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Molecular Similarity within Carcinogen and Guanine Cyclic Nucleotide Structures

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Abstract

Tumor promoters, apoptosis and autophagy modulators, chemotherapy drugs, and endogenous steroids demonstrate molecular similarity relative to cyclic nucleotide structure. This study explores relative molecular similarity within established human carcinogen structures using computational chemistry software. Molecular structures of conventional carcinogenic drugs and industrial agents demonstrate molecular similarity with a focus on the guanine base and nucleotide cyclized ring. Structures of volatile and gaseous anesthetic carcinogens do not conform to conventional 3-point pharmacophore-based fits characteristic of receptor-binding drugs. The results of this study provide some insight into how carcinogen structures may interact with endogenous compounds to disrupt cyclic nucleotide-driven homeostatic mechanisms.

Keywords

Anesthetics, Cancer, Carcinogens, Guanosine Cyclic Monophosphate, Molecular Similarity

1. Introduction

Purine nucleotides are enzyme substrates and receptor ligands of proteins with a crucial but poorly understood role in the development and regulation of cancer cells.

Guanine and adenine nucleotides participate in cyclical phosphate exchange and nucleotide-cyclic nucleotide transition mechanisms, facilitating cell signal transduction and maintaining energy and ion balances. Deregulation of these mechanisms may initiate carcinogenicity, or homeostatic mechanisms may go awry when cells become cancerous. Down-regulation in cGMP and phosphokinase activity, observed in cancer cell lines, relate to an up-regulation in phosphodiesterase (PDE) activity in cancer patients [1]. Increased synthesis of GTP,

activation of cGMP dependent kinases and promotion of metastases are evident in a mouse model of breast cancer metastasis [2]. The protective effects of PDE inhibitor treatment in breast cancer patients are attributed to the synergistic effects of drugs on initiating apoptosis and decreased chemotherapeutic drug efflux through inhibition of multidrug resistance (MDR) transporters [3]. Several MDR proteins have binding sites for cyclic nucleotides [4].

Mutations in Ras genes are commonly found in tumors in which disruption of GTPase mechanisms may have led to malignancy or cell death [5]. Mutations are also evident in P-class ATPases. Ion-channel transporter proteins, a potential target of chemotherapy, are influenced by tumor promoters [6] [7]. Thapsigargin reduces prostate tumor volume by inhibiting the microsomal Ca^{2+} ATPase [7]. Phorbol myristate acetate (PMA) inhibits ion transport across membranes and binds to guanine nucleotide exchange factors that activate small GTPases [8] [9]. A previous study found no consistent link between the molecular structures of carcinogens and ATPase inhibitors [10]. However, in regard to cyclic nucleotides, a considerable number of compounds of interest in cancer research (chemotherapeutics, apoptosis and autophagy modulators, drug resistance inhibitors) demonstrate relative molecular similarity, including some tumor promoters and probable human carcinogens (PMA, thapsigargin, dichlorvos) [11]. This investigation evaluates molecular similarity within the structures of established human chemical carcinogens and cGMP to further knowledge in this area.

2. Materials and Methods

The agents under investigation are identified as human carcinogens in the National Toxicology Program's 15th Report on Carcinogens 2021 [12]. Compounds are selected on the basis of their different chemical structures listed on the PubChem website (https://pubchem.ncbi.nlm.nih.gov/).

The Nemesis software program (Oxford Molecular version 2.1) is used to build molecular structures from the contents of the program fragment file and minimise structures by conformational analysis. Compound structures used for fitting are minimum energy conformers in an uncharged form. The conformation of the cGMP structure is described by the torsion angle C8N9C1'O9 -33 degrees (see **Figure 2(1)**). The computational program fits paired molecular structures on a three-point basis. Fitting-points, comprised of atoms of similar type and partial charge within compound and nucleotide structures, are identified in the text and table with respect to the nucleotide labels. Colour-coded atoms in the figures identify ligand fitting-points: carbon-green, nitrogen-blue, oxygen-red, and sulphur-yellow. Bond order within the molecular structures is not shown to improve on presentation. The Nemesis program computes goodness-of-fit values, in respect of inter-atomic distance at each fitting-point and root mean square (RMS) value. The sequence of fitting-points used for each carcinogen structure (given in **Table 1**, left to right) provides a fit with the lowest RMS value.

