the test compounds were compared by tail flick method.

| Treatment (mg/kg) | Average writhing | Inhibition of writhing (%) |
|------------------|------------------|---------------------------|
| Control (–)      | 23.20 ± 0.30     | –                         |
| Diclofenac (25)  | 3.90 ± 0.19***   | 83.19                     |
| 2 (25)           | 13.40 ± 0.37***  | 42.24                     |
| 2 (50)           | 8.50 ± 0.22***   | 63.36                     |
| 3 (25)           | 10.50 ± 0.27***  | 54.74                     |
| 3 (50)           | 4.70 ± 0.26***   | 79.74                     |
| 4 (25)           | 14.60 ± 0.40***  | 37.07                     |
| 4 (50)           | 9.30 ± 0.25***   | 59.91                     |
| 5 (25)           | 17.20 ± 0.52***  | 25.86                     |
| 5 (50)           | 10.80 ± 0.26***  | 53.45                     |
| 6 (25)           | 20.30 ± 0.71**   | 11.64                     |
| 6 (50)           | 17.60 ± 0.86***  | 24.14                     |

*P < 0.001, *P < 0.01, *P < 0.05 compared with negative control (one-way ANOVA followed by Dunnett’s test).
and the data are presented in Fig. 2. Compound 3 at the dose 50 mg/kg showed highest tail flicking reaction time of 2.55 ± 0.1 min after 30 min of oral administration among the test compounds which is comparable to morphine’s 3.28 ± 0.15 min. Compound 3 at 25 mg/kg and compound 2 at 50 mg/kg showed reaction time of 2.00 ± 0.16 and 1.74 ± 0.12 min, respectively, after 30 min of oral administration. After 60 min of oral administration, compound 3 at 25 mg/kg and 50 mg/kg showed reaction time of 1.51 ± 0.11 and 2.03 ± 0.16 min which is superior to other test compounds at the same dose and same time interval.

3.3. Anti-inflammatory activity of synthesized compounds

The mean paw volume (ml) and percentage of paw edema inhibition of the compounds are shown in Table 2. From the statistical evaluation, it is evident that all the compounds at 100 mg/kg dose showed reduction of paw volume from the first hour and onwards. All the compounds showed anti-inflammatory activity among which compound 2 and 6 had prominent activity. The paw edema inhibition of compound 2 was 15.79%, 58.68%, 77.32% and 91.15% and compound 6 was 14.29%, 58.68%, 79.38% and 95.13% in 1st, 2nd, 3rd and 4th hour, respectively, whereas paw edema inhibition was 31.58%, 70.66%, 86.08% and 97.79% in 1st, 2nd, 3rd and 4th hour, respectively, for positive control aceclofenac (100 mg/kg). Compound 3, 4 and 5 also showed significant reduction in paw edema volume.

3.4. Antimicrobial and antioxidant activity of synthesized compounds

Antimicrobial activity of the compounds 2, 3, 4, 5 and 6 were assessed by disk diffusion method on both Gram-positive and Gram-negative species. DPPH free radical scavenging method was applied to determine the antioxidant potential of the compounds. However, the synthesized compounds did not show notable antimicrobial and antioxidant properties (data not shown).

3.5. Molecular docking simulation

Molecular docking studies showed that the binding affinities of compound 2, 3, 4 and 5 with PLA2 were greater than the native ligand’s binding affinity, meaning PLA2 may be a target for these compounds for their anti-inflammatory activity. Compound 3 effectively interacted with PLA2 by forming hydrogen bond with Gly28A and Lys61A residue shown in Fig. 3. Among the four compounds, compound 3 had the highest affinity towards COX-1 which is also higher than the native ligand ibuprofen’s binding affinity. Compound 3 formed hydrogen bond with Arg120A residue of COX-1 as depicted in Fig. 4 and this Arg120A residue is required for high affinity binding of arachidonic acid with cyclooxygenase (Vecchio et al., 2012). These interactions of compound 3 with PLA2 and COX-1 may be accounted for significant anti-inflammatory and analgesic activity of this compound in biological study. However, the compounds showed lesser affinity towards COX-2 than COX-1 and binding patterns of the compounds with COX-2 are shown in Fig. 5. Compound 2 and 5 effectively bound with NIK as illustrated in Fig. 6 with binding affinities which are comparable to that of corresponding native ligand. Compound 2 formed hydrogen bond with Ser328B residue of IRAK-4 ATP binding site shown in Fig. 7 and had higher binding affinity than that of corresponding native ligand and other synthesized compounds. The binding affinities of synthesized molecules with these target proteins are shown in Table 3.

