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Keywords: colorectal cancer; oxidative stress; free radicals; antineoplastic drugs; quantum mechanics; drug screening

Abbreviations

OS oxidative stress, ROS reactive oxygen species, CRC Colorectal cancer, ET electron transfer, $\Delta G_{desolv,CDS}$ free energy of water desolvation, $\Delta G_{lipo,CDS}$ lipophilicity free energy, CDS cavity dispersion solvent structure of the first solvation shell, HOMO highest occupied molecular orbital, LUMO lowest unoccupied molecular orbital, R^2 multiple correlation coefficient, the F test of significance, SEE standards errors for the estimate, standard errors of the variables SE($\Delta G_{desolCDS}$), SE($\Delta G_{lipoCDS}$), SE(Dipole Moment), SE (Molecular Volume), LD_{50} drug toxicity, ID_{90} drug concentration that produces 90% inhibition of cell growth

Abstract

It has been found that a free radical mechanism is involved in the cytotoxicity of most of a panel of 12 anti-colorectal cancer drugs tested against a panel of 9 colorectal cancer variants. The colorectal cancers Ht-29, LoVo and WiDr do not appear to involve free radical oxidative stress processes, although such processes may be too small to be detected or are kinetically slower than cell membrane or other drug-target interactions.

The colorectal cancer variants 320, OM-1, 205 and SW-620 CRC show the strongest dependencies on free radical processes.

The toxicity LD_{50} of the tested anti-colorectal cancer drugs is linearly related to the HOMO-LUMO energy gap, and can be a useful theoretical screening tool for designing anti-colorectal cancer drugs.

Introduction

Colorectal cancer (CRC) or bowel or colon cancer develops in the bowel or rectum. It is the fourth most common cause of cancer death after lung, stomach, and liver cancer and is more common in developed countries with highest rates in Australia, New Zealand, Europe and the US.

Chemotherapy is usually offered for stage 3 and 4 CRC, including the use of capecitabine, fluorouracil, uracil, irinotecan, oxaliplatin. Some specific regimens used for CRC are CAPOX, FOLFOX, FOLFOXIRI. Antiangiogenic drugs such as bevacizumab can also be used in first line therapy.[1,2]

The role of redox processes in the development of CRC is well known. The role of reactive oxygen species (ROS) in the redox modification of lipids, proteins, DNA is involved in many biological processes related to diseases including CRC. ROS also play an important role in cellular signaling. The spread of lipid peroxidation from malignant into the adjacent non-malignant colon tissue also occurs. A low catalase activity is indicative of oxidative-antioxidative disorders. Oxidative stress is an imbalance between ROS production and the ability of naturally occurring biological antioxidants. ROS include free radicals such as the superoxide anion ($O_2\bullet^-$), hydroxyl radical (HO•) and the non-radical molecules such as hydrogen peroxide (H_2O_2), and singlet oxygen. The primary source of ROS is the mitochondria. ROS can invoke single and double strand DNA breaks, and the common genetic mutations include p53, KRAS, APC, and BRAF mutations. [3-7] Intracolonic production of ROS notably HO• has been physically observed.[8]

Because CRC has higher incidence rates in western countries, sporadic human CRC could be linked to various environmental and lifestyle factors, such as dietary habits, obesity, and physical inactivity. While there is much evidence of a link between ROS induced oxidative stress and carcinogenesis, there is some debate whether oxidative stress is a cause or consequence of CRC. [6] A comprehensive study of 81 primary CRCs in clinical stages II, III, and IV has shown that the progression of CRC is associated with enhanced oxidative stress and the gradual advancement of oxidative-antioxidative disorders is followed by progression of CRC. [7]

Cancer cells characteristically have a high antioxidant capacity that regulates ROS to levels that are compatible with cellular biological functions but still higher than in normal cells. Targeting these high antioxidant defence mechanisms is a strategy to kill cancer cells but not normal cells. [9-12] However, all antineoplastic agents generate some ROS as they induce apoptosis in cancer cells, because one of the pathways of drug-induced apoptosis involves the release of cytochrome C from mitochondria. When this occurs, electrons are diverted from the electron transfer system to oxygen by NADH dehydrogenase and reduced coenzyme Q10, resulting in the formation of superoxide radicals. [9]