Table 1. Values for fitting carcinogen structures to the cGMP template.

Compound	Fitting-points	Interatomic distances (Å)	RMS (Å)
4-aminodiphenyl	C4O6C2	0.05, 0.10, 0.10	0.0073
4-aminodiphenyl	C4'C3'O8	0.07, 0.06, 0.04	0.0014
arsenic trioxide	O5O7P1	0.17, 0.16, 0.02	0.0041
arsenic trioxide	N1N2N3	0.17, 0.16, 0.17	0.0019
azathioprine	C2N3C8	0.07, 0.11, 0.04	0.0168
azathioprine	O5O3C1'	0.10, 0.08, 0.06	0.0000
bis(chloromethyl)ether	O5P1O3	0.08, 0.13, 0.11	0.0052
bis(chloromethyl)ether	C2C6N1	0.04, 0.04, 0.08	0.0009
1,3-butadiene	C2'C3'O3	0.09, 0.09, 0.00	0.0016
1,4-butanediol dimethane sulphonate (busulfan)	O5O3C2	0.06, 0.02, 0.04	0.0050
chlorambucil	O6C4C2	0.11, 0.06, 0.09	0.0069
chlorambucil	C4'C3'O8	0.07, 0.06, 0.02	0.0006
cyclophosphamide	O5P1O3	0.02, 0.05, 0.06	0.0087
cyclophosphamide	N1N2N3	0.06, 0.07, 0.02	0.0069
diethylstilbestrol	C4O6C2	0.05, 0.08, 0.09	0.0023
diethylstilbestrol	C4'C3'O8	0.07, 0.06, 0.06	0.0008
ethanol	C4'C3'O3	0.00, 0.00, 0.00	0.0000
phenacetin	O6N1C2	0.01, 0.02, 0.02	0.0037
phenacetin	C4'C3'O8	0.07, 0.06, 0.05	0.0017
tamoxifen	O6C4C2	0.10, 0.04, 0.12	0.0098
tamoxifen	C4'C3'O8	0.07, 0.06, 0.04	0.0001
TCDD (dioxin)	C4O6C2	0.04, 0.09, 0.10	0.0073
TCDD (dioxin)	O9C3'P1	0.04, 0.09, 0.09	0.0021
o-toluidine	C4'C3'O8	0.07, 0.06, 0.04	0.0004
o-toluidine	C4,N2,N1	0.06, 0.06, 0.05	0.0031
sulphuric acid	O8P1O5	0.07, 0.05, 0.10	0.0125
sulphuric acid	N1N2N3	0.03, 0.03, 0.06	0.0013
thiotepa	O8O3O5	0.06, 0.04, 0.02	0.0105
thiotepa	N1N2N3	0.09, 0.10, 0.05	0.0022
thorium dioxide	O3P1O5	0.06, 0.08, 0.06	0.0152
thorium dioxide	N1C2N2	0.08, 0.11, 0.08	0.0021
trichloroethylene	C2'C3'O3	0.06, 0.18, 0.18	0.0116
vinyl chloride	C2'C3'O5	0.05, 0.19, 0.19	0.0127

3. Results

The fitting data derive from several categories of carcinogenic compounds. The

first figure gives the fits of seven carcinogen structures to the ribose-phosphate moiety (A) and guanine ring (B) of a cGMP template (Figures 1(1)-(14)). In the main, the compounds are small single ring structures and inorganic oxides that fit the nucleotide phosphate (1 - 7) and guanidino (11 - 14) groups. Fitting values (Table 1) of structures within groups A and B are similar; RMS values 0.0004 - 0.0152 Å and 0.0009 - 0.0069 Å respectively with intermolecular distance values of 0.02 - 0.17 Å. The structure of arsenic trioxide (7, 10) provides inferior fitting values than the other compounds. This arsenic compound forms acids with water that have structures more like sulphuric acid (5, 8). Carcinogen structures (15 - 20) are volatile or gaseous alkane-based compounds that are

