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Synthesis, ADMET and pharmacokinetic analysis of new isoxazolidine-pyrazole hybrids

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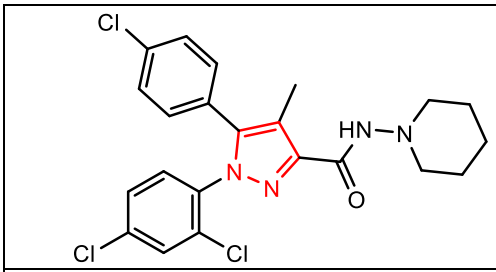
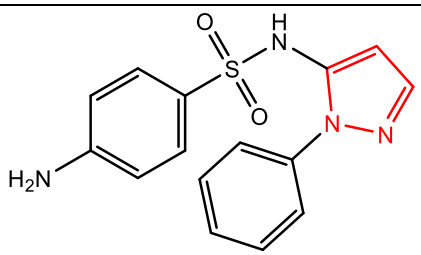
Abstract: Interest in hybrid compounds containing two or more different rings continues to grow due to the impressive biological results that have been derived in numerous examples. In this work, we have synthesized new isoxazolidine-pyrazole hybrids and study ADMET and pharmacokinetic analysis. These results outlined that the titled compounds have good ADMET properties, which are a requisite for a molecule to show acceptable drug-like and pharmacokinetic properties.

Keywords: pharmacokinetic analysis, ADMET, isoxazolidine, 1,3-dipolar cycloaddition, pyrazole.

1. INTRODUCTION

Interest in the synthesis of heterocycles continues to grow due to their multiple and diverse biological activities [1]. This is explained by the presence of a wide range of heterocycles containing one or more heteroatoms. For example, compounds containing pyrazole, a five-membered heterocycle containing two adjacent nitrogen atoms, have shown remarkable biological interest such as antitumor [2], antichagastic [3], antihyperglycemic [4], pesticidal [5], antifungal [6], antimicrobial [2,7], leishmanicidal [8-10] and anti-inflammatory activities [11]. Also, the pyrazole ring is found in biologically active natural compounds [12]. Recently, among the drugs approved and marketed by the FDA, we find those that contain the pyrazole ring (Figure 1). In this regard, pyrazole constitutes a research subject of great importance for organizational researchers.

	
Acomplia (Cannabinoid receptor)	Sulfaphenazole (Anti-bacterial)

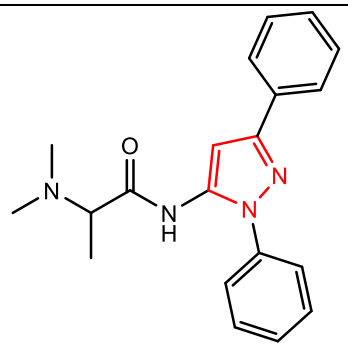
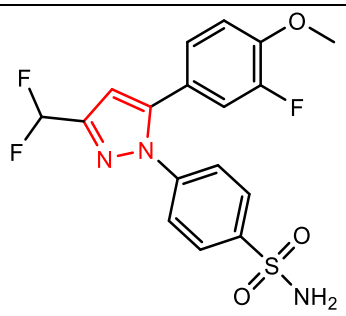
	
Diphenamizole (Analgesic)	Deracoxib (Anti-inflammatory)

Figure 1. Some pyrazole derivatives drugs approved and marketed by the FDA.

Furthermore, our research group has developed a wide variety of molecules derived from isoxazolidine possessing promising biological activities such as antimicrobial [13,14], antioxidant [14,15], antidiabetic [16-19], anticancer [20]. Based on the biological diversity of isoxazolidine and pyrazole, the idea of this article is to synthesize hybrid molecules between these two heterocycles in order to improve their biological potential.

2. MATERIALS AND METHODS

2.1. Chemistry

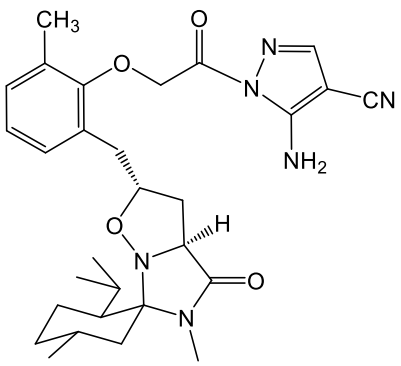
2.1.1. Synthesis of compounds 4a-d

Compounds **4a-d** were prepared according to the same operating procedure published by Alhawday et al. [18].

