BIOMEDICAL AND BIOSOCIAL ANTHROPOLOGY

Official Journal of the International Academy
of Integrative Anthropology

journal homepage: <http://bba-journal.com>

Investigation of pharmacokinetics of N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide various crystalline modifications in vivo

Bondarenko P. S.¹, Voloshchuk N. I.¹, Larionov V. B.², Fedoseenko H. O.³

¹National Pirogov Memorial Medical University, Vinnytsya, Ukraine

²A.V. Bogatsky Physico-Chemical Institute National Academy of Sciences of Ukraine, Odesa, Ukraine

³ALC "Interhim", Odesa, Ukraine

ARTICLE INFO

Received: 9 November 2020

Accepted: 10 December 2020

UDC: 615.014:615.451.35:615.015

CORRESPONDING AUTHOR

e-mail: voloshchuk@vnmu.edu.ua
Voloshchuk N. I.

The phenomenon of polymorphism, inherent in many biologically active substances, including drugs, is an extremely serious problem that requires close attention and comprehensive study. Various crystalline modifications of biologically active compounds, as well as excipients can dramatically change the biopharmaceutical properties and pharmacological characteristics of already known drugs. The bioactive compound N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide was obtained by bioisosteric substitution in the molecule of known non-steroidal anti-inflammatory drugs of the oxicam group in the form of three polymorphic modifications of the crystal. sticks (form A), plates (form B) and blocks (form C) with varying degrees of analgesic and anti-inflammatory effects. The aim of the study was to investigate the pharmacokinetic profile of these polymorphic forms of benzothiazine-3-carboxamide derivative by their oral administration in vivo. The concentration of compounds in the blood of mice was determined by HPLC and the main pharmacokinetic parameters were calculated. It was found that the polymorphic form in the form of plates (form B), in particular, having the largest surface area of crystals, has differences mainly at the stage of absorption with corresponding changes in absorption values, maximum concentration and time of its achievement, has the highest bioavailability and rapid elimination which correlates with the most optimal pharmacological and toxicological characteristics of this compound in comparison with other crystalline forms. This makes it possible to recommend the most active and safe polymorphic form of the compound (form B) for in-depth preclinical study and possible clinical trials as an analgesic and anti-inflammatory agent.

Keywords: N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide, polymorphic modifications, crystal habitus, parameters of pharmacokinetic.

Introduction

The phenomenon of polymorphism among biologically active substances occupies one of the leading places in the process of drug development and pharmaceutical development of the dosage form. To date, the phenomenon of polymorphism has been detected for more than 85 % of pharmaceutical substances and the process of relevant research continues [9]. The accumulation of experimental material and clinical experience has formed a clear understanding that polymorphism for pharmacy and medicine is an extremely serious and important problem that requires close attention and comprehensive study.

Fig. 1. Chemical formula of N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide.

Fig. 2. Crystalline forms of N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide, derived from ethyl acetate (A), meta-xylene (B) and ortho-xylene (C).

Various crystalline modifications of biologically active compounds, as well as their excipients can dramatically change (and not always for the better) technological, pharmaceutical and pharmacological characteristics of already known drugs [3, 7, 8, 15]. It is for these reasons that the phenomenon of pharmaceutical polymorphism is being actively studied and strictly controlled in many countries around the world [14].

The objects of the study were compounds - derivatives of 4-R-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxylic acids, synthesized by bioisosteric substitution in the molecule of oxicams at the Department of Pharmaceutical Chemistry of the National University of Pharmacy under the leadership of Professor Ukrayinets I. V. Preliminary results [16] proved the presence of derivatives of this chemical class of high analgesic activity and established a leader compound, namely N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide (Fig. 1).

In the process of chemical synthesis of this compound by crystallization from generally acceptable organic solvents (ethyl acetate, meta-xylene and ortho-xylene) three externally completely different types of completely colorless crystals were obtained - long sticks with a square cross section (form A), thin chisel-shaped plates (form B) or elongated blocks without a clear geometric shape (shape C), respectively (Fig. 2).