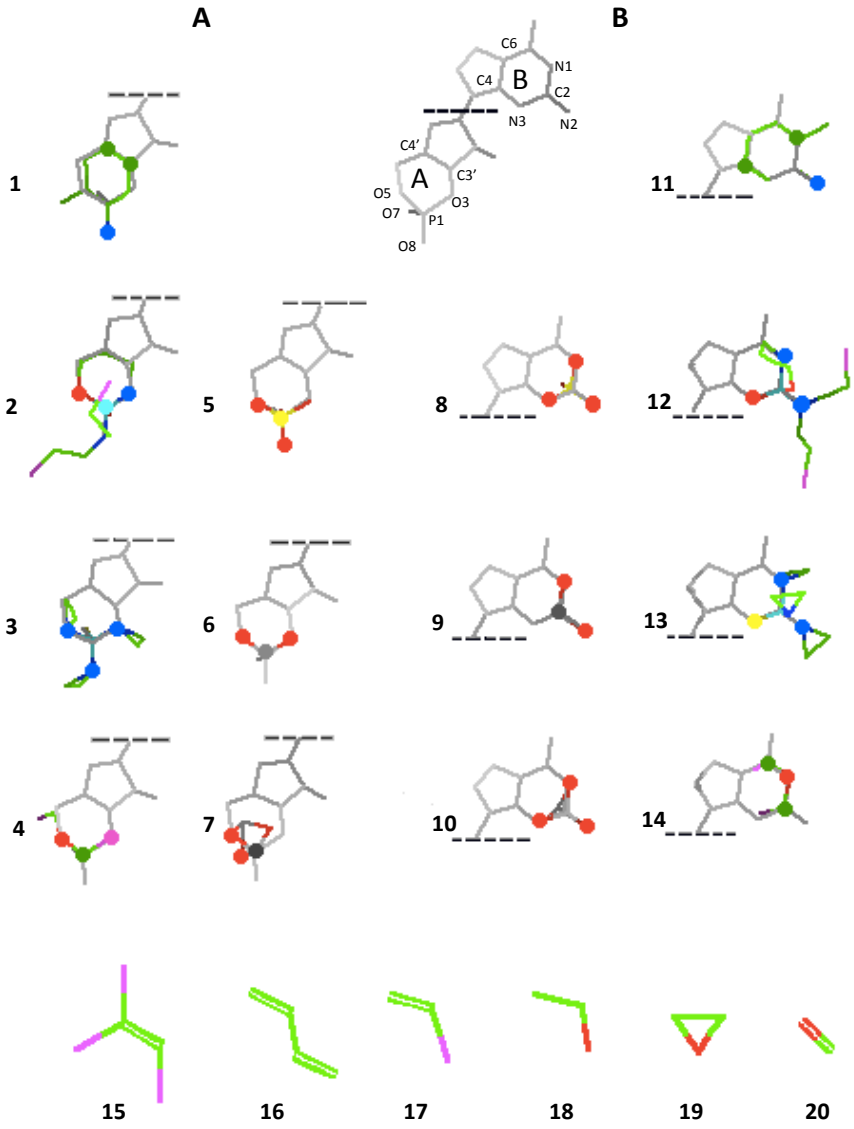

Figure 1. Fitting of compound structures to the cGMP template (grey), ribose-phosphate moiety A or guanine moiety B. 1, 11 o-toluidine; 2, 12 cyclophosphamide; 3, 13 thiotepa; 4, 14 bis(chloromethyl)ether; 5, 8 sulphuric acid; 6, 9 thorium dioxide; 7, 10 arsenic trioxide; 15 trichloroethylene; 16 1,3-butadiene; 17 vinyl chloride; 18 ethanol; 19 ethylene oxide; 20 formaldehyde.

enlarged 2× in comparison to other structures in the figure. The volatile and gaseous carcinogens align and superimpose at several points on the cGMP template but provide only weak or no 3-point pharmacophore fits. The fitting values of trichloroethylene and vinyl chloride are poor, even though the structures incorporate a double bond and chlorine atom. Ethanol provides an excellent conformation for fitting to the nucleotide template, whereas fits of butadiene are achieved only by pairing a sp2 carbon atom with oxygen or sp3 carbon atom; the latter (C2', C4', C3') providing poor-fitting values (0.09, 0.10, 0.18 Å; 0.0199 Å). Ethylene oxide and formaldehyde provide only two useful fitting-points. Consequently, fitting values are given only for structures (15 - 18) and template fits are not provided.