There is extensive evidence supporting involvement of electron transfer (ET), reactive oxygen species (ROS) and oxidative stress (OS) in the mechanism of many anticancer drugs. These free radical ET agents function catalytically in redox cycling with formation of ROS from oxygen. [13-15] The metabolism of a drug may generate a reactive intermediate that can reduce molecular oxygen directly to generate ROS. [16]

We have recently shown that the radical form of a wide range of current antineoplastic drugs is involved in oxidative stress processes in human DLD-1 human colorectal cancer cells as determined by the CellROX assay. The oxidative stress capability of these drugs is dependent on the lipophilicity and electron affinity of the drugs in water. All of the drugs examined are known to be involved in oxidative processes to some degree according to literature sources. The antioxidant properties of the radical drug species in the ORAC or ORAC/Cu assays are also dependent on the lipophilicity and electron affinity properties of the drugs. Linear equations have

been derived which can predict the oxidative stress in colorectal cancer cells and the redox behavior in the ORAC assays. [17]

Study objectives

Determine (a) whether the efficacy of a panel of anticancer drugs previously tested against a panel of CRCs can be ascribed to free radical chemotherapy and its effect on oxidative stress, and (b) if so develop a drug efficacy screening methodology using quantum mechanically derived molecular properties of the drugs

Results

Scheithauer [18] has previously used a panel of 12 anti-CRC drugs to examine their efficacy (the ratio of the concentration required to decrease cell growth to 10% of the control to 0.1 of the peak plasma concentration in humans, ie ID_{90}:0.1 peak plasma concentration) against 9 variants of CRC. Drugs were tested for 1 hour exposure as well as continuous exposure using standard clinical patient treatment formulations. Cell lines were propagated as monolayer cultures. Animal toxicology LD_{50} for the 12 drugs was also determined. A reasonable correlation ($P < 0.02$) between the percentage of in vitro response rate of the entire panel of cell lines for the 1 hour incubation and the known in vivo response data for the various drugs was observed (using a cut-off definition of in vitro drug efficacy of less than 30 for the ID_{90}:0.1 peak plasma concentration).

We have previously described a model that has been shown to apply to a wide range of drug transport, binding, metabolic and cytotoxicity properties of cells and tumours (Equation 1). The model is based on establishing linear free energy relationships between the four drug properties and various biological processes. Equation 1 has been previously applied to passive and facilitated diffusion of a wide range of drugs crossing the blood brain barrier, the active competitive transport of tyrosine kinase inhibitors by the hOCT3, OATP1A2 and OCT1 transporters, and cyclin-dependent kinase inhibitors and HIV-1 protease inhibitors. The model also applies to PARP inhibitors, the anti-bacterial and anti-malarial properties of fluoroquinolones, and active organic anion transporter drug membrane transport, and some competitive statin-CYP enzyme binding processes. There is strong independent evidence from the literature that $\Delta G_{desolvation}$, $\Delta G_{lipophilicity}$, the dipole moment and molecular volume are good inherent indicators of the transport or binding ability of drugs. [19-30]

Equation 1:

Transport or Binding or Cytotoxicity = $\Delta G_{desolv,CDS}$ + $\Delta G_{lipo,CDS}$ + Dipole Moment + Molecular Volume or Electron Affinity

Or

Transport or Binding or Cytotoxicity = $\Delta G_{desolv,CDS}$ + $\Delta G_{lipo,CDS}$ + Dipole Moment + Electron Affinity

Eq 1 uses the free energy of water desolvation ($\Delta G_{desolv,CDS}$) and the lipophilicity free energy ($\Delta G_{lipo,CDS}$) where CDS represents the non-electrostatic first solvation shell solvent properties. CDS may be a better approximation of the cybotactic environment around the drug approaching or within the protein receptor pocket, or the cell membrane surface or the surface of a drug transporter, than the bulk water environment outside the receptor pocket or cell membrane surface. The CDS includes dispersion, cavitation, and covalent components of hydrogen bonding, hydrophobic effects. Desolvation of water from the drug ($\Delta G_{desolv,CDS}$) before binding in the receptor pocket is required, and hydrophobic interactions between the drug and protein ($\Delta G_{lipo,CDS}$) is a positive contribution to binding. $\Delta G_{lipo,CDS}$ is calculated from the solvation energy in n-octane. In some biological processes, where biological reduction may be occurring, and the influence of molecular volume is small, the reduction potential (electron affinity) has been included in place of the molecular volume. In other processes, the influence of some of the independent variables is small and can be eliminated to focus on the major determinants of biological activity.

