Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 11 May 2022 doi:10.20944/preprints202205.0149.v1

Article

Cytotoxicity of Diphenyltin(IV) Diisopropyl Dithiocarbamate Compound on Acute Lymphoblastic Leukemia Cells, CCL-119 (CCRF-CEM)

Nur Rasyiqin Rasli, Normah Awang*, Nurul Farahana Kamaludin, Asmah Hamid and Nur Aini Yusof

Center for Toxicology & Health Risk Studies, Faculty of Health Sciences, Universiti Kebangsaan Malaysia, Jalan Raja Muda Abdul Aziz, 50300 Kuala Lumpur, Malaysia.
*Author for correspondence: E-mail: norm@ukm.edu.my

Abstract: Acute lymphoblastic leukemia (ALL) is the most common type of leukemia affecting children under the age of 15 years old in Malaysia. Chemotherapy is the primary treatment for cancer, which involves the intake of chemotherapeutic drugs to kill cancer cells. Glucocorticoids such as dexamethasone are chemotherapeutic agents used in the treatment of ALL. Although dexamethasone is highly effective, it is also associated with adverse effects such as bone fractures and organ toxicity. Therefore, there is a need to develop a new anticancer drug which milder side effects and better efficacy. Organometallic compounds such as organotin have a high potential to be developed as an antineoplastic agent and show high specificity towards cancer cells compared to normal cells. This study is done to evaluate the cytotoxic effects of diphenyltin(IV) diisopropyl dithiocarbamate (DPDT) against leukemic cells CCL-119 using the Trypan Blue exclusion (TBE) method at the intervals of 24, 48 and 72 h. Dexamethasone was used as a positive control. The cell's morphological changes were observed at 12, 24 and 48 h using the IC_{50} values obtained using TBE assay. Results show that DPDT has a lower IC_{50} value than dexamethasone against CCL-119 cells at 24 h with a value of $4.16 \pm 0.44 \mu M$ and a selectivity index of 2.02. Dexamethasone exhibited cytotoxic effects against CCL-119 but only IC_{25} and IC_{10} values were obtained. Cytotoxicity testing has shown that DPDT is toxic on CCL-119 cells with IC_{50} values of less than $10 \mu M$. Morphological changes in cells show characteristics of apoptosis such as cell shrinkage, blebbing and formation of apoptotic bodies. In conclusion, DPDT has the potential to be made into an antineoplastic agent but requires a more detailed study involving the molecular pathway of DPDT leading to cell death.

Keywords: organotin(IV) dithiocarbamate; childhood leukemia; antileukemia activity; anticancer potential; inhibitory effects

1. Introduction

According to World Health Organization (WHO), cancer is the main leading cause of death and morbidity globally, with 14 thousand new cases in 2012 and estimated to increase up to 70 % in two decades. Generally, leukemia is a blood cancer that causes abnormal and immature blood cells. This type of cancer usually affects white blood cells production. Globocan 2020 reports that leukemia is Malaysia's top 10 highest cancer cases. There are four main types of leukemia and acute lymphoblastic leukemia (ALL) is a significant type of blood cancer in childhood.

There are many treatment methods for leukemia and chemotherapy is one [1]. For pediatric leukemia, the primary treatment involved in the chemotherapeutic regiment is given orally or intervein [2]. However, the immediate problems patients face with the chemotherapy regimens are the side effects and drug resistance [3]. Also, the current chemotherapeutic drugs affect the cancer cells and affect healthy non-cancerous cells. So,

© 2022 by the author(s). Distributed under a Creative Commons CC BY license.

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 11 May 2022 doi:10.20944/preprints202205.0149.v1

it is crucial to find a novel antineoplastic agent with high specificity towards cancerous cells than non-cancerous cells with reduced or minimal side effects.

Following the discovery of cisplatin, a platinum-based antineoplastic agent, the more and more organometallic compounds had been studied its biological activities. Organotin(IV) is a stanum (Sn) based compound that can form a stable bond with carbon atoms[4]. Together with a stable ligand such as dithiocarbamate, this complex can enhance its properties and lipophilic characteristic to react with cellular components [5]. Many researches have proved the potent effect of organotin(IV) dithiocarbamate compounds on various cells in in vitro models[6, 7, 8].

In this study, the cytotoxicity potency of organotin(IV) dithiocarbamate compound known as diphenyltin(IV) diisopropyl dithiocarbamate (DPDT) (**Fig. 1**) were assessed on acute lymphoblastic leukemia cells, CCL-119 (CCRF-CEM).

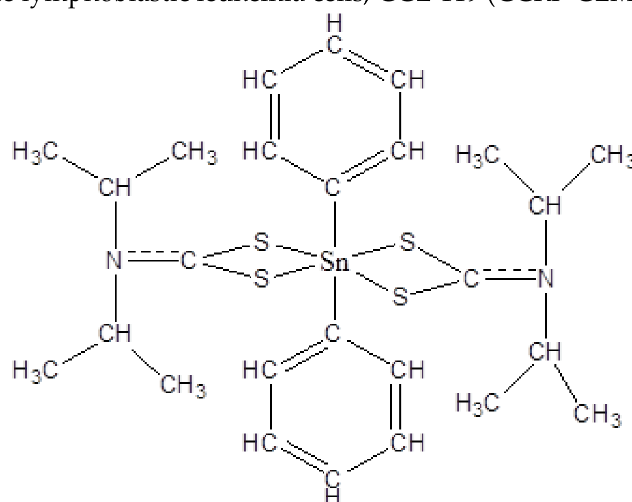


Figure 1. Diphenyltin(IV) diisopropyldithiocarbamate ($C_{26}H_{38}N_2S_4Sn$).

2. Materials And Methods

Diphenyltin(IV) diisopropyl dithiocarbamate (DPDT) compound, Dexamethasone (Nacalai Tesque, Japan), MTT powder (Sigma Aldrich, USA), Rosewell Park Memorial Institute (RPMI) 1640 Medium (Sigma Aldrich, USA), Fetal bovine serum (FBS) (Tico Europe, Netherlands), Penicillium-Streptomycin (Nacalai Tesque, Japan), Trypan blue (Sigma Aldrich, USA), Absolute ethanol, dimethyl sulphoxide (DMSO) (Sigma Aldrich, USA).