3.6. Lipinski’s rule of five prediction

Molecular weight, octanol–water partition coefficient, number of hydrogen bond donor, number of hydrogen bond acceptor and molar refractivity properties for the synthesized compounds are presented in Table 4. All the compounds 2, 3, 4, 5 and 6 passed the Lipinski’s rule of five screening with no violation of any of the acceptance criteria.
4. Discussion

In the current study synthesized α-D-ribofuranose derivatives were subjected to various pharmacological screening. Analgesic activity of synthesized compounds was evaluated by two different methods. In acetic acid induced writhing method, intra peritoneal administration of acetic acid prompts an acute inflammatory response followed by activation of the nociceptors (Dzoyem et al., 2017). The tail flick method is also broadly applied to assess anti-nociceptive property of compounds. Exposing the tail of mice to heat activates heat sensitive receptors, transient receptor potential vanilloid type 1 and 3 (TRPV1 and TRPV3) receptors promoting pain transmission (Leksiri et al., 2020). Compound 3 exhibited potent analgesic activity in both the tests. Compound 2, 4 and 5 also exhibited moderate analgesic activity in both the tests. Analgesic activity of compound 3, 4 and 5 is reported for the first time in this study. D-ribose derivatives and several nucleoside analogues containing modified ribose has exhibited notable anti-nociceptive effect (El-Gazzar et al., 2009; Gebhart and Jorgenson, 2004; Jarvis et al., 2002; Ueki et al., 2013). Structural resemblance

Table 2

| Treatment (mg/kg) | Mean Paw Volume (ml) (inhibition of edema %) |
|------------------|---------------------------------------------|
|                  | Basal 1st hr 2nd hr 3rd hr 4th hr           |
| Control (–)      | 0.57 ± 0.02 0.83 ± 0.01 0.90 ± 0.02 0.96 ± 0.02 1.02 ± 0.03 |
| Aceclofenac (100) | 0.51 ± 0.01 0.69 ± 0.01*** (31.58) 0.61 ± 0.02*** (70.66) 0.56 ± 0.03*** (86.08) 0.52 ± 0.02*** (97.79) |
| 2 (100)          | 0.51 ± 0.02 0.73 ± 0.02** (15.79) 0.64 ± 0.02*** (58.68) 0.60 ± 0.02*** (77.32) 0.55 ± 0.02*** (91.15) |
| 3 (100)          | 0.52 ± 0.01 0.76 ± 0.02 (11.28) 0.70 ± 0.02*** (46.71) 0.65 ± 0.02*** (67.01) 0.58 ± 0.02*** (86.73) |
| 4 (100)          | 0.57 ± 0.02 0.80 ± 0.03 (13.53) 0.71 ± 0.03*** (58.08) 0.67 ± 0.03*** (73.71) 0.63 ± 0.04*** (88.00) |
| 5 (100)          | 0.56 ± 0.01 0.80 ± 0.02 (12.03) 0.74 ± 0.01*** (46.71) 0.70 ± 0.02*** (63.40) 0.66 ± 0.02*** (78.32) |
| 6 (100)          | 0.52 ± 0.01 0.74 ± 0.02** (14.29) 0.65 ± 0.02*** (58.68) 0.60 ± 0.02*** (79.38) 0.54 ± 0.01*** (95.13) |

Paw volume values are presented as mean ± S.E.M. (n = 5); ***P < 0.001, **P < 0.01, *P < 0.05 compared to control (–) by Dunnett’s test.