Detailed studies of their antinociceptive effects in five models of pain perception have shown that, despite their chemical identity, they differ significantly in the strength of the analgesic effect, as well as acute toxicity and gastric damage [19]. Therefore, the question arose to establish the reasons that may explain the existence of these differences.

The aim of the study was to investigate the features of the pharmacokinetic behavior of three crystalline modifications of the benzothiazine-3-carboxamide derivative by their oral administration in vivo.

Materials and methods

The study was performed on 156 white nonlinear male mice kept on a standard vivarium diet, access to water *ad*

libitum and 12/12 day/night. All studies were performed in accordance with the requirements of the Helsinki Declaration on the Humane Treatment of Laboratory Animals and the current laws of Ukraine.

Determination of the content of the test compound was performed on a combined HPLC-MS system - liquid chromatograph 1260 Infinity with a detector 6530 Accurate Mass Q-TOF, "Agilent Technologies". The test compounds were administered orally at a dose of 10 mg/kg as a suspension stabilized with methylcellulose (0.5 %). At certain intervals after administration (0.25, 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 4.0, 5.0, 6.0, 8.0, 10.0, 12.0 h), the animals were subjected to chloroform anesthesia and decapitated by collecting blood in centrifuge tubes. Blood samples were kept for 0.5 h at 37 °C, blood serum was separated by centrifugation (15 min, 6000 rpm). To 200 µl of serum was added 300 µl of a mixture of 7 % perchloric acid - acetonitrile (60:40) to precipitate proteins, stirred and centrifuged (8000 rpm, 5 °C, 20 min). The supernatant was filtered through a microporous filter (20 µm), the bridge of the test substance was determined by HPLC under these conditions with a sample injection volume of 20 µl. The calculation of the concentration of the compound in the serum (C, µg/cm³) was carried out according to the formula:

$$C = Q \cdot ((V_1 + V_2) \cdot 1000) / (V_1 \cdot V_3),$$

where: Q is the amount of compound determined in the injection sample according to the calibration schedule; V₁ - serum volume, µl; V₂ - volume of perchloric acid solution with acetonitrile V₃ - volume of injection, µl; 1000 - conversion factor per 1 cm³ [6, 17].

Statistical processing of the obtained results was performed by non-parametric methods using the software package STATISTICA 10.0 with the calculation of the mean value, standard error of the mean. Differences were considered plausible in the case of p < 0.05.

Results

After oral administration of a benzothiazine-3-carboxamide derivative in the form of various polymorphic

Fig. 3. Concentration profile of N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide (A form) after its oral administration (10 mg/kg, $M \pm m$, $n=4$).

Fig. 4. Concentration profile of N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide (B form) after its oral administration (10 mg/kg, $M \pm m$, $n=4$).

Fig. 5. Concentration profile of N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide (C form) after its oral administration (10 mg/kg, $M \pm m$, $n=4$).

forms, its content in blood plasma is determined in rather high concentrations, and the concentration profile corresponds to the distribution kinetics according to the single-chamber model with absorption. It should be noted, however, that the type of concentration curves of the content of compounds A, B and C after administration in the form of polymorphic forms, are quite different both in profile and in the values of certain concentrations (Fig. 3-5). Thus, the introduction of polymorphic form B leads to higher concentrations of it in blood plasma (see Fig. 4) in

comparison with the introduction of polymorphic form A (see Fig. 3). Also for polymorphic form B there is a narrower peak of reaching the maximum concentration with, respectively, higher levels of the compound in the blood plasma than for polymorphic form A. Regarding form C, its concentration profile (see Fig. 5) shows intermediate indicators, which resemble those that can be obtained by introducing a mixture of polymorphic forms A and B and inherent in the additivity of the processes of mass transfer of the compound in the body.

To quantify the pharmacokinetic processes of polymorphic forms, the corresponding pharmacokinetic parameters for polymorphic forms A, B and C were calculated after their single oral administration (Table 1).

It was found that the pharmacokinetic parameters that characterize the processes of distribution of the compound are quite similar for all three forms (see Table 1). The constant volume of distribution (V_{dss}) for polymorphic forms did not have a statistically significant difference, although the difference between form B and polymorphs A and C at the trend level was recorded. This demonstrates the lack of influence of polymorphic modification on the processes of compound distribution in the body.