We have recently used this model to develop a predictive model of the transport and efficacy of hypoxia specific cytotoxic analogues of tirapazmine and the effect on the extravascular penetration of tirapazamine into tumours. [19] It was found that the multiparameter model of the diffusion, antiproliferative assays IC_{50} and aerobic and hypoxic clonogenic assays for a wide range of neutral and radical anion forms of tirapazamine (TPZ) analogues showed: (a) extravascular diffusion is governed by the desolvation, lipophilicity, dipole moment and molecular volume, similar to passive and facilitated permeation through the blood brain barrier and other cellular membranes, (b) hypoxic assay properties of the TPZ analogues showed dependencies on the electron affinity, as well as lipophilicity and dipole moment and desolvation, similar to other biological processes involving permeation of cellular membranes, including nuclear membranes, (c) aerobic assay properties were dependent on the almost exclusively on the electron affinity, consistent with electron transfer involving free radicals being the dominant species.

The model has also been recently applied to triple negative breast and ovarian cancers where transient and stable free radicals are involved in the cytotoxic oxidative stress processes. The electron affinity of the various drugs, along with the water desolvation, lipophilicity and dipole moment, has been shown to be an important predictor of cytotoxic efficacy. [28-30]

In our recent study of ORAC and CellROX free radical anticancer drugs and oxidative stress in colorectal cancer cells [17] we found that eq 2 was applicable.

Eq 2.

Oxidative Stress or Oxidative Properties = $\Delta G_{desolv,CDS}$ + $\Delta G_{lipo,CDS}$ + Dipole Moment + Electron Affinity

Screening methodology

Given the successful anti-tumorigenic model as described by eq 1 and 2 for wide range of anticancer drugs, the screening model used was to calculate five molecular parameters for the panel of 12 anti-CRC drugs from Scheithauer [18]: (1) the free energy of water desolvation ($\Delta G_{desolv,CDS}$), (2) the lipophilicity free energy ($\Delta G_{lipo,CDS}$), (3) the dipole moment in water, (4) the molecular volume in water, and (5) the adiabatic electron affinity, AEA, (or reduction potential) in water. These values are shown in Table 1 along with the anti-tumour efficacy (ID_{90}:0.1 peak plasma concentration) against the 9 CRC tumours for the 1 hour exposure as well as continuous exposures.

Stepwise multiple regression was then applied to seek out which drug molecular properties (Table 1) had the largest effect on the CRC variants shown in Table 1(a) for the 1 hour exposures and Table 1(b) for the continuous exposures. These results are shown in Table 2(a) and 2(b) which shows the molecular properties which had significant impacts on the CRC variants for 1 hour and continuous exposures respectively.

The statistical significance of the linear regression equations in Table 2(a) and 2(b) are shown (F test) and which relationships showed major statistical outliers, particularly BCNU and Vincristine, to which a number of CRC variants were resistant. The 1 hour exposures (approximating a bolus injection of drugs) are possibly more informative than the continuous exposures (approximating intravenous injection) since processes whereby drugs have to first permeate the cell membrane, then interact with the appropriate cystolic target is confounded when continuous exposure regimes are used. We have previously shown that desolvation, lipophilicity, molecular volume and dipole moment all play roles in both cell membrane permeation and drug-target interaction in cytotoxic processes.

The molecular properties in Table 1 have been scaled as indicated to provide a direct comparison of the linear regression coefficients to allow a direct relative comparison of the absolute magnitude of which molecular properties are most significant.

The findings from Table 2(a) and (b) are:
(1) CRC variants 320, OM-1, 205 and SW-620 CRC show the greatest effect on AEA for both 1 hour and continuous exposures.
(2) Ht-29, LoVo and WiDr do not show any significant dependency on AEA for 1 hour and continuous exposures. It is known that WiDR is a derivative of the colon adenocarcinoma cell line Ht-29. [31]
(3) Variants 205, DLD-1 and HOT-3 show only minor partial dependencies on AEA but major dependencies on desolvation, lipophilicities or molecular volumes of the drugs for 1 hour exposures.
(4) Where there is no effect of drug AEA on the CRC variants, there are dependencies on drug desolvation, lipophilicity, molecular volume or dipole moment for both exposure regimes.