Fig. 3. Interaction of synthesized compounds (a) 2, (b) 3, (c) 4, (d) 5 and (e) 6 with PLA2 (4UY1) active site. Green: conventional hydrogen bond; pink-violet: hydrophobic; light green: Van der Waals; cyan: carbon hydrogen; orange: pi-anion/pi-cation; grey: metal acceptor; red: electron donor–donor interaction.
Fig. 4. Interaction of synthesized compounds (a) 2, (b) 3, (c) 4, (d) 5 and (e) 6 with COX-1 (1EQG) active site. Green: conventional hydrogen bond; pink-violet: hydrophobic; light green: Van der Waals; cyan: carbon hydrogen bond/π-donor hydrogen bond; orange: π-cation; brown: π-sulfur interaction.

Fig. 5. Interaction of synthesized compounds (a) 2, (b) 3, (c) 4, (d) 5 and (e) 6 with COX-2 (5IKT) active site. Green: conventional hydrogen bond; pink-violet: hydrophobic; light green: Van der Waals; cyan: carbon hydrogen bond/π-donor hydrogen bond; orange: π-anion interaction.
Fig. 6. Interaction of synthesized compounds (a) 2, (b) 3, (c) 4, (d) 5 and (e) 6 with NIK (4IDV) active site. Green: conventional hydrogen bond; pink-violet: hydrophobic; light green: Van der Waals; cyan: carbon hydrogen bond/π-donor hydrogen bond; orange: π-cation; brown: π-sulfur; red: electron donor–donor interaction.

Fig. 7. Interaction of synthesized compounds (a) 2, (b) 3, (c) 4, (d) 5 and (e) 6 with IRAK-4 (5KX7) active site. Green: conventional hydrogen bond; pink-violet: hydrophobic; light green: Van der Waals; cyan: carbon hydrogen bond/π-donor hydrogen bond interaction.
of the synthesized compounds with ribose and its nucleosides with analgesic effect favors the results of anti-nociceptive tests. However, to understand the mechanism by which these compounds exhibit analgesic effects need more study and further structural modifications of these compounds may lead to better analogs.

Carrageenan induced paw edema test was carried out on laboratory animals to observe the anti-inflammatory effect of the compounds. Intra plantar injection of 1% carrageenan induces local edema that increases with time releasing different inflammatory mediators in two phases. In the first phase that may last up to 1.5 h after injection of carrageenan involves the release of histamine, 5-hydroxytryptamine, platelet activating factor, bradykinin whereas in the second phase prostaglandin and various cytokines dominates (Mehrzadi et al., 2020). All the compounds exhibited analgesic effects need more study and further structural modifications of these compounds may lead to better analogs.