2.1.2. Synthesis of isoxazolidine derivatives 5a-d

Ethoxymethylene malononitrile (1 eq.) was added gently to a solution of **4a-c** (1 eq.) in absolute ethanol. The reaction mixture was maintained at reflux for 18h, cooled, concentrated and subsequently chromatographed on a silica gel column (7/3 Cyclohexane/EtOAc) to access isoxazolidine-pyrazole hybrids **5a-d** (colorless oil).

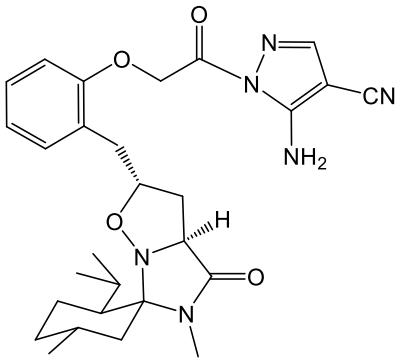
(**5a**)



84%; IR (cm^{-1}): 2252 (C≡N), 3241 (NH_2). NMR (1H, $CDCl_3$, 400MHz) 0.76(d, 3H, J=6.4Hz, Me); 0.87(d, 3H, J=6.4Hz, Me); 0.88(d, 3H, J=6.4Hz, Me); 0.87-0.88(m, 1H); 1.12(t, 1H, J=12.4Hz); 1.32-1.60 (m, 3H) 1.65-1.73 (m, 3H); 1.76-1.83 (m, 1H); 1.85-1.98 (m, 1H); 2.12 (ddd, 1H, J=2.4, 8.6, 10.8Hz); 2.15 (s, 3H, CH_3); 2.64-2.67 (m, 1H), 2.70 (s, 3H, NMe); 2.84 (brd, 2H, J=12.0Hz); 3.79-3.84(m, 1H); 4.00(d, 1H, 8.4Hz); 4.59 (brs, 2H); 6.91(dd, 1H, J=2.2, 8.0Hz); 7.32 (dd, 1H, J=2.0, 8.0Hz); 7.66 (t, 1H, J=8.1Hz); 9.56 (s, 1H).

NMR (13C, $CDCl_3$, 100 MHz) 18.9; 20.1; 20.2; 20.6; 21.0; 21.1; 23.4; 26.7; 29.5; 29.7; 31.4; 34.4; 37.8; 40.3; 44.02; 56.6; 67.3; 73.7; 95.0; 111.5; 115.1; 119.2; 124.1;129.5; 133.7; 141.8; 152.8; 165.1; 171.0.

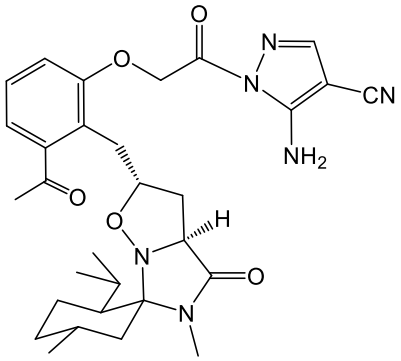
(**5b**)



86%; IR (cm^{-1}): 2250 (C≡N), 3242 (NH_2). NMR (1H, $CDCl_3$, 400MHz) 0.77(d, 3H, J=6.4Hz, Me); 0.87(d, 3H, J=6.4Hz, Me); 0.88(d, 3H, J=6.4Hz, Me); 0.87-0.88(m, 1H); 1.04(t, 1H, J=12.4Hz); 1.34-1.62 (m, 3H) 1.66-1.74 (m, 3H); 1.77-1.81 (m, 1H); 1.85-1.99 (m, 1H); 2.11 (ddd, 1H, J=2.4, 8.6, 10.8Hz); 2.65-2.68 (m, 1H), 2.71 (s, 3H, NMe); 2.83 (brd, 2H, J=12.0Hz); 3.81-3.87(m, 1H); 4.01(d, 1H, 8.4Hz); 4.68 (brs, 2H); 6.85-6.91(m, 3H); 7.35 (dd, 1H, J=2.0, 8.0Hz); 9.71 (s, 1H).

