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Prediction of the most probable biological activities for imidazo[2,1-c][1,2,4]triazine derivatives

Imidazotriazines display a wide spectrum of biological activity. Till now all known imidazo[2,1-c][1,2,4]triazines have been prepared and tested as cardiovascular agents (6), bactericides (5), central nervous system stimulants (1). The other derivatives of this ring system have been received as the Maillard reaction inhibitors. Nowadays it is considered that the Maillard reaction takes part in various diseases relating to diabetes and ageing. Some of imidazo[2,1-c]triazines described in the literature have been synthesized for the treatment and /or prevention of various diabetes complications such as coronary disease, periphery circulatory disorder, renal disease, cerebrovascular disorder, diabetic neurosis, retinities, articular sclerosis or the diseases caused by ageing such as senile cataract, atherosclerosis etc. by inhibiting the Maillard reaction (4).

Imidazo[2,1-c][1,2,4]triazines reported herein contain in their chemical structure certain similar features, (potential pharmacophore formations: the phenyl ring, the additional carbonyl group as the potential acceptor centre of hydrogen bond) to many morphine-like analgesics such as fentanyl, petidine, benzitramide and selective ligand of d-opioid receptors (SNC-80). These similar features, according to pharmacophore model introduced by Beckett with its further modifications, can play an important role in expressing pharmacological activity, especially the analgesic action. The presence of carbonyl group in the structure of the obtained compounds can probably play a supporting role in binding with receptor due to the high negative potential present on the oxygen atom. On the other hand, the obtained imidazo[2,1-c][1,2,4]triazines have no basic nitrogen atom. The lack of this atom could play a role in the receptor activation stage and was also observed in the first selective and very potent kappa opioid peptide receptor agonist – salvinorin A, the main active ingredient of Salvia divinorum, which has no action at the 5-HT_2 receptors, the principle molecular target responsible for the actions of classical for opioids side effects (3).

The potential pharmacological activity of reported imidazotriazoles was confirmed based on experimental behavioural animal tests conducted in the Department of Pharmacodynamics of Medical University of Lublin. Some of these compounds exhibited significant antinociceptive activity as the result of the writhing and hot plate tests indicated. Their activity was correlated with relatively low acute toxicity (LD_{50} values ranging from 1100 to over 2000 mg/kg b.w. via i.p.) (7).

The following compounds obtained due to the reaction of 1-arylimidazolidin-2-one hydrazones by cyclization reaction with ethyl oxalate were evaluated for the most possible biological activities using computer PASS programme:

I. 8-phenyl-3,4-dioxo-2H,8H-6,7-dihydroimidazo[2,1-c][1,2,4]triazine;
II. 8-(2-methylphenyl)-3,4-dioxo-2H,8H-6,7-dihydroimidazo[2,1-c][1,2,4]triazine;
III. 8-(4-methylphenyl)-3,4-dioxo-2H,8H-6,7-dihydroimidazo[2,1-c][1,2,4]triazine;
IV. 8-(2-methoxyphenyl)-3,4-dioxo-2H,8H-6,7-dihydroimidazo[2,1-c][1,2,4]triazine;
V. 8-(4-methoxyphenyl)-3,4-dioxo-2H,8H-6,7-dihydroimidazo[2,1-c][1,2,4]triazine;
VI. 8-(3-chlorophenyl)-3,4-dioxo-2H,8H-6,7-dihydroimidazo[2,1-c][1,2,4]triazine;
VII. 8-(4-chlorophenyl)-3-hydroxy-6,7-dihydro-4H-imidazo[2,1-c][1,2,4]triazin-4-one;
VIII. 8-(3,4-dichlorophenyl)-3,4-dioxo-2H,8H-6,7-dihydroimidazo[2,1-c][1,2,4]triazine.

The structure of the above mentioned compounds was confirmed by elemental analysis and spectral data: nuclear magnetic resonance (^{1}H NMR, ^{13}C NMR) and mass spectra (MS). The purity of all the compounds synthesized was checked by TLC. Thin-layer chromatography was carried out on commercial Merck SiO_2 60 F_{254} plates having fluorescence indicator; the spots were visualized with UV light ë = 254 nm. These compounds were characterized by solubility in dimethylformamide and dimethylsulfoxide. These compounds showed antinociceptive activity in animal behavioral tests and relatively low acute toxicity (LD_{50} value in range from 1100 to over 2000 mg/kg b.w. via i.p.) (7).