Carrageenan-induced paw edema test was carried out on laboratory animals to observe the anti-inflammatory effect of the compounds. Intra plantar injection of 1% carrageenan induces local edema that increases with time releasing different inflammatory mediators in two phases. In the first phase that may last up to 1.5 h after injection of carrageenan involves the release of histamine, 5-hydroxytryptamine, platelet activating factor, bradykinin whereas in the second phase prostaglandin and various cytokines dominates (Mehrzadi et al., 2020). All the compounds exhibited promising anti-inflammatory activity with paw edema inhibition ranging from 78.32% to 95.15% in 4th hour. Anti-inflammatory effects of compound 3, 4, 5 and 6 are reported for the first time in the current study. Since the synthesized compounds exhibited considerable anti-inflammatory effect, we may assume that the compounds may interact with the inflammatory mediators or interfere in any of the mechanistic pathways associated with these mediators such as antagonistic action on these mediator’s receptor or interference in the biosynthesis procedure of these mediators. The observed activity of the compounds might be rationalized that the compounds are derivatives of D-ribose and possess structural similarity to some adenosine kinase inhibitors (Jarvis et al., 2004). PLA2 enzymes have roles in the arachidonic acid metabolic pathway and biosynthesis of eicosanoid in both physiological condition and in inflammation (Burke and Dennis, 2009). NIK is commonly related to signaling mechanism of various cytokines and regulation of inflammation-angiogenesis (Pflug and Stickelner, 2020), IRAK-4, a protein kinase, is necessary in numerous inflammatory signaling pathways (Lye et al., 2004). Synthesized compounds showed efficient binding with these molecular targets. Compound 2 had higher binding affinities toward COX-1 and NIK than the corresponding native ligands. Compound 3 interacted with Arg120A residue of COX-1 which is required for high affinity binding of arachidonic acid with cyclooxygenase (Vecchio et al., 2012). PLA2 enzyme has role in the arachidonic acid metabolic pathway and biosynthesis of eicosanoid in both physiological condition and in inflammation (Burke and Dennis, 2009). NIK is commonly related to signaling mechanism of various cytokines and regulation of inflammation-angiogenesis (Pflug and Stickelner, 2020), IRAK-4, a protein kinase, is necessary in numerous inflammatory signaling pathways (Lye et al., 2004). Synthesized compounds showed efficient binding with these molecular targets. Compound 2 had higher binding affinities toward COX-1 and NIK than the corresponding native ligands. Compound 3 interacted with Arg120A residue of COX-1 which is required for high affinity binding of arachidonic acid with cyclooxygenase (Vecchio et al., 2012). Compound 3 also interacted with PLAr and IRAK-4 active site effectively and these interactions may be accounted for significant anti-inflammatory and analgesic activity of this compound in biological study.

Lipinski’s rule of five illustrates molecular properties of candidate compounds related to pharmacokinetics for successful outcomes in drug development process. It indicates that when a molecule transgresses these rules it is likely to have poor pharmacokinetic property and not favorable as a lead for drug development (Lipinski, 2004). All synthesized compounds complied with acceptable criteria, therefore, these compounds may have good pharmacokinetic properties for consideration in drug development (Wang et al., 2015).

Table 3

| Protein (PDB code) | Native ligand* | 2 | 3 | 4 | 5 | 6 |
|-------------------|----------------|---|---|---|---|---|
| PLA2 (4UY1)       | −7.6           | −8.5| −8.9| −8.6| −8.1| −7.2|
| COX-1 (1EQC)      | −7.7           | −6.2| −8.1| −7.4| −7.0| −7.0|
| COX-2 (5IKT)      | −8.6           | −5.4| −6.4| −6.5| −6.0| −6.1|
| NIK (4IDV)        | −9.9           | −8.2| −7.8| −7.3| −8.0| −7.0|
| IRAK-4 (5KX7)     | −8.7           | −9.1| −8.9| −8.3| −8.2| −6.9|

* Native ligands of PLA2 (4UY1), COX-1 (1EQC), COX-2 (5IKT), NIK (4IDV), IRAK-4 (5KX7) are 5-(2,5-dimethyl-3-thienyl)-1H-pyrazole-3-carboxamide (PubChem CID: 78673871), ibuprofen (PubChem CID: 3672), tolfenamic acid (PubChem CID: 610479), 4-[3-(2-amino-5-(2-methoxyethoxy)pyrimidin-4-yl)]-1h-indol-5-yl)-2-methylbut-3-yn-2-ol (PubChem CID: 58221375), (n)-(3-aminoacarbonyl-1-methyl-pyrazol-4-yl)-6-(1-methylpyrazol-4-yl)pyridine-2-carboxamide (PubChem CID: 31693309), respectively.

Table 4

| Compounds (ligands) | Molecular weight | Acceptor of H-bond | Donor of H-bond | Molar Refractivity | LogP | No. of Violations |
|---------------------|------------------|--------------------|-----------------|--------------------|------|------------------|
| 2                   | 400.46           | 6                  | 1               | 106.40             | 1.16 | 0                |
| 3                   | 442.50           | 7                  | 0               | 116.14             | 1.91 | 0                |
| 4                   | 373.43           | 6                  | 2               | 98.94              | 0.72 | 0                |
| 5                   | 416.46           | 7                  | 1               | 108.68             | 1.09 | 0                |
| 6                   | 270.28           | 6                  | 4               | 64.99              | −0.95| 0                |

Acceptance criteria: molecular weight < 500, octanol–water partition coefficient (expressed as LogP) < 5, number of hydrogen bond donor < 5, number of hydrogen bond acceptor < 10, molar refractivity between 40 and 130.