NMR (13C, $CDCl_3$, 100 MHz) 19.3; 20.2; 20.6; 21.0; 21.1; 23.3; 26.5; 29.3; 29.5; 31.4; 34.4; 37.8; 40.3; 44.0; 56.8; 68.3; 74.3; 94.0; 111.4; 115.0; 119.0; 123.1; 128.2; 133.5; 143.9; 151.8; 163.9; 171.7.

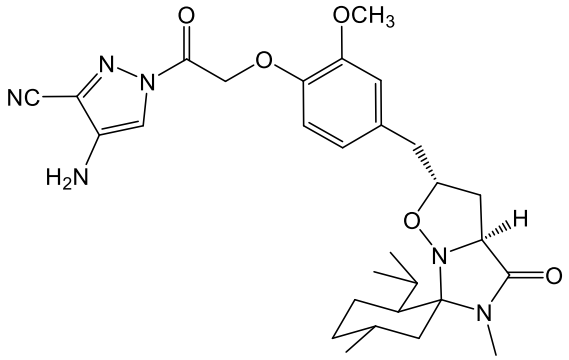
(**5c**)



88%; IR (cm^{-1}): 2254 (C≡N), 3241 (NH_2). NMR (1H, $CDCl_3$, 400MHz) 0.73(d, 3H, J=6.4Hz, Me); 0.77(d, 3H, J=6.4Hz, Me); 0.79(d, 3H, J=6.4Hz, Me); 0.77-0.79(m, 1H); 1.02(t, 1H, J=12.4Hz); 1.31-1.57 (m, 3H) 1.61-1.70 (m, 3H); 1.73-1.81 (m, 1H); 1.83-1.95 (m, 1H); 2.09 (ddd, 1H, J=2.4, 8.6, 10.8Hz); 2.51 (s, 3H, CH_3); 2.61-2.66 (m, 1H), 2.69 (s, 3H, NMe); 2.81 (brd, 2H, J=12.0Hz); 3.77-3.81(m, 1H); 4.02(d, 1H, 8.4Hz); 4.61 (brs, 2H); 7.09(dd, 1H, J=2.0, 7.6Hz); 7.44 (dd, 1H, J=2.0, 7.6Hz); 7.55 (t, 1H, J=7.6Hz); 9.58 (s, 1H).

NMR (13C, $CDCl_3$, 100 MHz) 18.7; 20.3; 20.5; 21.1; 21.2; 23.4; 26.7; 29.5; 29.7; 30.1; 31.4; 34.4; 37.8; 40.3; 44.02; 56.6; 67.7; 74.7; 95.4; 111.5; 115.0; 119.1; 124.1;129.5; 134.9; 141.6; 153.8; 166.1; 171.0; 201.0.

(**5d**)