Table 3 shows the relationship between animal toxicology of the panel of drugs and the HOMO-LUMO gap for the drugs. The linear relationship is $LD_{50} = 14273.2 \text{ (HOMO-LUMO)} -1805.2$ (R^2 0.416, SEE 907.3, $SE_{(HOMO-LUMO)}$ 5346, F 7.13, Significance 0.023. No other linear

dependencies were separately observed with desolvation, lipophilicity, molecular volume, dipole moment or AEA.

Discussion

Scheithauer [18] found that the response rate for the panel of drugs for 1 hour exposure was similar to in vivo clinical data. This study finds a strong or partial dependency on AEA for 5 of the 9 drugs for 1 hour exposure. Similar findings were found for continuous exposure, but SW-620 showed dependency on AEA but not for the 1 hour exposure. Overall only Ht-29, LoVo and WiDr showed no dependency on AEA, indicating possibly that a different non-oxidative stress mechanism occurs when treated by the panel of drugs, or kinetic effects related to cell membrane permeability or cell target interaction are dynamically dominating over free radical processes. Scheithauer [18] found that 320DM, OM-1 and Ht-29 closely resembled in vivo clinical results. This study finds that 320DM and OM-1 cytotoxicities are strongly dependent on AEA for both 1 hour and continuous exposure.

These results are consistent with our previous study where it was shown that the radical form of a wide range of current antineoplastic drugs is involved in oxidative stress processes in human DLD-1 human colorectal cancer cells as determined by the CellROX assay. The oxidative stress and redox capability of these drugs was dependent on the lipophilicity and electron affinity of the drugs in water. [17]

Scheithauer [18] found a good correlation between the peak plasma level in humans and the LD_{50} values in normal (non-tumor-bearing) mice ($P < 0.01$), indicating that LD_{50} is useful for screening investigational drugs for their activity in colorectal cancer. However we have recently shown that LD_{50} of a series of Pt(II)- and Pt(IV)-nitroxide conjugate drug treatments of healthy and leukemic mice is inversely linearly related to the HOMO-LUMO energy gap of the drugs. Also in another series of common neutral Pt(II) and Pt(IV) drugs, we also found that the LD_{10} was directly related to the HOMO-LUMO gap, unlike the Pt-Nitroxides, which are stable free radicals, and which showed a negative LD_{50} relationship with the HOMO-LUMO gap, consistent with antioxidant behaviour having a positive lowering effect on lethality. [32]

This study also demonstrates that the LD_{50} of the panel of 12 anti-CRC drugs is quite strongly linearly related to the HOMO–LUMO gap, indicating that the HOMO-LUMO gap can be a useful theoretical screening tool for designing anti-CRC drugs.

Conclusions

It has been found that a free radical mechanism is involved in the cytotoxicity of most of a panel of 12 anti-colorectal cancer drugs tested against a panel of 9 colorectal cancer variants. The colorectal cancers Ht-29, LoVo and WiDr do not appear to involve free radical oxidative stress processes, although such processes may be too small to be detected or are kinetically slower than cell membrane or other drug-target interactions.

The colorectal cancer variants 320, OM-1, 205 and SW-620 CRC show the strongest dependencies on free radical processes.

The toxicity LD_{50} of the tested anti-colorectal cancer drugs is linearly related to the HOMO-LUMO energy gap, and can be a useful theoretical screening tool for designing anti-colorectal cancer drugs.

Experimental

All calculations were carried out using the Gaussian 09 package. Energy optimizations were at the DFT/B3LYP/6-31G(d,p) (6d, 7f) or DFT/B3LYP/3-21G (for larger molecules) level of theory for all atoms. Selected optimizations at the DFT/B3LYP/6-311$^+$G(d,p) (6d, 7f) level of theory gave very similar results to those at the lower level. Optimized structures were checked to ensure energy minima were located, with no negative frequencies. Energy calculations were conducted at the DFT/B3LYP/6-31G(d,p) (6d, 7f) for neutral compounds and DFT/B3LYP/6-31+G(d) (6d, 7f) level of theory for anions with optimized geometries in water, using the IEFPCM/SMD solvent model. With the 6-31G* basis set, the SMD model achieves mean unsigned errors of 0.6 - 1.0 kcal/mol in the solvation free energies of tested neutrals and mean unsigned errors of 4 kcal/mol on average for ions. [33] The 6-31G** basis set has been used to calculate absolute free energies of solvation and compare these data with experimental results for more than 500 neutral and charged compounds. The calculated values were in good agreement with experimental results across a wide range of compounds. [34,35] Adding diffuse functions to the 6-31G* basis set (ie 6-31$^+$G**) had no significant effect on the solvation energies with a difference of less than 1% observed in solvents, which is within the literature error range for the IEFPCM/SMD solvent model. HOMO and LUMO calculations included both delocalized and localized orbitals (NBO).