The magnitude of the plasma elimination constant is usually due to the similarity of the physicochemical properties of the compound itself (water solubility, reactivity, binding to plasma proteins, etc.). Our results showed that the elimination constant for polymorphic form B when administered orally was 1.48 times higher compared to form A and C, and the half-life of form B was 34.7-31.6 % less than other polymorphic forms ($p < 0.05$), which indicates about the higher rate of excretion of compound B from the body. These values are lower than those for the compound when administered intravenously, which was $0.511 \pm 0.068 \text{ h}^{-1}$ (data not published), but the difference is not statistically significant and is probably due to the lack of absorption during intravenous administration. Another factor that leads to a partial decrease in the rate of elimination of the compound may also be a combination of active transport and passive diffusion in the overall elimination process, so that at high plasma concentrations the studied compound is excreted by both mechanisms, while at lower concentrations achieved after oral administration, a significant contribution is due only to active transport. Accordingly, the values of the half-life ($T_{el}^{1/2}$) of the compound from the body are also close.

The amount of total clearance, which depends on the volume of distribution and elimination of compounds in compound B, was also smaller compared to the other two forms (18-25 % on average), but this difference also did not reach probable values.

At the same time, it should be noted that the process of absorption of polymorphic forms is significantly different, which is reflected in the indicators of the corresponding pharmacokinetic parameters. First of all, it is noticeable (see Table 1) that the process of absorption of N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide in polymorphic form B occurs

Table 1. Pharmacokinetic parameters of N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide after its oral administration in the form of polymorphs A, B and C (10 mg/kg, M±m, n=4).

Pharmacokinetic parameter	A form	B form	C form
Absorption constant, k_{abs} , h^{-1}	0.669±0.067	0.750±0.077*/**	0.624±0.081
Elimination constant, k_{el} , h^{-1}	0.270±0.018	0.401±0.024*/**	0.274±0.017
Constant volume of distribution, V_{dss} , l/kg	6.000±1.310	4.610±0.476	5.612±1.421
Maximum concentration, C_{max} , mcg/ml	671.4±21.8	809.5±71.4*	786.1±10.1
Time to reach maximum concentration, T_{max} , h	3.000	1.500	2.510
Area under the pharmacokinetic curve, $AUC_{0-\infty}$, mcg·h/ml	4687±798	4281±586	5308±720
The area under the first moment of the pharmacokinetic curve, $AUMC_{0-\infty}$, mcg·h ² /ml	35529±8882	18495±1755	40152±3989
The average retention time in the body, MRT, h	7.581±0.286	4.322±0.719*	7.376±2.076
Total clearance, Cl_{tot} , l/kg·h	1.611±0.019	1.900±0.110	1.511±0.100
Semi-elimination time, $T_{el}^{1/2}$, h	2.621±0.210	1.712±0.119*/**	2.502±0.200
Half-absorption time, $T_{abs}^{1/2}$, h	1.042±0.112	0.919±0.093	1.110±0.142
Bioavailability, f	0.719± 0.063	0.810±0.178*/**	0.122±0.013

Notes: * - statistically significant differences ($p < 0.05$) to the form A; ** - statistically significant differences ($p < 0.05$) relative to form C.

with a much larger rate than in the form of polymorphic form A, because the absorption constant of form B statistically exceeds the same rate of compound A by 12.1 %, and compound C - by 20.2 %. Accordingly, the average half-absorption time ($T_{abs}^{1/2}$) is on average 11.5 % lower for polymorphic form B compared to the other two crystal forms. Shorter absorption time for form B causes a total shorter residence time of this compound in the body (by 43.0 %) compared to forms A and C. The rate of absorption and the ratio of absorption and elimination constants are also known to affect the value (C_{max} , µg/ml) and time (T_{max} , h) to reach maximum concentration. Polymorphic form B of the benzothiazine-3-carboxamide derivative shows a higher maximum concentration, which is 20.6 % higher than form A ($p < 0.05$) and unreliable ($p > 0.05$) of compound C, and the time to reach the maximum concentration of compound B was twice as fast as in form A (see table. 1).