2.1. Stock preparation:

The stock concentration of diphenyltin(IV) diisopropyl dithiocarbamate was prepared at 20 mM. Exactly 0.013 g of diphenyltin(IV) diisopropyl dithiocarbamate were dissolved in 1.0 mL of dimethyl sulphoxide (DMSO). The stocks solutions were stored at 4 °C, and fresh dilutions were made by adding the medium before the experiment. The formula used to calculate the mass needed to prepare the stock solution is as below;

Mass (g) = Concentration (mM) x Volume (mL) x Relative molecular weight (g/mol)

Dexamethasone was used as a positive control in this study. The stock concentration of dexamethasone was prepared at 100 mM by dissolved in absolute ethanol. The stocks solutions were stored at 4 °C, and fresh dilutions were made by adding the medium before the experiment.

2.2. Cell culture:

The cell line was purchased from the American Type Culture Collection. Both CCRF-CEM (ATCC® CCL-119™) and WIL2-NS (ATCC® CRL-8155™) are suspension types of cells. These cells were cultured in Roswell Park Memorial Institute Medium 1640, RPMI-

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 11 May 2022 doi:10.20944/preprints202205.0149.v1

1640 with supplementation of 10% Fetal Bovine Serum (FBS) and 1% penicillin-streptomycin to obtain the complete growth medium. The cells were incubated at 37 °C with 5% CO_2 in an incubator. The cell culture was observed every day under an inverted microscope to monitor the growth and contamination. The cell was cultured in Biocompatibility and Toxicology Lab, Faculty of Health Sciences, Universiti Kebangsaan Malaysia, Kuala Lumpur.

2.3. Trypan blue exclusion (TBE) method:

Cytotoxicity evaluation was done by using the trypan blue exclusion method. The principle of trypan blue stain is relatively straight forward, where the non-viable cell will take up the blue colour while the viable cell will not. Once the cultured cell has reached the confluency, the cell was divided into three study groups which are negative control (untreated cells), positive control (cells treated with dexamethasone; concentration range: 0.625-10.0 µM) and treatment group (cells treated with DPDT; concentration range: 50-150 µM). Cell lines were seeded in 96-well culture plates with a final concentration of 5×10^4 cells/mL and incubated for 48 h. After the incubation period, the cells were treated and further incubated for 24, 48 and 72 h for reaction to take place. After the treatment period ended, the cell suspension from each well was stained with trypan blue with a dilution factor of 1:1. The viable and non-viable cells were observed and counted under an inverted microscope. The formula for percentage of cell viability was shown below;

$$\text{Percentage of cell viability}(\%) = \frac{\text{Average of viable cells}}{\text{Average of total cells (viable and non viable)}} \text{ X } 100\ \%$$

2.4. Selectivity index (SI) evaluation:

The selectivity index refers to the value of treatment selectivity towards cancerous cells and non-cancerous cells. The value of IC_{50} obtained in the TBE assay was used in this evaluation. SI more than 2 [9] showed the compound is more selective towards cancer cells than non-cancerous cells. The formula for SI was shown below;

$$\text{SI} = \frac{\text{IC}_{50} \text{ of WIL2} - \text{NS cells}}{\text{IC}_{50} \text{ of CCL} - 119 \text{ cells}}$$

2.5. Morphology observation:

The morphology of the cells was observed with different concentrations of treatment and different treatment times (12, 24 and 47 h). The cell lines were seeded in 96-well culture plates with a 5×10^4 cells/mL final concentration and incubated for 48 h. After the incubation period, the cells were treated with DPDT (IC_{50} values) and dexamethasone (highest concentration). The cells were further incubated at different time points. After the incubation period, the cell suspension was observed under an inverted microscope.

2.6. Statistical analysis:

The cytotoxicity assay was repeated for 4 times (n=4). Statistical analysis and evaluations of the percentage of viable cells and the concentration of compounds used to treat the cells were calculated using GraphPad Prism 8.0 software by employing a one-way analysis of variance (ANOVA). A p-value of < 0.05 was considered statistically significant.

3. Results

3.1. Cytotoxicity evaluation

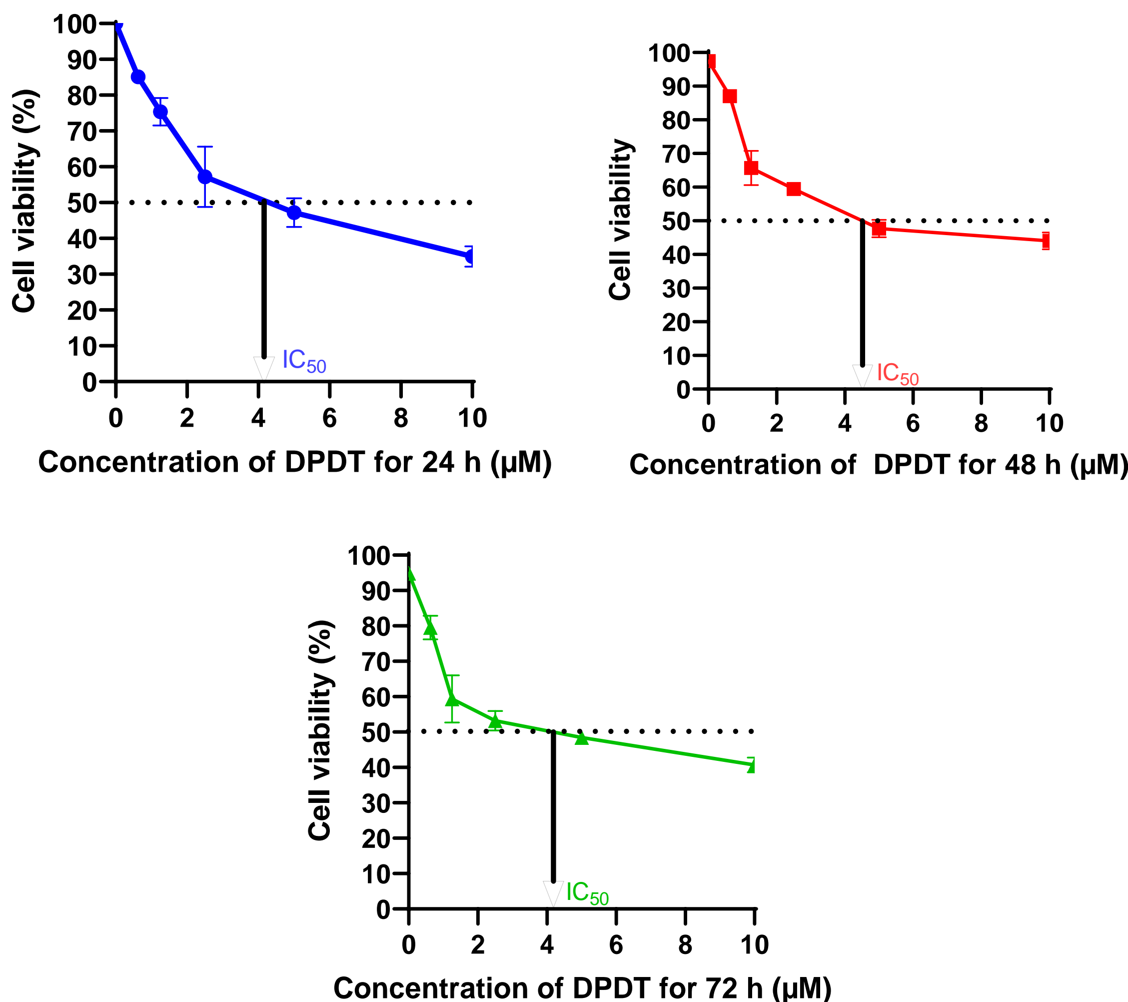
Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 11 May 2022 doi:10.20944/preprints202205.0149.v1