MATERIAL AND METHODS

Prediction of potential biological activity. Potential biological properties of the investigated imidazo[2,1-c][1,2,4]triazines were evaluated using computer programme PASS (Prediction of Activity Spectra for Substances); http://www.ibmh.msk.su/service.htm. This programme predicts the biological activity spectrum for a compound on the basis of its structural formula. It estimates the probability for the molecule to be active (Pa) and inactive (Pi) for each type of activity from the biological activity spectrum. The most probable biological activities of above mentioned molecules predicted by PASS programme are shown in Table 1.

Table 1. The most probable types of biological activity of imidazo[2,1-c][1,2,4]triazines predicted by PASS programme and those comparison with experimental data

Comp.	Pa	Pi	Predicted activity	Experimental data [a]
I	96.4	0.1	GABA A receptor antagonist	antinociception in a dose
	94.7	0.1	Antineurogenic pain	140 mg/kg (0.1 LD_{50});
	85.9	0.4	Cyclic GMP phosphodiesterase inhibitor	reversion of antinociception by
	84.1	0.7	Analgesic, non opioid	5 mg/kg dose of naloxone
	81.6	0.2	GABA receptor antagonist	
II	94.7	0.1	GABA A receptor antagonist	not tested
	92.0	0.2	Antineurogenic pain	
	81.1	0.5	Cyclic GMP phosphodiesterase inhibitor	
	77.6	0.3	GABA receptor antagonist	
	73.2	1.2	Analgesic, non opioid	
III	95.1	0.1	GABA A receptor antagonist	antinociception in a dose
	92.6	0.2	Antineurogenic pain	180 mg/kg (0.1 LD_{50});
	81.6	0.5	Cyclic GMP phosphodiesterase inhibitor	
	79.4	0.8	Analgesic, non opioid	
	78.5	0.3	GABA receptor antagonist	
IV	94.4	0.1	GABA A receptor antagonist	antinociception in a dose
	91.0	0.2	Antineurogenic pain	110 mg/kg (0.1 LD_{50})
	83.9	0.4	Cyclic GMP phosphodiesterase inhibitor	
	76.3	0.3	GABA receptor antagonist	
	76.1	0.9	Analgesic, non opioid	

V	94.9 91.6 82.2 77.5 77.3	0.1 0.2 0.4 0.3 0.9	GABA A receptor antagonist Antineurogenic pain Cyclic GMP phosphodiesterase inhibitor GABA receptor antagonist Analgesic, non opioid	significant antinociception in doses 37.5-150 mg/kg (0.025-01 LD_{50})
VI	95.1 91.4 82.6 82.2 79.4	0.1 0.2 0.4 0.7 0.3	GABA A receptor antagonist Antineurogenic pain Cyclic GMP phosphodiesterase inhibitor Analgesic, non opioid GABA receptor antagonist	strong antinociception in doses 12.5-200 mg/kg (0.00625-0.1LD_{50}); reversion of antinociception by 5mg kg dose of naloxone
VII	96.7 84.8 81.4	0.1 0.2 0.5	GABA A receptor antagonist GABA receptor antagonist Cyclic GMP phosphodiesterase inhibitor	lack of antinociceptive effect, reduction of number "head twitch episodes" after 5HTP administration
VIII	95.2 92.2 83.2 82.3 80.0	0.1 0.2 0.7 0.4 0.3	GABA A receptor antagonist Antineurogenic pain Analgesic, non opioid Cyclic GMP phosphodiesterase inhibitor GABA receptor antagonist	not tested