Due to the higher rate of absorption of the compound N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide in the form of polymorphic form B, significantly increases the amount of substance entering the internal environment of the body - the integral value of the amount of compound entering the blood (area under the pharmacokinetic curve "concentration-time", $AUMC_{0-\infty}$) is 4281±586 µg·h²/ml, while for form A - 4687±798 µg·h²/ml.

The higher absorption rate also causes the expected increased bioavailability (f) for polymorphic form B - indeed, for form B this indicator was significantly higher by 12.5 and 24.6 % than for polymorphic forms A and C. This indicates that form B has greater bioavailability. due to the higher degree of absorption.

Discussion

Inherent in many substances, including biologically active, polymorphism is an extremely serious problem. The

phenomenon of polymorphism is explained by the fact that the same atoms of matter can create different stable crystal lattices. This phenomenon was first described in 1788 by the German chemist Claport M.G. In 1942, more than 1,200 organic polymorphic compounds were known, now there are more than 10,000. Polymorphism of the active pharmaceutical substance (APS) can cause pharmaceutical inequality and, as a consequence, pharmacokinetic and therapeutic non-equivalence of drugs in solid dosage form. There are significant differences in the solubility of polymorphic modifications of APS can lead to differences in the kinetics of their dissolution in vivo and, as a consequence, the bioavailability of drugs. The bioavailability / bioequivalence of drugs depends on a number of factors that determine the rate and degree of their absorption, such as dissolution kinetics and penetration through cell membranes, gastrointestinal motility, metabolism. These factors are taken into account in the concept of biopharmaceutical classification system (BCS) APS, which is already accepted in the manuals of regulators in industrialized countries [1].

At present, polymorphism is found in more than 70 % of drugs in almost all pharmacological groups. It is an extremely important factor that modifies the therapeutic properties of pharmaceutical substances and dosage forms, significantly affects the parameters of their biological activity, as well as pharmacokinetics. Polymorphic changes in drugs can cause rapid inactivation of drugs, changes in the physical parameters of finished drugs, chemical incompatibility of ingredients in the same dosage form. By changing the pharmacokinetics of the drug, the polymorphism may affect its toxicity. Pharmaceutical substances have the ability to form polymorphic modifications that differ from each other in crystallographic parameters, physicochemical properties and the like.

Different in strength and toxicity, different polymorphs and stereoisomers are able to form antibacterial drugs (chloramphenicol, ampicillin, tetracycline), local anesthetics and analgesics (ibuprofen, ketoprofen), hormonal and cardiovascular drugs methylprednisolone, insulin, progesterone [2, 4, 9, 10, 11, 20].

However, in recent years, no less important characteristic that significantly affects the properties of biologically active substances and drugs, their habit (appearance) - needles, plates, cubes, prisms, shapeless crystals and others. Samples of the active substance may differ in the degree of wettability, compressibility, hygroscopicity, stability, solubility and other physicochemical properties, which ultimately affects the quality of the final product. That is why these moments are currently receiving a lot of attention [4, 5, 11, 12, 18, 20]. The crystalline modifications of N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide presented in this work are unique in that they differ only in appearance, whereas the X-ray diffraction analysis showed their identical molecular and crystalline structure. These crystals differ only in some geometric characteristics of the studied molecules - the length of the connections, the angle of deviation of atoms and the total value of the valence angles [19]. Studies of biological activity in different models of pain perception have shown that these compounds differ in both strength and speed of onset of effect. Thus, under the conditions of oral administration in the model of acetic acid cramps in mice and thermal irritation of the tail (tail flick and tail emersion), the most powerful effect was shown by compound B, while compound A was significantly inferior in strength and compound C was intermediate. An in-depth study of the rate of onset and duration of analgesic effect, which was studied in a model of electrical irritation, showed that the effect of compound B occurs most rapidly, reaches a maximum at 2 hours, but then its effect gradually decreases. On the other hand, compound A, acts slowly with a maximum at the 4th hour, does not reach the level of compound B by force, but its duration of action is much longer than the last one. Based on the working hypothesis that such a difference is due to the different rate of absorption in the gastrointestinal tract and greater bioavailability of crystalline form B compared to the other two - was fully confirmed in this work. Given the route of administration of the compound, as well as the fact that it was administered as polymorphs and under conditions that preserve the crystal lattice, it can be assumed that the type of polymorph and its effect on the biopharmaceutical