Table 1 showed the cytotoxic effect of DPDT towards CCL-119 and WIL2-NS cell lines at three periods of exposure (24, 48 and 72 h). The cytotoxic effect was in a dose-dependent manner. The IC_{50} values of DPDT for CCL-119 cells at 24, 48 and 72 h of treatment were 4.16 μM, 4.50 μM, and 4.18 μM, respectively. For the control cell, the IC_{50} values obtained at 24, 48 and 72 h of treatment were 8.41 μM, 3.65 μM, and 2.51 μM, respectively.

Table 1. Ic_{50} values of dpdt compound after treatment at 24, 48, and 72 h against ccl-119 and wil2-ns cells.

Treatment time (h)	**CCL-119**	**WIL2-NS**
	IC_{50} (μM)	**IC_{50} (μM)**
24	4.16 ± 0.44	8.41 ± 0.45
48	4.50 ± 0.52	3.65 ± 0.18
72	4.18 ± 0.96	2.51 ± 0.16

The graph in **Fig. 2** showed the graph of cytotoxic effects of DPDT on CCL-119 cells after 24, 48 and 72 h of treatment. The graph proved that the DPDT treatment could reduce the percentage of CCL-119 viability at all-treatment time. The highest concentration was 10 μM whereas the lowest was 0.625 μM. For 24 h of treatment, the lowest concentration showed a declining trend of cell viability compared to negative control with the value of 85.12 ± 0.51 %. The cell viability keeps decreasing to 34.97 ± 2.81 % for the highest concentration. The cell viability for DPDT treatment at 48 and 72 h also shows a similar declining trend. For the lowest concentration, the cell viability for 48 h is 87.06 ± 0.50 % while for 72 h is 79.58 ± 1.67 %. The cell viability keeps decreasing to 44.02 ± 1.25 % at a concentration of 5μM (48 h) and 40.71 ± 1.05 % at the highest concentration (72 h).



Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 11 May 2022 doi:10.20944/preprints202205.0149.v1

Figure 2. The cytotoxicity of DPDT compound against CCL-119 cells upon 24, 48 and 72 h of treatment using TBE method. Data represents the mean (± SEM) of four independent experiments (n=4).

The graph in **Fig. 3** showed the graph of cytotoxic effects of DPDT on WIL2-NS cells after 24, 48 and 72 h of treatment. The graph showed that the DPDT treatment causes WIL2-NS cell death time-dependent. The highest concentration was 10 µM whereas the lowest was 0.625 µM. For 24 and 48 h of treatment, the lowest concentration showed a declining trend of cell viability compared to negative control with a value of 91.34 ± 0.48 % and 89.38 ± 1.60 %, respectively. The cell viability keeps decreasing to 44.51 ± 1.38 % (24 h) and 31.13 ± 2.72 % (48 h) for the highest concentration. The cell viability for DPDT treatment at 72 h showed a decreasing and increasing trend. The cell viability for the lowest concentration (0.625µM) is 80.50 ± 2.30 %, decrease at 5 µM and increase to 27.97 ± 1.49 % at 10 µM.

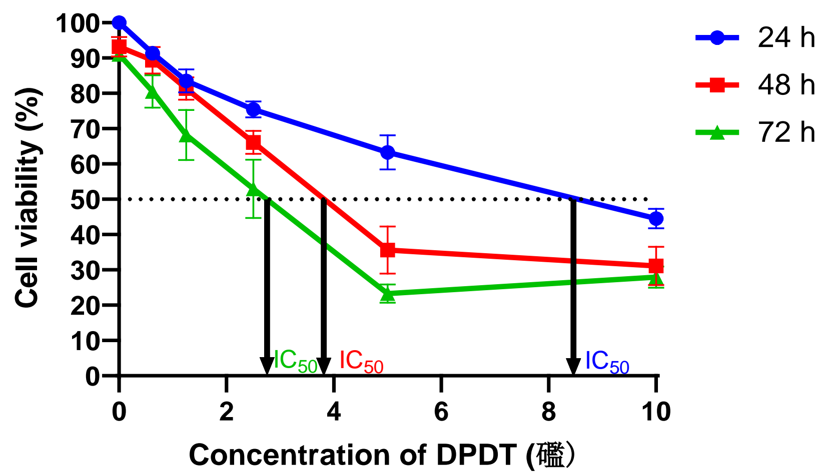


Figure 3. The cytotoxicity of DPDT compound against WIL2-NS cells upon 24, 48 and 72 h of treatment using TBE method. Data represents the mean (± SEM) of four independent experiments (n=4).

Fig. 4 showed the trend of cell viability of CCL-119 and WIL2-NS cells, respectively, treated with dexamethasone. The treatment revealed that this glucocorticoid agent caused a cytotoxic effect on both cells (CCL-119 and WIL2-NS) only at IC_{10} and IC_{25}. **Table 2** showed the summarized value of dexamethasone cytotoxic effect on CCL-119 and WIL2-NS cells for 24, 48 and 72 h of treatment time.