Data concerning antinociceptive activity from ref. 7

RESULTS AND DISCUSSION

In connection with proved significant activity on the central nervous system of the above mentioned compounds, it seemed worthwhile to predict the most possible biological activity spectrum for them. Based on PASS programme for all compounds the most probable it seemed to be GABA A receptor antagonistic activity (Pa about 95.3%, Pi 0.1%). Also, the inhibition of cyclic guanosine monophosphate (cGMP) was predicted as highly possible (Pa in the range 81.1–85.9%) for them. From the literature data it follows that imidazo[5,1-f][1,2,4]triazin-4(3H)-ones are highly cGMP- PDE-5 selective and show IC_{50} values for PDE-3 and PDE-4 greater than 50 nM. In comparison to well known pyrazolo[4,3-d]pyrimidin-7-one phosphodiesterase inhibitors (for instance Sildenafil) the imidazotriazines reported in the literature indicate improved selectivity over PDE-1 and substantially improved PDE-5 inhibition (2). Also substituted purinones as analogues of cGMP are potent phosphodiesterase (PDE) inhibitors *in vitro*. For seven compounds, except compound VII, analgesic activity was found as high possible with Pa>70 %. According to PASS programme the probability for compounds to be analgesic is in the order: I>VIII>VI>III>V>IV>II. For compound VI i. e. 8-(3-chlorophenyl-3,4-dioxo-2H,8H-6,7-dihydroimidazo[2,1-c][1,2,4]triazine the probability to be analgesic (82.2%) is greater than for compound V i. e. 8-(4-methoxyphenyl)-3,4-dioxo-2H,8H-6,7-dihydroimidazo[2,1-c][1,2,4]triazine (77.3%). It is in agreement with the experimental data: compound VI revealed strong antinociception in doses 12.5-200 mg/kg (0.00625-0.1 LD_{50}), whereas compound V caused significant antinociception in doses 37.5-150 mg/kg (0.025-0.1 LD_{50}) (7). The probability for compound VII to be analgesic was not predicted by PASS programme. Also, lack of analgesic effect was observed in experimental studies. In this case the inactivity of this compound could be explained on the basis of the molecular modelling results (this compound exists predominantly in the 3-hydroxy form, all other in 3-oxo one (7). Compounds VIII and II also indicated Pa>70% in the category of analgesic with values 83.2 and 73.2%, respectively. Therefore, these compounds will be tested in the future for their potential analgesic activity.

The PASS programme may be useful in the prediction of the most possible activity spectrum for potential bioactive molecules and lead to limit the organic syntheses. It may also be used for a comparison of the theoretical and experimental data results like it has been shown in this case.

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SUMMARY

The purpose of this study was to evaluate the most possible biological activities of certain analgesic active imidazo[2,1-c][1,2,4]triazines, using computer PASS programme. These compounds contain in their chemical structure potential pharmacophore formations and features similar to those present in morphine-like analgesics and opioid receptor agonists with no basic nitrogen atom, showing no typical opioid side effects. In experimental trials these compounds showed significant antinociceptive activity on the central nervous system of the tested animals, correlated with relatively low acute toxicity values (LD_{50} ranging from 1100 to above 2000 mg/kg b.w. via i. p.). The most possible biological activities of investigated imidazo[2,1-c][1,2,4]triazines were predicted by PASS programme. For all the compounds the most possible it seemed to be GABA A receptor antagonistic activity, inhibition of cGMP phosphodiesterase and analgesic activity.

Określenie najbardziej prawdopodobnych typów aktywności imidazo[2,1-c][1,2,4]triazyn

Celem pracy jest określenie najbardziej prawdopodobnych typów aktywności imidazo[2,1-c][1,2.4]triazyn, przy zastosowaniu programu komputerowego PASS. W strukturze otrzymanych związków występują potencjalne ugrupowania farmakoforowe analogiczne do występujących w narkotycznych lekach przeciwbólowych, a także agonistach receptora opioidowego bez zasadowego atomu azotu, pozbawionych typowych objawów ubocznych. W badaniach eksperymentalnych związki te wykazały znaczącą aktywność antynocyceptywną w ośrodkowym układzie nerwowym i niską toksyczność (wartość LD_{50} wynosząca od 1100 do 2000 mg/kg m.c przy podaniu dootrzewnowym). Określono najbardziej prawdopodobne typy aktywności pochodnych imidazo[2,1-c][1,2,4]triazyny przy zastosowaniu programu komputerowego PASS. Najbardziej prawdopodobne spektrum aktywności badanych związków obejmuje: działanie jako antagoniści receptora GABA, hamowanie fosfodiesterazy cGMP, działanie przeciwbólowe.