phase (dissolution after oral administration and subsequent absorption in the gastrointestinal tract) cause a difference in pharmacokinetic parameters and, accordingly, in the manifestation of pharmacological effects. Given that these polymorphs are obtained by crystallization from different solvents, it can be assumed that the crystalline structures that are formed are subject to varying degrees to wetting with hydrophilic aqueous medium and the formation of hydrogen connections with the solvent, which precedes their subsequent dissolution. Accordingly, crystalline polymorphic form B, which dissolves more rapidly in the gastrointestinal tract, is absorbed at a higher rate (and degree of absorption) in the intestine and has a pharmacological effect. Toxicological studies (acute lethality and ulcerogenic action) have shown that the safest is the polymorphic modification B, ie crystals in the form of plates [19], which serves as a basis for further in-depth studies of this modification N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide.

Further research in this direction will serve to develop optimal conditions for the development and creation of new biologically active compounds and generic substances to optimize the pharmacodynamic and toxicological parameters of drugs.

Conclusion

1. The results of the study expand the understanding of the role of spatial structure in the manifestations of pharmacological activity and safety of drugs.

2. Based on the analysis of pharmacokinetic parameters of polymorphic forms of N-(4-trifluoromethylphenyl)-4-methyl-2,2-dioxo-1H-2λ6.1-benzothiazine-3-carboxamide, it can be concluded that the polymorphic form in the form of plates (form B), having the largest surface area of the crystal, affects mainly the absorption processes with corresponding changes in the values of absorption, maximum concentration and time of its achievement, has the highest bioavailability and rapid elimination, which correlates with the most optimal pharmacological and toxicological characteristics of this compound compared to other crystalline forms.

3. The results of an in-depth study of the pharmacodynamics and pharmacokinetics of compounds with different crystalline modifications will make it possible to recommend the most active and safe compound for in-depth preclinical study and possible clinical trials as an analgesic and anti-inflammatory agent.

References

- [1] Ahr, G., Voith, B., & Kuhlmann, J. (2000). Guidances related to bioavailability and bioequivalence: European industry perspective. *European journal of drug metabolism and pharmacokinetics*, 25(1), 25-27. doi: 10.1007/BF03190052
- [2] Babilev, F. V., & Andronic, I. Ya. (1981). *Полиморфизм лекарственных веществ [Polymorphism of drug substances]*. Кишинев: Штиинца - Chisinau: Shtiintsa.
- [3] Bernstein, J. (2011). Polymorphism - A perspective. *Crystal Growth & Design*, 11(3), 632-650. doi: 10.1021/cg1013335
- [4] Bukovec, P., Benkić, P., Smrkolj, M., & Vrećer, F. (2016). Effect of crystal habit on the dissolution behavior of simvastatin crystals and its relationship to crystallization solvent properties. *Pharmazie-An International Journal of Pharmaceutical Sciences*, 71(5), 263-268. PMID: 27348970