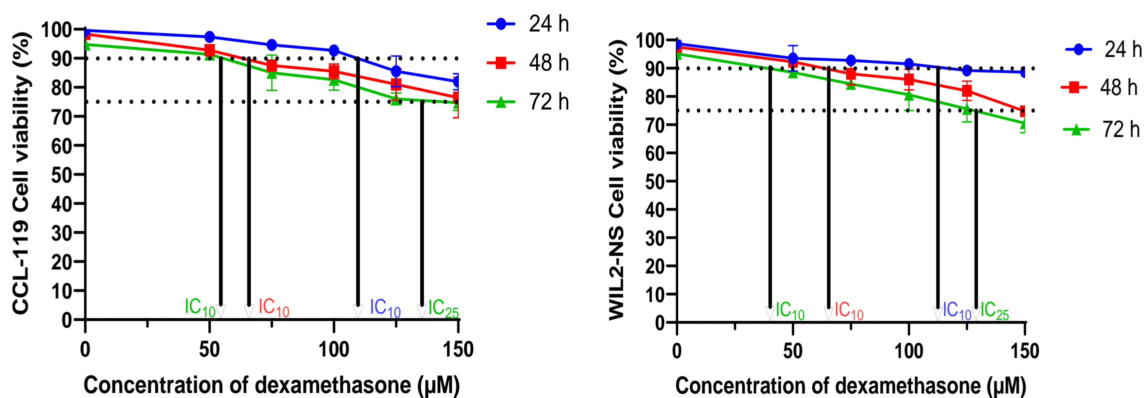


Figure 4. The cytotoxicity of dexamethasone against CCL-119 and WIL2-NS cells upon 24, 48 and 72 h of treatment using TBE method. Data represents the mean (± SEM) of four independent experiments (n=4).

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 11 May 2022 doi:10.20944/preprints202205.0149.v1

Table 2. Ic_{10} and ic_{25} values of dexamethasone after treatment at 24, 48, and 72 h against ccl-119 and wil2-ns cells.

Treatment time (h)	CCL-119		WIL2-NS	
	IC_{10} (μM)	IC_{25} (μM)	IC_{10} (μM)	IC_{25} (μM)
24	109.66 ± 3.63	-	104.62 ± 15.42	-
48	66.46 ± 6.70	-	70.35 ± 7.70	-
72	56.19 ± 7.74	133.04 ± 4.50	42.06 ± 3.08	129.97 ± 11.49

The treatment of dexamethasone on CCL-119 and WIL2-NS showed the decreasing trend of cell viability percentage for all treatment times. The highest concentration was 150 μM whereas the lowest was 50 μM. For the treatment of dexamethasone on CCL-119 cells, the lowest concentration (50 μM) shows the cell viability of 97.34 ± 0.79 %, 94.45 ± 1.54 % and 91.84 ± 2.73 % for 24, 48 and 72 h of treatment time, respectively. The percentage keep decreasing to 81.96 ± 1.36 % (24 h), 76.42 ± 3.50 % (48 h) and 71.97 ± 0.88 % (72 h) at the highest concentration. As for the treatment of dexamethasone on the control cell, WIL2-NS, the trend of cell viability is similar to CCL-119. At 50 μM, the percentage of cell viability are 93.52 ± 1.60 % (24 h), 92.26 ± 1.79 % (48 h) and 88.57 ± 1.16 % (72 h) and drop to 88.65 ± 0.92 %, 74.77 ± 0.86 % and 70.55 ± 1.68 % at concentration of 150μM for 24, 48 and 72 h, respectively.

3.2. Selectivity index (SI)

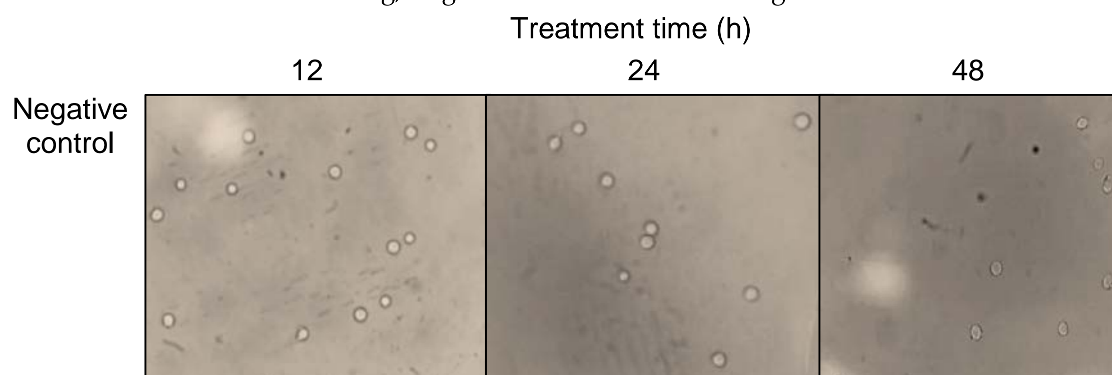
Selectivity index done based on the value of IC_{50} for CCL-119 cells and IC10 for WIL2-NS obtained from TBE assay. The evaluation shows that the treatment of DPDT at 24 h was selective towards CCL-119 cells (SI>2) and general selectivity at 48 and 72 h of treatment. Meanwhile, all treatment time, the dexamethasone treatment showed general selectivity towards CCL-119 cells (SI<2). Summary of SI obtained shown in **Table 3**.

Table 3. Si values of ccl-119 cells treated with dpdt compound and dexamethasone after treatment at 24, 48, and 72 h.

Treatment time (h)	SI	
	DPDT compound	Dexamethasone
24	2.02	0.95
48	0.81	1.06
72	0.60	0.98

Morphological observation

Fig. 5 and **6** showed the morphological changes on CCL-119 and WIL2-NS cells, respectively, treated with DPDT compound at 12, 24 and 48 h of treatment. The IC_{50} obtained for CCL-119 cells are 4.16 μM, 4.50 μM, and 4.18 μM for 24, 48 and 72 h respectively. Whereas, the IC_{50} obtained for WIL2-NS cells are 8.41 μM, 3.65 μM, and 2.51 μM for 24, 48 and 72 h respectively. The result showed the characteristic of apoptotic cell death such as cell blebbing, fragmentation and cell shrinkage.



Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 11 May 2022 doi:10.20944/preprints202205.0149.v1

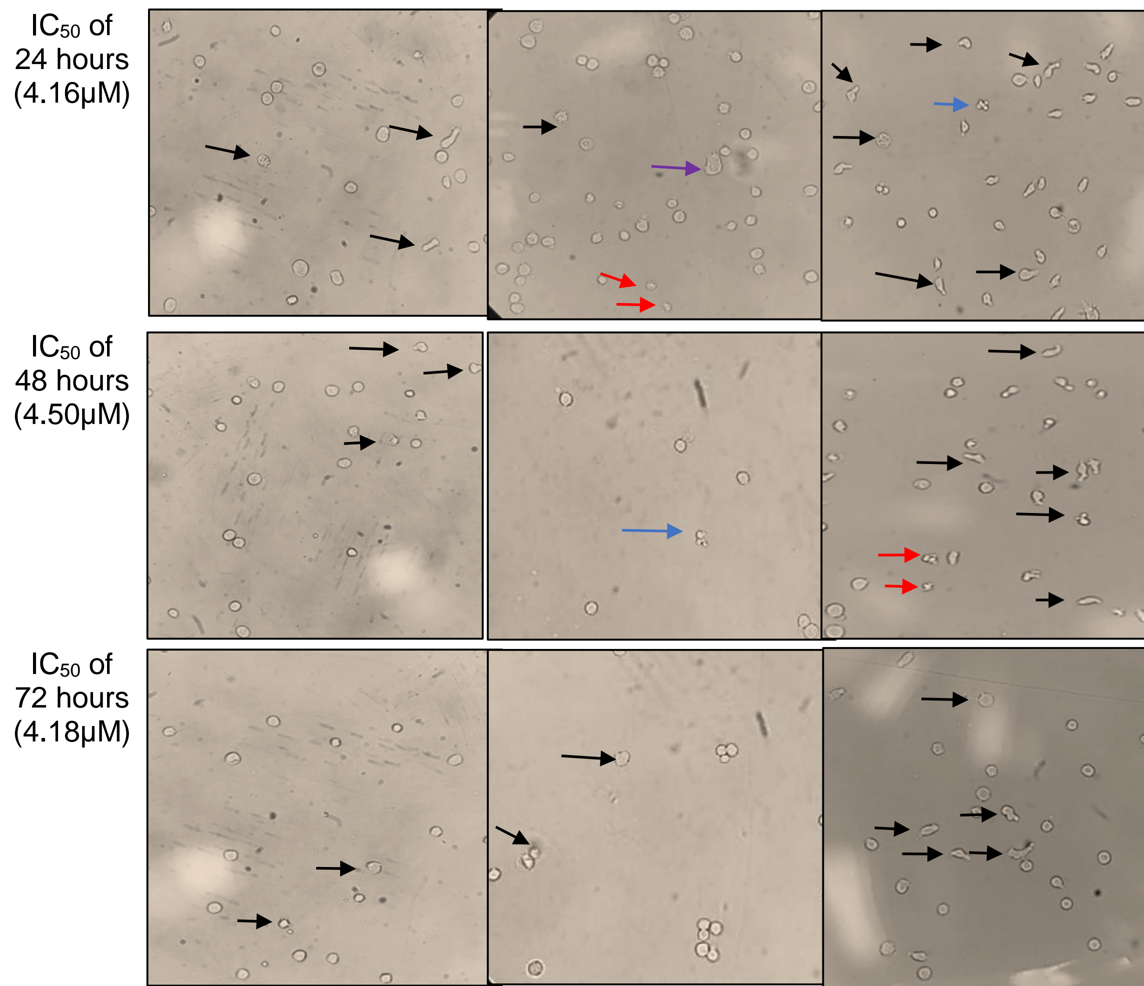
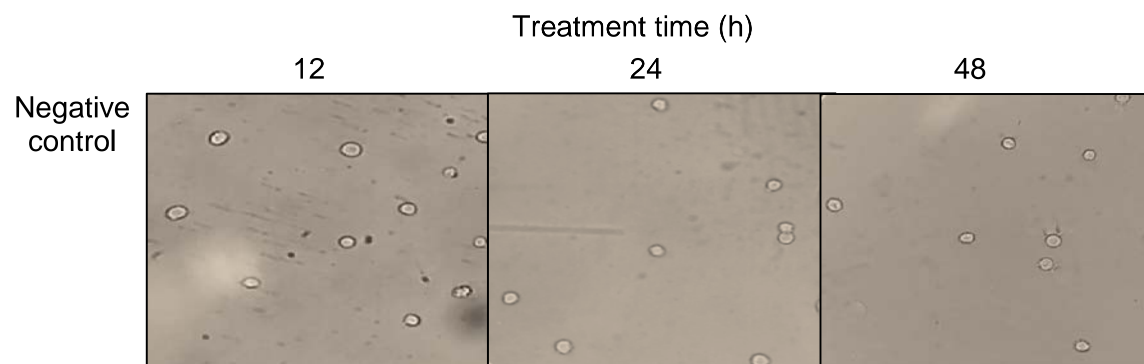


Figure 5. Morphological observation of CCL-119 cells after 12, 24 and 48 h for the negative control, treated with DPDT with IC_{50} value for 24 h (4.16µM), treated with DPDT with IC_{50} value for 48 h (4.50µM) and treated with DPDT with IC_{50} value for 72 h (4.18µM). (Magnification x40). Indicator: black arrow (cell blebbing), a blue arrow (fragmentation), a purple arrow (cell enlargement), a red arrow (cell shrinkage).



Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 11 May 2022 doi:10.20944/preprints202205.0149.v1

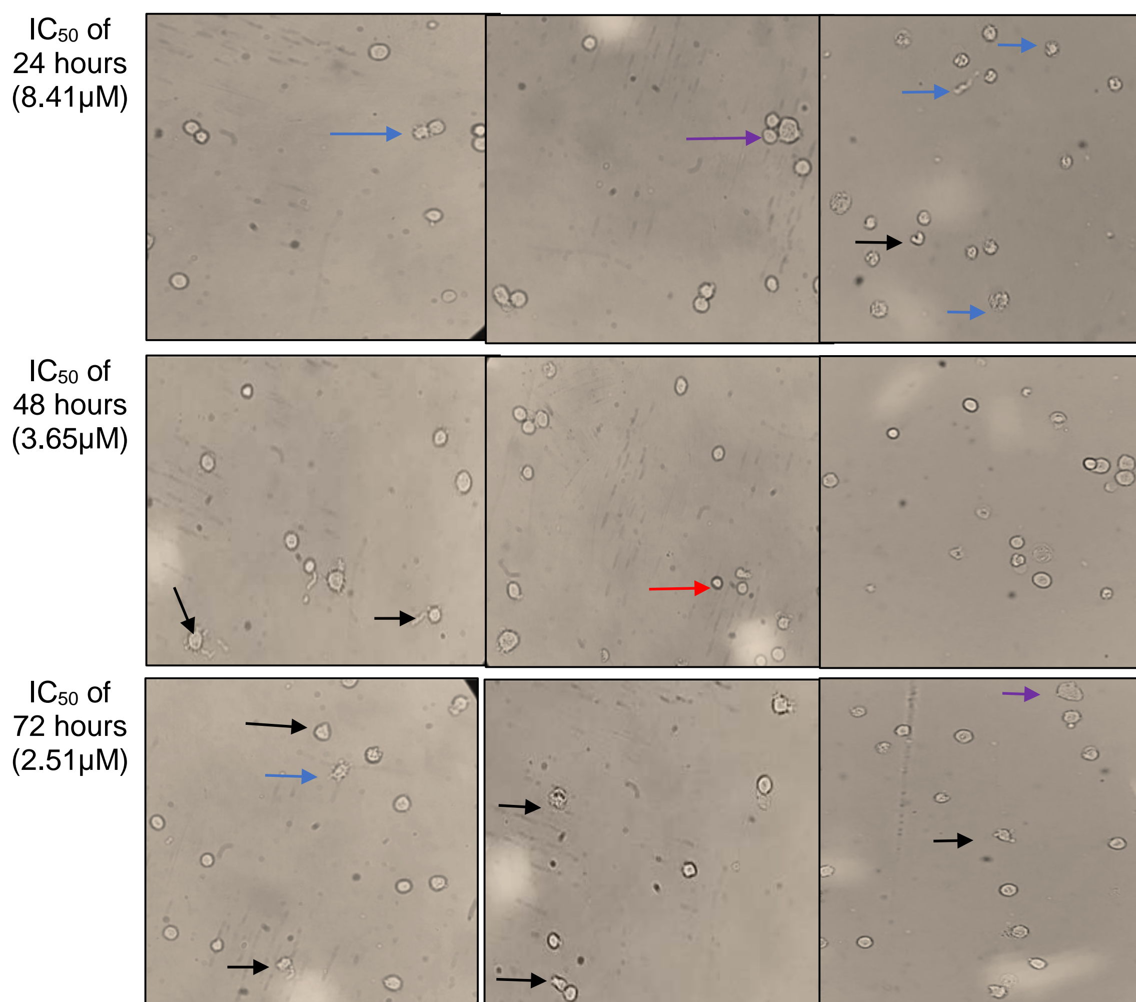
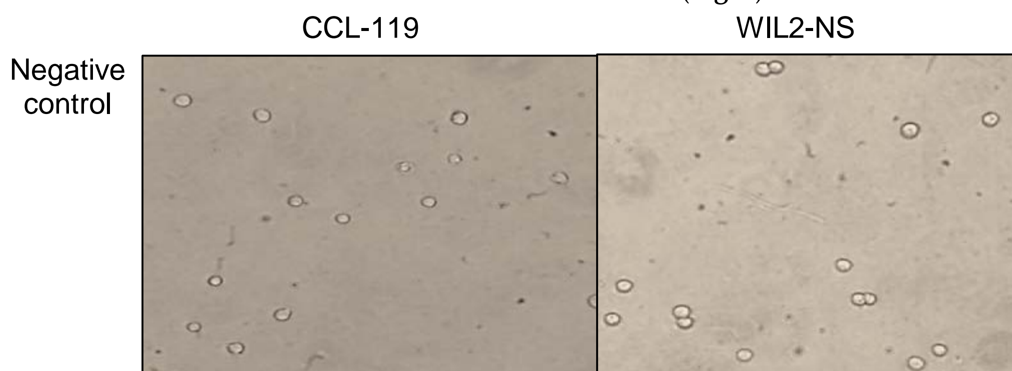


Figure 6. Morphological observation of WIL2-NS cells after 12, 24 and 48 h for the negative control, treated with DPDT with IC_{50} value for 24 h (4.16µM), treated with DPDT with IC50 value for 48 h (4.50µM) and treated with DPDT with IC_{50} value for 72 h (4.18µM). (Magnification x40). Indicator: black arrow (cell blebbing), a blue arrow (fragmentation), a purple arrow (cell enlargement), a red arrow (cell shrinkage).

For the morphological observation of the cells treated with glucocorticoid (dexamethasone), the treatment was done based on the highest concentration since the IC_{50} is undetected at 150 µM. The characteristic of apoptotic cells was observed at 12, 24 and 48 h of treatment for CCL-119 and WIL2-NS cells (**Fig. 7**).



Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 11 May 2022 doi:10.20944/preprints202205.0149.v1