5. Conclusion

In conclusion, five α-D-ribofuranose analogues were synthesized and evaluated for their pharmacological potential. The compound 3 manifested significant analgesic effect in peripheral and central analgesic assay whereas the other analogues demonstrated mild to moderate analgesic activity. In carrageenan-induced hind paw edema method both 2 and 6 exerted significant anti-inflammatory effect. In molecular docking study, compound 3

Sahiba Enam Spriha, Fahad Imtiaz Rahman and S. M. Abdur Rahman

Saudi Pharmaceutical Journal 29 (2021) 981–991

989
showed highest binding affinity with COX-1, COX-2 and PLA2 whereas compound 2 manifested highest binding affinity with NIK and IRAK-4. Further study and structural modifications are required to learn more about the biological properties and underlying mechanisms which is the subject of our future investigation.

Funding

This work was supported by the Ministry of Science, Information and Communication Technology, Government of the People’s Republic of Bangladesh (grant no. 39.00.0000.012.002.04.19.09).

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

Adamo, M., Pergoli, R., 2008. Synthesis and medicinal properties of 2-deoxysugar and ribose C-nucleoside. Curr. Org. Chem. 12, 1544–1569. https://doi.org/10.2174/1389207087878318.

Adukwu, E.C., Bowles, M., Edwards-Jones, V., 2016. Synthesis and anti-inflammatory activities of 4H-chromene and bicyclonucleoside monomers, oligomerisation, and unprecedented nucleic acid recognition. Tetrahedron. 74 (14), 3051–3063. https://doi.org/10.1016/j.tet.2018.02.034.

Aguero, J.M., 2002. Effective syntheses of C-nucleosides with 2′- and 3′-nucleoside analogues as potential anti-inflammatory agents. Heteroc. Chem. 12, 52–56. https://doi.org/10.1016/S0018-0671(01)00919-5.

Ahmed, S., El-Gazzar, A.-R., Hafez, H.N., Abbas, H.-A., 2009. S- and C-nucleosidoquinazoline as metabolic, inflammatory, infectious and systemic Diseases. Elsevier Inc., pp. 175–191.

Adukwu, E.C., Michielin, O., Zoete, V., 2017. SwissADME: A free web tool to evaluate drug-likeness. J. Cheminform. 3, 18. https://doi.org/10.1186/s13321-011-0018-1.

Al-Rehaily, A.J., Alqahtani, A.S., Hidayatullah, S., Rehman, M.T., Ahmed, S., Al-Rehaily, A.J., Alam, P., 2016. Synthesis and anti-inflammatory activities. Inflammation 44, 186–193. https://doi.org/10.1007/s10753-012-9810-7.

Ambroz, P., Vajragupta, O., Rojsitthisak, P., Mols, D., van den Bergh, L.S., 2015. Effective syntheses of 2′-deoxy-3′-fluoro-β-d-guanosine via 2,3-dideoxy-3-fluoro-α-d-ribose 1-phosphate. Tetrahedron Lett. 44 (14), 2899–2901. https://doi.org/10.1016/j.tetlet.2013.03.028.

Andrews, L.P., 2008. The role of interleukin 1 receptor-associated kinase-4 (IRAK-4) kinase activity in IRAK-4-mediated signaling. ASBMB. 279 (39), 40637–40658. https://doi.org/10.1016/j.apsb.2011.04.003.

Aminov, R.I., Kuzmin, A., 2016. Synthesis of nucleosides from unprotected or 5-O-monoprotected D-ribose. Org. Lett. 17, 4604–4607. https://doi.org/10.1021/acs.orglett.5b02332.