Electron affinities (EA) in eV in water were calculated by the SCF difference between the optimised/relaxed neutral and optimised radical species method as previously described. [19,28-30] It has been shown that the B3LYP functional gives accurate electron affinities when tested against a large range of molecules, atoms, ions and radicals with an absolute maximum error of 0.2 eV. [36-38]

It is noted that high computational accuracy for each species in different environments is not the focus of this study, but comparative differences between various species is the aim of the study. The literature values for drug efficacy and toxicity used in the stepwise multiple regressions have much higher experimental uncertainties than the calculated molecular properties. The statistical analyses include the multiple correlation coefficient R^2, the F test of significance, standards errors for the estimates (SEE) and each of the variables SE($\Delta G_{desolCDS}$), SE($\Delta G_{lipoCDS}$), SE(Dipole Moment), SE (EA) as calculated from "t" distribution statistics. Residual analysis was used to identify outliers.

Table 1 Molecular properties of anti-CRC drugs

	$\Delta G_{desolv,CDS}$ Water kcal/mol	$\Delta G_{lipo,CDS}$ kcal/mol	Dipole Moment Water D	Molecular Volume/20 Water cm^3/mol	5.AEA Water eV
BCNU	-4.84	-4.44	4.28	6.25	16.2
Bisantrene	2.4	-13.56	11.04	13.3	14.9
Cis-Platinum vert	-4.57	-0.47	17.08	5.2	9.1
Doxorubicin	-11.11	-9.69	9.35	19.9	17.3
5-Fluorouracil	-5.76	-2.87	6.62	4	11.45
FUDR	-7.12	-5.07	6	8.4	11.5
Melphalan vert	-5.66	-6.41	10.7	10.25	4.55
Methotrexate	-6.11	-11.81	3.47	16.4	13.85
Mitomycin C	-4.54	-3.64	9.9	10.55	18.05
MitoxantroneH$^+$H$^+$	-12.54	-10.87	16.23	16.45	36.3
PALA	-11.17	-4.09	9.91	6.85	10.3
Vincristine	-15.04	7.65	18.5	32.55	7.65

Footnotes: BCNU: l,3-bis(2-chloroethyl)-1-nitrosourea; FUDR: 5-fluoro-2-deoxyuridine; PALA: N-(phosphonacetyl)-L-aspartate; Cis-Platinum and Malphalan given as vertical electron affinity as adiabatic electron affinities showed elongation of Pt-Cl and CH$_2$-Cl bonds, MitoxantroneH$^+$H$^+$ is di-protonated at physiological pH. Molecular volumes divided by 20, AEAs multiplied by 5

Table 1(a) Drug efficacy ID$_{90}$:0.1 peak plasma concentration

1 Hour Exposure	320DM	OM1	Ht29	205	DLD1	HOT3	LoVo	WiDr	SW620
BCNU	146.2	840.2	588.5	289.3	275.7	1094.7	632.8	232.7	132.6
Bisantrene	59.8	16.7	6.1	6.1	8.7	19.9	7.8	7.9	20.5
CisPt vert	42.1	54.5	48.3	61	25.5	82.4	41.3	30.5	20.6
Doxorubicin	227.8	120	90.3	126.5	6.5	307.8	8.8	6.5	193.7
5-FU	10	11	5.6	18.3	5.4	24.6	1.4	1.3	30.1
FUDR	64.1	9.5	1.2	30.6	32.1	29.5	8.1	12.7	50.9
Melphalan vert	28.1	21.3	25.5	144.2	28.4	82.3	28.7	17.8	47.3
Methotrexate	26.1	47.7	37.5	24.8	21.4	251.4	450.6	6.2	102.2
Mitomycin C	81.2	52.7	6.7	80.5	16.1	393.8	8.7	49.5	9.3
MitoxantroneH$^+$H$^+$	165.2	90.8	32	118	36.2	321.6	6.4	50.6	10
PALA	234.8	76.3	246.9	444.2	318.8	53	112.8	385.6	44.9
Vincristine	25.37	1045	517	912	616	9999	719	6	1019