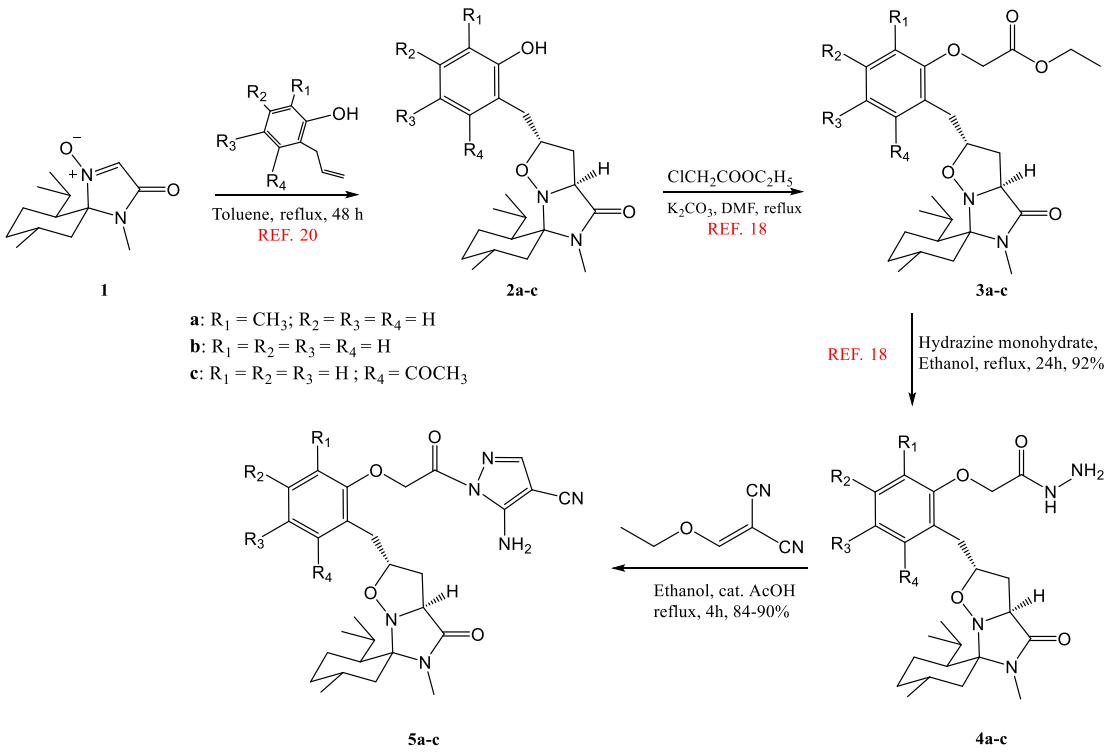


90%; IR (cm^{-1}): 2249 (C≡N), 3245 (NH_2). NMR (1H, $CDCl_3$, 400MHz) 0.69(d, 3H, J=6.4Hz, Me); 0.84(d, 3H, J=6.4Hz, Me); 0.85(d, 3H, J=6.4Hz, Me); 0.83-0.86(m, 1H); 1.12(t, 1H, J=12.4Hz); 1.31 (dd, 2H, J=2.4, 12Hz); 1.35-1.42 (m, 1H); 1.62-1.72 (m, 3H); 1.76-1.79 (m, 1H); 1.86-1.98 (m, 1H); 2.21 (ddd, 1H, J=2.0, 8.8, 10.8Hz); 2.66-2.68 (m, 1H), 2.69 (s, 3H, NMe); 2.80 (brd, 2H, J=11.6Hz); 3.80-3.85(m, 1H); 4.00(d, 1H, 8.4Hz); 4.65 (brd, 2H, J=1.2Hz); 6.75-6.77(m, 2H); 6.87 (d, 1H, J=8.0Hz); 9.67 (s, 1H).

NMR (13C, $CDCl_3$, 100 MHz) 18.3; 22.1; 22.2; 24.1; 24.3; 26.0; 29.7; 29.74; 34.4; 38.6; 38.8; 40.3; 48.0; 55.8; 66.3; 70.3; 87.4; 90.0; 113.0; 117.0; 121.2; 134.5; 145.9; 149.8; 164.8; 172.7.