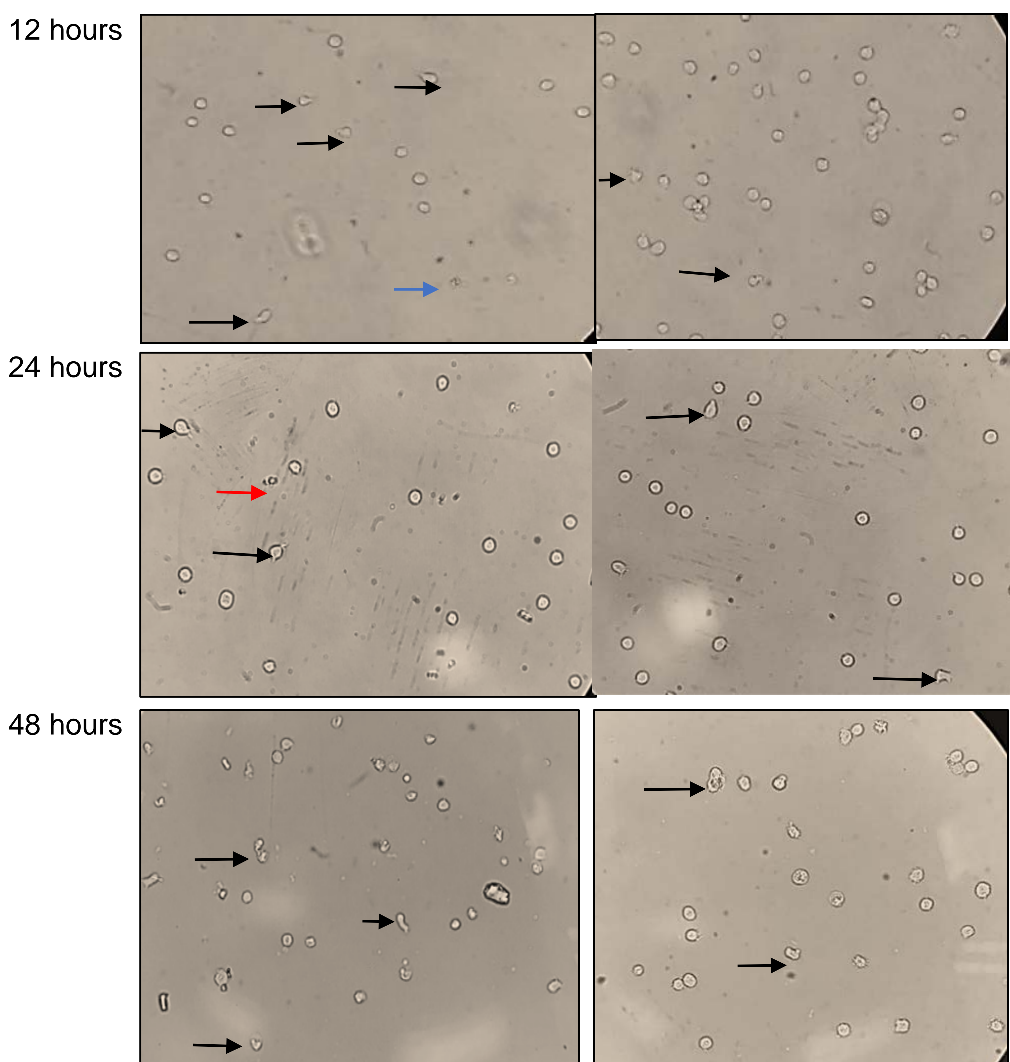


Figure 7. Morphological observation of CCL-119 and WIL2-NS cells after 12, 24 and 48 h for negative control and treated with dexamethasone at the highest concentration (150µM). (Magnification x40). Indicator: black arrow (cell blebbing), blue arrow (fragmentation), red arrow (cell shrinkage).

4. Discussion

Acute lymphoblastic leukemia (ALL) is the primary type of leukemia in childhood age and can originate from B or T cell. One of the treatments for ALL patients involved giving a glucocorticoid drug such as dexamethasone [10]. However, the usage of chemotherapeutic drugs such as dexamethasone, prednisone, methylprednisolone and hydrocortisone come along with side effect such as rashes, dizziness and emotional changes. Excessive use of dexamethasone causes resistance to the patients [11, 12]. So, there is a need in searching for new anticancer drugs with fewer side effects and are more selective towards cancer and non-cancerous cells.

The organotin compounds showed high potential to be developed as an antineoplastic agent and proved to give high cytotoxic effects compared to platinum-based organometallic complex such as cisplatin [13]. Many studies done showed that the organotin compound could inhibit cell growth at low doses. A study done by Kamaludin et al. showed that organotin(IV) N-butyl-N-phenyldithiocarbamate causes inhibitory concentration below 6 µM towards Jurkat E6.1, K562 and HL-60 cells [14].

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 11 May 2022 doi:10.20944/preprints202205.0149.v1

It was suggested that both structure of organotin and dithiocarbamate cause synergetic effects on biological structure [6]. The phenol structure attached to the tin center enhances the lipophilic properties of the organotin structure [15]. The nature of the dithiocarbamate structure produced better solubility in organic solvents than in water, thus helping to transport the compound across the cell membrane [14, 16].

Inhibitory concentration 50 or IC_{50} refers to the concentration of a substance that can inhibit 50 % of the cell population. This value has always been used to measure the efficacy of a drug. Besides that, the IC_{50} value gives information related to the potency of a drug or substance in a pharmacology study [17]. Sebaugh stated that this value could become a benchmark for drug toxicity because the lower the IC_{50}, the higher the toxicity effects [18]. Other than IC_{50} value, IC_{10} and IC_{25} also suggested being biologically significant to measure the cytotoxicity efficacy due to its impact on cells' growth and cycle [19].

The results obtained from this study indicated that DPDT could inhibit CCL-119 cells growth. Treatment of DPDT on leukemia cells, CCL-119, reduced the cell viability to 50 % after 24, 48 and 72 h of incubation with values of IC_{50} 4.16 μM, 4.50 μM and 4.18 μM, respectively. The lowest IC_{50} detected for CCL-119 cells is at 24 h of treatment. For the result of DPDT treated non-cancerous cells, WIL2-NS, the lowest IC_{50} value was detected after 72 h of incubation (2.51 ± 0.16μM). Pellerito et al. suggested that the best incubation for organotin(IV) compound are between 24 to 48 h[20].

In this study, dexamethasone was used as a positive control. Based on the TBE assay, treatment using this drug couldn't produce a value of IC_{50} on both CCL-119 and WIL2-NS cells even at the highest concentration (150 μM), and the cytotoxic effect was observed better by the treatment of DPDT. However, this treatment showed a declining trend with increasing the concentration and treatment time and producing values of IC_{10} and IC_{25}. This result was opposed to the study done by Basri et al. [21]. In that study, the result of dexamethasone on CCL-119 cells for 72 h of treatment was able to produce IC_{50} at a low dose which is between 0.5-2.5 μM. So, for this study, it was suggested to increase the concentration range since there's declining trend of cell viability but couldn't obtained IC_{50} values using current concentration range.

As an antineoplastic agent, high selectivity index (SI) is needed to produce maximum effects in cancerous cells with minimum effects in healthy non-cancerous cells [22]. In this study, the highest SI was detected at 24 h of treatment (SI=2.02). By increasing the treatment exposure period, the SI obtained is decreases (48 h=0.81; 72 h=0.60). This showed that the CCL-119 develops resistance at exposure more than 24 h and the DPDT is toxic to both cancerous and non-cancerous cells. This result was opposed to the study done by Pillai et al. where they showed that the treatment of organotin(IV) compound showed high selectivity to cancerous cells compared to non-cancerous cells[23].