Aoki, K., Imaizumi, M., Obika, S., 2009. Synthesis of several types of bridged bicyclonucleosides as potential anti-inflammatory agents. Heteroc. Chem. 12, 1195–1215. https://doi.org/10.1016/j.hetero.2008.12.017.

Aoyagi, Y., Takeda, T., 2006. Effective syntheses of 2′-deoxy-3′-fluoro-β-D-guanosine via 2,3-dideoxy-3-fluoro-α-D-ribose 1-phosphate. Tetrahedron Lett. 44 (14), 2899–2901. https://doi.org/10.1016/j.tetlet.2003.05.015.

Arai, A., 2003. LNA (Locked Nucleic Acids): synthesis of the 2′-O-methyl and 2′-O-allyl 2′-deoxy-L-ribofuranosyl derivatives. Res. Chem. Inf. 42 (2), 1195–1215. https://doi.org/10.1016/j.rci.2003.05.027.

Arai, A., 2003. LNA (Locked Nucleic Acids): synthesis of the 2′-O-allyl and 2′-O-methyl 2′-deoxy-L-ribofuranosyl derivatives. Res. Chem. Inf. 42 (2), 1195–1215. https://doi.org/10.1016/j.rci.2003.05.027.

Arai, A., 2003. LNA (Locked Nucleic Acids): synthesis of the 2′-O-allyl and 2′-O-methyl 2′-deoxy-L-ribofuranosyl derivatives. Res. Chem. Inf. 42 (2), 1195–1215. https://doi.org/10.1016/j.rci.2003.05.027.

Arai, A., 2003. LNA (Locked Nucleic Acids): synthesis of the 2′-O-allyl and 2′-O-methyl 2′-deoxy-L-ribofuranosyl derivatives. Res. Chem. Inf. 42 (2), 1195–1215. https://doi.org/10.1016/j.rci.2003.05.027.

Arai, A., 2003. LNA (Locked Nucleic Acids): synthesis of the 2′-O-allyl and 2′-O-methyl 2′-deoxy-L-ribofuranosyl derivatives. Res. Chem. Inf. 42 (2), 1195–1215. https://doi.org/10.1016/j.rci.2003.05.027.

Arai, A., 2003. LNA (Locked Nucleic Acids): synthesis of the 2′-O-allyl and 2′-O-methyl 2′-deoxy-L-ribofuranosyl derivatives. Res. Chem. Inf. 42 (2), 1195–1215. https://doi.org/10.1016/j.rci.2003.05.027.
in mice. Tohoku J. Exp. Med. 229 (3), 195–201. https://doi.org/10.1620/tjem.229.195.
Vecchio, Alex J., Orlando, Benjamin J., Nandagiri, Ritwik, Malkowski, Michael G., 2012. Investigating substrate promiscuity in cyclooxygenase-2 the role of Arg-120 and residues lining the hydrophobic groove. J. Biol. Chem. 287 (29), 24619–24630. https://doi.org/10.1074/jbc.M112.372243.
Wagner, S., Herrick, J., Shecterle, L.M., St. Cyr, J.A., 2009. D-ribose, a metabolic substrate for congestive heart failure. Prog. Cardiovasc. Nurs. 24, 59–60. https://doi.org/10.1111/j.1751-7117.2009.00033.x.
Wang, Y., Xing, J., Xu, Y., Zhou, N., Peng, J., Xiong, Z., Liu, X., Luo, X., Luo, C., Chen, K., Zheng, M., Jiang, H., 2015. In silico ADME/T modelling for rational drug design. Q. Rev. Biophys. 48, 468–515. https://doi.org/10.1017/S0033583515000190.
Zhang, Bin, Wu, Jia-Ting, Zheng, Cai-Juan, Zhou, Xue-Ming, Yu, Zhang-Xin, Li, Wan-Shan, Chen, Guang-Ying, Zhai, Guo-Yuan, 2021. Bioactive cyclohexene derivatives from a mangrove-derived fungus Cladosporium sp. JMJ22. Fitoterapia. 149, 104823. https://doi.org/10.1016/j.fitote.2020.104823.