The compound structures given in Figure 2 relate best to the cGMP nucleotide (1) in respect of molecular size. The fits of carcinogenic structures (2 - 9) are based on those of the smaller structures in Figure 1, in that each compound provides a structure that fits the ribose-phosphate ring and another that fits the purine ring. This double-fit provides continuity between the small and large

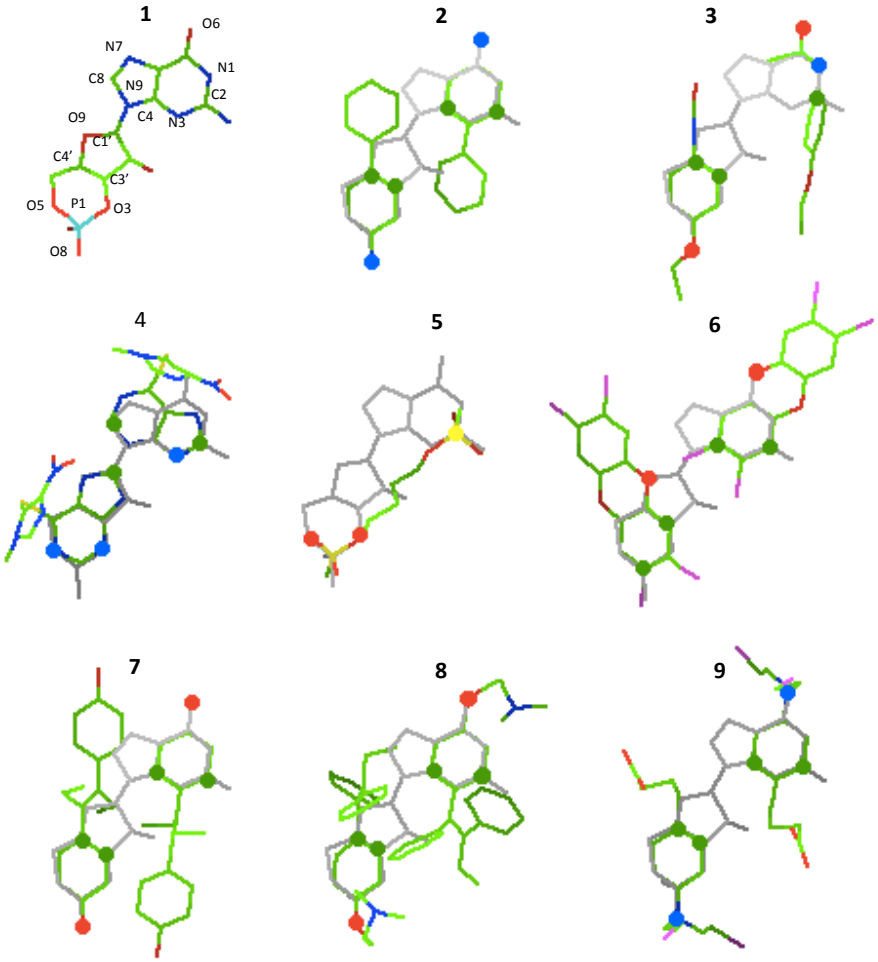

Figure 2. Fitting of compound structures to the cGMP template (grey). 1 cGMP; 2 4-aminodiphenyl; 3 phenacetin; 4 azathioprine; 5 1,4-butanediol dimethane sulphonate; 6 TCDD; 7 diethylstilbestrol; 8 tamoxifen; 9 chlorambucil.

structures and may explain why some small structures fitting solely to the ribose-phosphate moiety may not necessarily be carcinogens. In the main, structures (2 - 9) fit to the ribose-phosphate and guanine moieties via phenol-like rings. Azathioprine (4) and butanediol dimethane sulphonate (5) provide unique fits; a single structure of the latter compound covers both ring systems. Respective compound fitting values to the ribose-phosphate and guanine moieties range from 0.04 - 0.10 Å (intermolecular distance) 0.0000 - 0.0021 Å (RMS) and 0.01 - 0.12 Å (intermolecular distance), 0.0023 - 0.0168 Å (RMS).