- [5] Chernyshev, V. V., Morozov, Y. N., Bushmarinov, I. S., Makoed, A. A., & Sergeev, G. B. (2016). New polymorph of dehydroepiandrosterone obtained via criomodification. *Crystal Growth & Design*, 16(2), 1088-1095. doi: 10.1021/acs.cgd.5b01666
- [6] Dhillon, S., & Gill, K. (2006). *Basic Pharmacokinetics*. Clinical Pharmacokinetics, Pharmaceutical Press, London.
- [7] Fandaruff, C., Silva, M. A. S., Bedor, D. C. G., de Santana, D. P., Rocha, H. V. A., Rebuffi, L., ... & Cuffini, S. L. (2015). Correlation between microstructure and bioequivalence in Anti-HIV Drug Efavirenz. *European Journal of Pharmaceutics and Biopharmaceutics*, 91, 52-58. doi: 10.1016/j.ejpb.2015.01.020
- [8] Gelbrich, T., Meischberger, I., & Griesser, U. J. (2015). Two polymorphs of 5-cyclohexyl-5-ethylbarbituric acid and their packing relationships with other barbiturates. *Acta Crystallographica Section C: Structural Chemistry*, 71(3), 204-210. doi: 10.1107/S2053229615002880
- [9] Gildeeva, G. N. (2017). Полиморфизм: влияние на качество лекарственных средств и актуальные методы анализа [Polymorphism: Impact on Drug Quality and Current Analysis Methods]. *Качественная клиническая практика - Good clinical practice*, 1, 56-60.
- [10] Hilfiker, R. (2006). *Polymorphism in pharmaceutical industry*. Weinheim: Wiley-VCH.
- [11] Hooper, D., Clarke, F. C., Docherty, R., Mitchell, J. C., & Snowden, M. J. (2017). Effects of crystal habit on the sticking propensity of Ibuprofen - A case study. *International journal of pharmaceutics*, 531(1), 266-275. doi: 10.1016/j.ijpharm.2017.08.091
- [12] Leuner, C., & Dressman, J. (2000). Improving drug solubility for oral delivery using solid dispersions. *Pharmaceutics and Biopharmaceutics*, 50(1), 47-60. doi: 10.1016/s0939-6411(00)00076-x
- [13] Lutay, M. I., Lysenko, A. F., & Moiseenko, O. I. (2009). Использование оптических изомеров известных сердечно-сосудистых средств - путь к повышению их эффективности и переносимости [The use of optical isomers of known cardiovascular drugs is a way to increase their effectiveness and tolerance]. *Український кардіологічний журнал - Ukrainian Journal of Cardiology*, (4), 13-20.
- [14] Morissette, S. L., Soukasene, S., Levinson, D., Cima, M. J., & Almarsson, O. (2003). Elucidation of crystal form diversity of the HIV protease inhibitor ritonavir by high-throughput crystallization. *Proceedings of the National Academy of Sciences*, 100(5), 2180-2184. doi: 10.1073/pnas.0437744100
- [15] Muresan-Pop, M., Braga, D., Pop, M. M., Borodi, G., Kacso, I., & Maini, L. (2014). Crystal structure and physicochemical characterization of Ambazone monohydrate, anhydrous, and acetate salt solvate. *Journal of pharmaceutical sciences*, 103(11), 3594-3601. doi: 10.1002/jps.24151
- [16] Petrushova, L. A., Ukrainets, I. V., Dzyubenko, S. P., & Grinevich, L. A. (2015). Synthesis and the biological activity of 4-hydroxy-2,2-dioxo-1H-2λ6,1-benzothiazin-3-carboxylic acids trifluoromethyl-substituted anilides. *Journal of Organic and Pharmaceutical Chemistry*, 13(1(49)), 44-48. doi: 10.24959/ophcj.15.826
- [17] Santos, O. M. M., Freitas, J. T. J., Cazedey, E. C. L., Araujo, M. B. D., & Doriguetto, A. C. (2016). Structure, solubility and stability of Orbifloxacin crystal forms: hemihydrate versus anhydrate. *Molecules*, 21(3), 328. doi: 10.3390/molecules21030328
- [18] Stoica, C., Verwer, P., Meekes, H., Van Hoof, P. J. C. M., Kaspersen, F. M., & Vlieg, E. (2004). Understanding the effect of a solvent on the crystal habit. *Crystal growth & design*, 4(4), 765-768. doi: 10.1021/cg0342314
- [19] Ukrainets, I. V., Petrushova, L. A., Fedosov, A. I., Voloshchuk, N. I., Bondarenko, P. S., Shishkina, S. V., ... & Sim, G. (2020). Crystal Habits and Biological Properties of N-(4-Trifluoromethylphenyl)-4-Hydroxy-2, 2-Dioxo-1H-2λ6, 1-Benzothiazine-3-Carboxamide. *Scientia Pharmaceutica*, 88(1), 45-60. doi: 10.3390/scipharm88010001
- [20] Watanabe, A., Yamaoka, Y., & Takada, K. (1982). Crystal habits and dissolution behavior of aspirin. *Chemical and Pharmaceutical Bulletin*, 30(8), 2958-2963. doi: 10.1248/cpb.30.2958