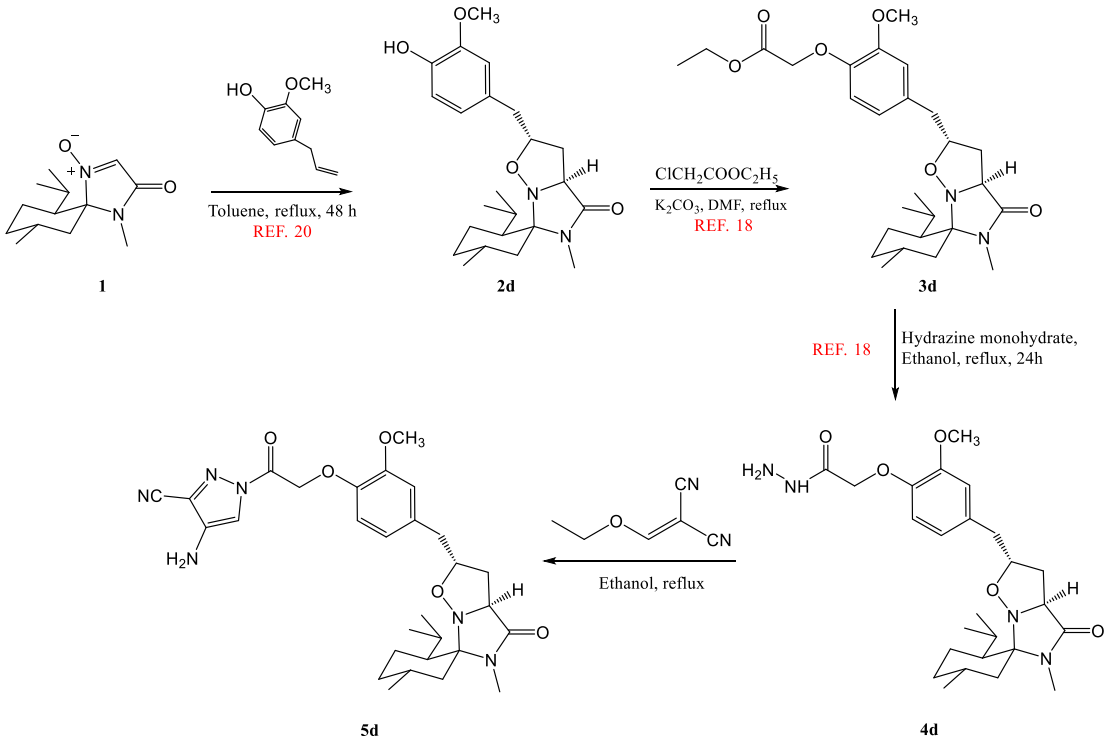
3. RESULTS AND DISCUSSION

The synthesis begins with the 1,3-dipolar cycloaddition between chiral nitrone and a series of for terminal alkenes as described by Alminderej et al [20]. The stereochemistry of cycloadducts has already been discussed by Alminderej et al [20]. The obtained cycloadducts undergo an alkylation reaction in the presence of chloromethyl ethyl ester followed by condensation with hydrazine monohydrate to give hydrazides **4a-c** [18]. The desired products **5a-c** were prepared by reaction of intermediates **4a-c** with ethoxymethylene-malononitrile (scheme 1).



Scheme 1. Synthesis of compounds **5a-c**

The same procedure was applied to 4-allyl-2-methoxy phenol to access isoxazolidine-pyrazole hybrid **5d** (scheme 2).



Scheme 2. Synthesis of compounds **5d**

The 1H NMR spectra of the isoxazolidine-pyrazole hybrids **5a-d** showed a singlet around 9.68 ppm attributed to the $C_{sp2}H$ proton of the pyrazole ring. The 13C nmr spectra of the isoxazolidine-pyrazole hybrids **5a-d** revealed two peaks around 164.8 ppm and 172.7 ppm attributed to the two carbonyls and a peak around 116.9 ppm corresponds to carbonitrile (CN). IR reveals a thin band of average intensity around 2252 cm^{-1} corresponding to the C≡N group and a band around 3241cm^{-1} attributed to the NH_2 group.

ADMET and pharmacokinetic analysis

Prediction of the properties of absorption, distribution, metabolism, excretion, and toxicity (ADMET) from a compound is an essential and pivotal step in the pharmaceutical stage of drug development because several dugs fail due to their poor pharmacokinetics and toxicity problems [21-24]. Thereby, to avoid wasting time, in silico ADMET methods are the first step in this pipeline process to predict pharmacokinetics features. All the tested molecules show negative solubility in in accordance with the limits and potent intestinal absorption (90%). Caco-2 permeability revealed that only compounds **5a** and **5b** will be easily absorbed and better than those of **5c** and **5d** respectively. The likelihood of these compounds to be skin permeable have been explored and revealed moderate values. To characterize their distribution, volume of distributions (VDss (human)), fraction unbound (human), blood-brain barrier (BBB) and CNS permeabilities were analyzed. All molecules are predicted to have moderate and negative blood-brain barrier (BBB) permeability values, justifying their penetration through BBB. Low volume of distributions was found for **5c** and **5d** and moderate for the rest of molecules justifying their ability to be circulated at an equal level of blood plasma and to be distributed to tissue rather than plasma. Regarding the CNS permeability values. Results show that **5a** and **5b** were predicted to be moderately penetrate the CNS, unlikely those of **5c** and **5d** which are unable to penetrate the (logPS is < -3). For metabolism parameters, the possibility of these compounds to be metabolized in the liver show variable and exciting results based on s predicted based on the CYP models (CYP2D6, CYP3A4, CYP1A2, CYP2C19, CYP2C9, CYP2D6, and CYP3A4), with the most relevant are CYP2D6 and CYP3A4. From Table 1, it can be seen that all the title compounds were substrate for CYP3A4, not substrate for CYP2D6, not inhibitor of CYP1A2 and CYP2D6, and are inhibitor of CYP3A4 inhibitors. Excretion behavior of our compounds was estimated based on the total clearance model (rate of drug elimination divided by its plasma concentration) and renal OCT2 substrate and were found to be not affect OCT2 substrates revealing their potential contraindications and clearance. According to the summarized toxicity results (Table 1), the selected compounds show no AMES toxicity, no hERG 1 inhibition properties as well as no skin sensitization.