4. Discussion

The data generated by this investigation demonstrate that the cGMP structure accommodates the fitting of small molecular weight organic and inorganic carcinogen compounds but not those of volatile and gaseous carcinogens. The latter compounds represent a coherent group that does not conform to the 3-point pharmacophore fits of more conventional drug-like structures. Apart from formaldehyde, the volatile and gaseous carcinogenic group compounds have recognised anesthetic properties. The relationship of compounds such as formaldehyde and ethylene oxide to the cGMP structure can only be considered here in terms of molecular alignment and superimposition, making it difficult to speculate on the likelihood of a common carcinogenic mechanism. Although structural differences in the carcinogenic structures are easily observed, there are also less evident similarities that facilitate fitting to the cGMP template. The triangular pharmacophore of small carcinogen structures is based on a bond angle of 60 degrees and is similar to ethylene oxide. The alternative boomerang-like pharmacophore with a bond angle of 110 degrees is equivalent to the ethanol molecule and the C4'C3'O3 sequence of cGMP. Bond angles in trichloroethylene, 1,3-butadiene and vinyl chloride of approximately 120 degrees are represented in cGMP by C6C5C4 and C5C4N3 sequences.

Weak molecular forces within anesthetic-like molecules, characterized by hydrophobic and hydrophilic interactions necessary for reversible binding [13] may be compensated for by interaction at multiple sites, as determined by the small dimensions of these molecules and available sites on a protein receptor. Current theories on the action of these simple, inert molecules relate to the lipid environment of protein receptors, hydrophobic sites or pockets in proteins, neuron microtubules, lipid rafts and molecular adsorption and modulation of membrane properties [14] [15]. There is no reason to assume that the structural properties of anesthetic compounds differ in their anesthetic and carcinogenic modes. Volatile anesthetics associate with multiple and selective sites on human serum albumin [16] and promote cellular disturbance characteristic of more conventional chemical carcinogens: oxidative stress, mitochondrial dysfunction and chemoresistance [17] [18]. Anesthetic inhibition of the P-glycoprotein ATPase and drug efflux from MDR proteins in cell cultures occurs via membrane fluidization and not through conventional inhibition of enzyme activity [19]. Volatile anesthetics

modulate oxidative stress and nitric oxide levels in patients undergoing surgery, although there is no evidence of a deleterious influence on cancer progression following tumor resection [20] [21].

The simplicity of the small organic and inorganic carcinogen structures establishes a fitting pattern for the phenol-like rings of the larger structures. Phenol is not itself classed as a human carcinogen by the International Agency for Research on Cancer (IARC) as the statistical analysis of cancer cases fails to demonstrate a significant cancer risk [22]. The dual-fits of carcinogen structures to the ribose-phosphate and guanine rings would better antagonize the cGMP structure and nucleotide receptors and may provide an explanation for the lack of carcinogenicity shown by α-lipoic, phytic and folic acids, vitamin E and fraxetin; compounds that fit the ribose-phosphate ring, inhibit apoptosis and are considered as providing protection against cancer [23]. The nucleotide cyclized ring is also a fitting site for ML-9, a compound responsible for effects on store-operated Ca^{2+} entry and endoplasmic reticulum stress [23].

The focus on guanine cyclic nucleotide in this study, rather than the adenine base equivalent, is justified by greater interest in cGMP's modulation of tumour proliferation *in vitro* via the NO-cGMP pathway and the potential benefits of cGMP- targeted medication in cancer patients [1] [2] [3], cGMP homeostatic control of intracellular Ca^{2+} levels [11] and the molecular similarity evident in cGMP, glucocorticoid and sex hormone steroid structures [24]. Nucleotide-binding domains on cell-signaling proteins are common and enzyme-promoted signaling cascades are cyclic nucleotide dependent. Cyclic nucleotide domains are evident on cyclic nucleotide-gated channels and MDR proteins. Genetic polymorphism in the MDR1 gene is associated with an increased risk of breast cancer [25]. MDR proteins provide a cellular environment where carcinogens, chemotherapeutic drugs and endogenous steroids may interact. Disruption of cyclic nucleotide function by compounds of similar molecular structure may initiate the carcinogenic process. The presented findings therefore provide additional evidence for the role of cGMP in carcinogenesis, attributable to the susceptibility of the nucleotide as a target of simple epigenetic chemical carcinogens. These findings should be tested on much larger chemical data sets to assess their use in predictive modeling, as computational methods for investigating the carcinogenic potential of chemicals have major advantages. Furthermore, the results provide a stimulus for developing cGMP-based therapies for the treatment of cancers.