Footnote: from ref 18

Table 1(b) Drug efficacy ID$_{90}$:0.1 peak plasma concentration

Continuous Exposure	320DM	OM1	Ht29	205	DLD1	HOT3	LoVo	WiDr	SW620
BCNU	91.5	290	330.5	120.6	108.6	335.1	62.4	203.8	91.7
Bisantrene	5.8	7.3	0.4	0.4	3.3	75.6	0.4	0.3	0.4
CisPt vert	0.9	7.2	36.2	14.5	7.2	484.8	6.9	5.6	76.9
Doxorubicin	7	158	124.7	10	7.3	1083.7	6.5	4.8	6.5
5-FU	0.6	1.7	5.2	1.2	0.3	39.9	1.1	4.6	1.5
FUDR	0.9	5.2	35.4	1.2	5.7	12	2.8	1.2	15.6
Melphalan vert	19.2	10	36.3	35.5	34.9	405.5	7.3	9.5	287
Methotrexate	0.9	33	18.2	16.6	163.5	140.7	134.6	0.3	2432.8
Mitomycin C	3.5	0.9	42	5.4	0.1	140.5	0.1	2.6	0.1
MitoxantroneH$^+$H$^+$	4	114	78.4	6.4	18.8	924.4	0.1	3.4	5.4
PALA	11.1	0.5	221	7.1	19	33	29.8	146.6	139
Vincristine	1412	2011	10	10	408	2935	8	75	9971

Footnote: from ref 18

Table 2(a) Major dependencies of drug efficacies on molecular properties 1 hour exposures

Colorectal Cancer Cell Line	Tumour Origin & Cell Differentiation	Antineoplastic Effect: 1 Hour Exposure	Ratio relative to AEA	Significance F Test	Statistical Outliers (Resistance)
320DM	Primary Moderate	AEA		0.148	
OM-1	Metastatic Well	AEA		0.127	
Ht-29	Primary Well	Lipo+DM+Vol		0.06	excl BCNU
205	Ascites Poor	Desolv+AEA	<-40:-12>*	0.023	excl BCNU
DLD-1	Primary Poor	Desolv+AEA	<-12:-4>	0.307	excl BCNU, Vincristine
HOT-3	Metastratic Well	Lipo+Vol+AEA	<-39:33:7>**	0.015	excl BCNU, Vincristine
LoVo	Lymph Node Well	Lipo+DM+Vol		0.081	
WiDr	Primary Moderate	Desolv+Vol		0.187	
SW-620	Lymph Node Moderate	Lipo+DM+Vol		0	

Footnote: * Regression coefficients desolvation:AEA ratio is -40:-12 indicating that desolvation effect is greater than Adiabatic Electron Affinity effect by 40/12 times; ** lipophilic free energy:molecular volume:AEA coefficients in ratios 39:33:7

Table 2(b) Major dependencies of drug efficacies on molecular properties continuous exposures

Colorectal Cancer Cell Line	Tumour Origin & Cell Differentiation	Antineoplastic Effect: Continuous Exposure	Significance F Test	Statistical Outliers (Resistance)
320DM	Primary Moderate	AEA	0.054	excl BCNU, Vincristine
OM-1	Metastatic Well	AEA	0.393	excl BCNU
Ht-29	Primary Well	Desolv	0.155	excl BCNU
205	Ascites Poor	AEA	0.254	excl BCNU
DLD-1	Primary Poor	Lipo+DM+Vol	0.002	
HOT-3	Metastratic Well	Lipo+DM+Vol	0	
LoVo	Lymph Node Well	Lipo+DM	0.096	
WiDr	Primary Moderate	Lipo+DM	0.341	
SW-620	Lymph Node Moderate	AEA	0.103	exclude Methotrexate, Vincristine

Table 3 Drug toxicology

	LD_{50}	HOMO - LUMO Gap
	mg/kg	eV
BCNU	42	4.930814
Bisantrene	245	2.87903
CisPt vert	26.8	4.968911
Doxorubicin	24	2.824606
5-FU	171	5.3526
Melphalan vert	29.6	5.194771
Methotrexate	69	3.494021
Mitomycin C	12	2.824606
MitoxantroneH^+H^+	86.5	2.566092
Vincristine	5.8	3.951182
FUDR	550	5.32811
PALA	4000	7.015254

Footnote: LD_{50} values from ref 18

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