Table 1. ADMET properties of compounds the synthesized compounds using pkCSM.

Entry	5a	5b	5c	5d	Reference
Absorption					
Water solubility	-5.132	-4.944	-5.097	-4.471	-
Caco2 permeability	1.156	1.092	0.165	0.71	>0.9
Intestinal absorption (human)	92.127	91.787	90.987	97.039	<30% is poorly
Skin Permeability (log Kp)	-2.921	-2.916	-2.847	-2.799	>-2.5 is low
Distribution					
VDss (human)	-0.009	-0.063	-0.16	-0.204	Low is <-0.15, High is >0.45
Fraction unbound (human)	0.017	0.012	0	0	-
BBB permeability	-0.676	-0.662	-0.841	-0.785	Poorly is <-1, High is >0.3
CNS permeability	-2.563	-2.634	-3.28	-3.475	Penetrate is >-2, Unable is <-3
Metabolism					
CYP2D6 substrate	No	No	No	No	No
CYP3A4 substrate	Yes	Yes	Yes	Yes	-
CYP1A2 inhibitor	No	No	No	No	No
CYP2C19 inhibitor	No	Yes	No	No	No
CYP2C9 inhibitor	Yes	Yes	Yes	Yes	No
CYP2D6 inhibitor	No	No	No	No	No
CYP3A4 inhibitor	Yes	Yes	Yes	Yes	No
Excretion					
Total Clearance	0.291	0.344	0.217	0.262	-
Renal OCT2 substrate	No	No	No	No	-
Toxicity					
AMES toxicity	No	No	No	No	No
Max. tolerated dose (human)	-0.742	-0.884	-0.915	-0.459	Low is ≤ 0.477, High is > 0.477
hERG I inhibitor	No	No	No	No	No
Oral Rat Acute Toxicity (LD50)	2.515	2.464	2.458	2.173	-
Oral Rat Chronic Toxicity	1.445	1.454	1.442	1.84	-
Skin Sensitisation	No	No	No	No	No

4. Conclusion

In summary, four isoxazolidine-pyrazole hybrids were synthesized and characterized. The ADMET study and pharmacokinetic analysis showed that the title compounds have good ADMET properties, which are essential for a molecule to exhibit acceptable pharmacokinetic and pharmacokinetic properties.

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الملخص باللغة العربية

اصطناع وتحليل ADMET والتحليل الدوائي الحركي لهجينات جديدة من إيزوكسازوليدين وبيرازول

سوار الغناي
المملكة العربية السعودية، القصيم، بريدة، جامعة القصيم، كلية العلوم، قسم الكيمياء

يستمر الاهتمام بالمركبات الهجينة التي تحتوي على حلقتين مختلفتين أو أكثر في النمو بسبب النتائج البيولوجية الرائعة التي تم التوصل إليها في العديد من الأمثلة. في هذا العمل، قمنا بتصنيع هجينات جديدة من إيزوكسازوليدين-بيرازول ودراسة تحليل ADMET والدوائية الحركية. أوضحت هذه النتائج أن المركبات التي تحمل عنوانًا لها خصائص ADMET جيدة، وهي شرط أساسي لإظهار الجزيء لخصائص دوائية مقبولة.

الكلمات المفتاحية: تحليل الحركية الدوائية، ADMET، إيزوكسازوليدين، إضافة حلقية 1،3-ثنائية القطب، بيرازول