Conflicts of Interest

The author declares no conflicts of interest regarding the publication of this paper.

References

[1] Catalano, S., Campana, A., Giordano, C., Gyorffy, B., Tarallo, R., Rinaldi, A., Bruno, G., Ferraro, A., Romeo, F., Lazino, M., Naro, F., Bonofiglio, D., Ando, S. and Ba-

rone, I. (2015) Expression and Function of Phosphodiesterase Type 5 in Human Breast Cancer Cell Lines and Tissues: Implications for Targeted Therapy. *Clinical Cancer Research*, **22**, 2271-2282. https://doi.org/10.1158/1078-0432.CCR-15-1900

[2] Lv, Y., Wang, X., Li, X., Xu, G., Bai, Y., Wu, J., Piao, Y., Shi, Y., Xiang, R. and Wang, L. (2020) Nucleotide *De Novo* Synthesis Increases Breast Cancer Stemness and Metastasis Via cGMP-PKG-MAPK Signaling Pathway. *PLOS Biology*, **18**, e3000872. https://doi.org/10.1371/journal.pbio.3000872

[3] Di Iorio, P., Ronci, M., Giuliani, P., Caciagli, F., Ciccarelli, R., Caruso, V., Beggiato, S. and Zuccarini, M. (2022) Pros and Cons of Pharmacological Manipulation of cGMP-PDEs in the Prevention and Treatment of Breast Cancer. *International Journal of Molecular Sciences*, **23**, Article No. 262. https://doi.org/10.3390/ijms23010262

[4] Honorat, M., Terreux, R., Falson, P., Di Pietro, A., Dumontet, C. and Payen, L. (2013) Localization of Putative Binding Sites for Cyclic Guanosine Monophosphate and the Anti-Cancer Drug 5-Fluoro-2'-Deoxyuridine-5'-Monophosphate on ABCC11 *in Silico* Models. *BMC Structural Biology*, **13**, Article No. 7. https://doi.org/10.1186/1472-6807-13-7 http://www.biomedcentral.com/1472-6807/13/7

[5] Prior, I.A., Hood, F.E. and Hartley, J.L. (2020) The Frequency of Ras Mutations in Cancer. *Cancer Research*, **80**, 2969-2974. https://doi.org/10.1158/0008-5472.CAN-19-3682

[6] Dang, D. and Rao, R. (2016) Calcium-ATPases: Gene Disorder and Dysregulation in Cancer. *Biochimica et Biophysica Acta*, **1863**, 1344-1350. https://doi.org/10.1016/j.bbamcr.2015.11.016

[7] Themistocleous, S.C., Yiallouris, A., Tsioutis, C., Zaravinos, A., Johnson, E.O. and Patrikios, I. (2021) Clinical Significance of P-Class Pumps in Cancer (Review). *Oncology Letters*, **22**, Article No. 658. https://doi.org/10.3892/ol.2021.12919

[8] Gagnon, F., Hamet, P. and Orlov, S.N. (1999) Na^+, K^+ Pump and Na^+-Coupled Ion Carriers in Isolated Mammalian Kidney Epithelial Cells: Regulation by Protein Kinase C. *Canadian Journal of Physiology and Pharmacology*, **77**, 305-319. https://doi.org/10.1139/y99-041

[9] Lorenzo, P.S., Kung, J.W., Bottorff, D.A., Garfield, S.H., Stone, J.C. and Blumberg, P.M. (2001) Phorbol Esters Modulate the Ras Exchange Factor RasGRP3. *Cancer Research*, **61**, 943-949.

[10] Williams, W.R. (2003) Relative Molecular Similarity in Selected Chemical Carcinogens and the Nucleoside Triphosphate Chain. *Pharmacology & Toxicology*, **92**, 57-63. https://doi.org/10.1034/j.1600-0773.2003.920202.x

[11] Williams, W.R. (2020) Tumour Initiation, Store-Operated Calcium Entry (SOCE) and Apoptosis: Cyclic Nucleotide Dependence. *General Physiology and Biophysics*, **39**, 419-435. https://doi.org/10.4149/gpb_2020020

[12] U.S. Department of Health and Human Services (2021) National Toxicology Program.

[13] Bertaccini, E.J. (2010) The Molecular Mechanisms of Anesthetic Action: Updates and Cutting Edge Developments from the Field of Molecular Modeling. *Pharmaceuticals*, **3**, 2178-2196. https://doi.org/10.3390/ph3072178

[14] Cukras, J. and Sadlej, J. (2021) Towards Quantum-Chemical Modeling of the Activity of Anesthetic Compounds. *International Journal of Molecular Sciences*, **22**, Article No. 9272. https://doi.org/10.3390/ijms22179272

[15] Cantor, R.S. (2018) Understanding Anesthetic Mechanisms: Analysis of the Com-

plex Kinetics of Ligand-Gated Ion Channels. *Methods in Enzymology*, **602**, 97-110. https://doi.org/10.1016/bs.mie.2018.02.002

[16] Liu, R. and Eckenhoff, R.G. (2005) Weak Polar Interactions Confer Albumin Binding Site Selectivity for Haloether Anesthetics. *Anesthesiology*, **102**, 799-805. https://doi.org/10.1097/00000542-200504000-00016

[17] Kishikawa, J.I., Inoue, Y., Fujikawa, M., Nishimura, K., Nakanishi, A., Tanabe, T., Imamura, H. and Yokoyama, K. (2018) General Anesthetics Cause Mitochondrial Dysfunction and Reduction of Intracellular ATP Levels. *PLoS ONE*, **13**, e0190213. https://doi.org/10.1371/journal.pone.0190213

[18] Merk, R., Hebelbach, K., Osipova, A., Popadic, D., Schmidt-Heck, W., Kim, G.J., Gunther, S., Pineres, A.G., Merfort, I. and Humar, M. (2020) Particulate Matter ($PM_{2.5}$) from Biomass Combustion Induces an Anti-Oxidative Response and Cancer Drug Resistance in Human Bronchial Epithelial BEAS-2B Cells. *International Journal of Environmental Research and Public Health*, **17**, Article No. 8193. https://doi.org/10.3390/ijerph17218193

[19] Regev, R., Assaraf, Y.G. and Eytan, G.D. (1999) Membrane Fluidization by Ether, Other Anesthetics, and Certain Agents Abolishes P-Glycoprotein ATPase Activity and Modulates Efflux from Multidrug Resistant Cells. *European Journal of Biochemistry*, **259**, 18-24. https://doi.org/10.1046/j.1432-1327.1999.00037.x

[20] Dharmalingam, S.K., Amirtharaj, G.J., Ramachandran, A. and Korula, M. (2021) Volatile Anesthetic Preconditioning Modulates Oxidative Stress and Nitric Oxide in Patients Undergoing Coronary Artery Bypass Grafting. *Annals of Cardiac Anaesthesia*, **24**, 319-326. https://www.annals.in/text.asp?2021/24/3/319/320937

[21] Liu, X. and Wang, Q. (2022) Application of Anesthetics in Cancer Patients: Reviewing Current Existing Link with Tumor Recurrence. *Frontiers in Oncology*, **12**, Article ID: 759057. https://doi.org/10.3389/fonc.2022.759057

[22] Public Health England (2016) Phenol: Toxicological Overview.

[23] Williams W.R. (2021) Cyclic Nucleotide Structural Differentiation of Compounds Modulating Apoptosis and Drug Resistance. *Journal of Biosciences and Medicines*, **9**, 10-28. https://doi.org/10.4236/jbm.2021.98002

[24] Williams, W.R. (2022) Contributors to Cancer Susceptibility, Development and Treatment: Cyclic Nucleotides, Steroids and Autophagy Modulators. *Journal of Biosciences and Medicines*, **10**, 65-86. https://doi.org/10.4236/jbm.2022.102008

[25] Jaramillo-Rangel, G., Ortega-Martinez, M., Cerda-Flores, R.M. and Barerra-Saldana, H.A. (2018) C3435T Polymorphism in the MDR1 Gene and Breast Cancer Risk in Northeastern Mexico. *International Journal of Clinical and Experimental Pathology*, **11**, 904-909.