The organotin(IV) compound can cause a more effective reaction on cancer cells with less toxic effects on non-cancerous cells by chemical modification[24]. This statement was supported by a study done by Tiekink[25]. In that study, the IC_{50} value obtained by treating phenyltin(IV) dithiocarbamate is 0.3 μM. However, when the chloride structure was added to the structure, the level of toxicity was reduced. With that, the organotin(IV) dithiocarbamate compound has the potential to be developed as an anti-cancer agent due to the different toxicity effects produced by adding or removing one atom structure.

The DPDT compound used in this study was suggested to induce cell death through apoptosis. Apoptosis is programmed cell death while necrosis is a mode of death that occurs due to various stressors that can cause inflammation. Morphologically, the cells that died via apoptosis showed characteristics of membrane blebbing, size shrinkage and apoptotic body formation[19]. A similar result was displayed in the study done by Awang et al. using triorganotin(IV) dithiocarbamate on Jurkat E6.1 cells[26]. Even with all these morphological changes observed, it was not enough to conclude that the DPDT compound can induce apoptosis in CCL-119 and WIL2-NS cells. So, it was suggested to do a more accurate study to detect the mode of cell death by using the Annexin V-FITC/PI staining method.

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 11 May 2022 doi:10.20944/preprints202205.0149.v1

In conclusion, the organotin(IV) dithiocarbamate compound used in this study (DPDT) was able to cause cytotoxic effects towards CCL-119 cells at low concentrations and the IC_{50} value was obtained at all time points (24, 48 and 72 h). Besides that, DPDT shows selectivity and specificity to leukemia cells compared to non-leukemia cells at a treatment time of 24 hours (SI=2.02). The positive control (dexamethasone) used was able to cause general cytotoxic and selectivity effects (SI<2) for 24, 48 and 72 h. Morphological observation shows characteristics of apoptotic cells such as cell shrinkage, blebbing, and apoptotic bodies. In conclusion, the potential of DPDT as an anti-neoplastic agent requires more detailed studies involving the molecular pathway of DPDT leading to cell death.

Acknowledgments: the authors would like to thank the Ministry of Higher Education (FRGS/1/2020/STG02/UKM/03/1) for supporting this study and special thanks to the management of the Biocompatibility and Toxicology Lab, Faculty of Health Sciences, UKM Kuala Lumpur for providing the essential laboratory facilities. The authors would also like to thank all the Faculty of Health Sciences laboratory assistants for their technical support.

Conflict of interest statement: The authors declare no conflict of interest.

REFERENCES

1. Holohan C, Van Schaeybroeck S, Longley DB, Johnston PG. Cancer drug resistance: an evolving paradigm. Nature Reviews Cancer 2013; 13(10): 714-726.
2. Cookies V, Terms A, Terms, H. 2020. Available at: http://www.cancer.org/cancer/leukemiainchildren/detailedguide/childhood-leukemia-treating-chemotherapy
3. Nurgali K, Jagoe RT, Abalo R. Frontiers in Pharmacology 2018; 1–3.
4. Ali MA, Mirza AH, Hamid MHSA, Bernhardt, PV. Diphenyltin (IV) complexes of the 2-quinolinecarboxaldehyde Schiff bases of S-methyl-and S-benzyldithiocarbazate (Hqaldsme and Hqaldsbz): X-ray crystal structures of Hqaldsme and two conformers of its diphenyltin (IV) complex. Polyhedron 2005; 24(3): 383–390.
5. Cvek B, Dvorak Z. Targeting of nuclear factor-κB and proteasome by dithiocarbamate complexes with metals. Current Pharmaceutical Design 2007; 13(30): 3155–3167.
6. Adeyemi JO, Onwudiwe DC, Singh M. Synthesis, characterization, and cytotoxicity study of organotin (IV) complexes involving different dithiocarbamate groups. Journal of Molecular Structure 2019; 1179: 366–375.
7. Odularu AT, Ajibade PA. Dithiocarbamates: challenges, control, and approaches to excellent yield, characterization, and their biological applications. Bioinorganic Chemistry and Applications 2019; 1-15.
8. Kamaludin NF, Awang N, Baba I, Hamid A, Meng, CK. Synthesis, characterization and crystal structure of organotin (IV) N-butyl-N-phenyldithiocarbamate compounds and their cytotoxicity in human leukemia cell lines. Pakistan Journal of Biological Sciences 2013; 6(1): 12-21.
9. Koch A, Tamez P, Pezzuto J, Soejarto D. Evaluation of plants used for antimalarial treatment by the Maasai of Kenya. Journal of ethnopharmacology 2005; 101(1-3): 95-99.
10. Möricke A, Zimmermann M, Valsecchi MG, Stanulla M, Biondi A, Mann G, Locatelli F, Cazzaniga G, Niggli F, Arico M, Bartram CR. Dexamethasone vs prednisone in induction treatment of pediatric ALL: results of the randomized trial AIEOP-BFM ALL 2000. The Journal of the American Society of Hematology 2016; 127(17): 2101-2112.
11. Bachmann PS, Gorman R, MacKenzie KL, Lutze-Mann L, Lock RB. Dexamethasone resistance in B-cell precursor childhood acute lymphoblastic leukemia occurs downstream of ligand-induced nuclear translocation of the glucocorticoid receptor. Blood 2005; 2519-2526.
12. Inaba H, Pui CH. Glucocorticoid use in acute lymphoblastic leukaemia. The lancet oncology 2010; 11(11):1096-106.
13. Adeyemi JO, Onwudiwe DC. Organotin (IV) dithiocarbamate complexes: Chemistry and biological activity. Molecules 2018; 23(10): 1–27.
14. Kamaludin NF, Zakaria SA, Awang N, Mohamad R, Pim NU. Cytotoxicity assessment of organotin (IV)(2-metoxyethyl) methyldithiocarbamate compounds in human leukemia cell lines. Oriental Journal of Chemistry 2017; 33(4).
15. Hussain S, Ali S, Shahzadi S, Tahir MN, Shahid M. Synthesis, characterization, biological activities, crystal structure and DNA binding of organotin (IV) 5-chlorosalicylates. Journal of Coordination Chemistry 2015; 68(14): 2369-2387.

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 11 May 2022 doi:10.20944/preprints202205.0149.v1

16. Cvek B, Dvorak Z. Targeting of nuclear factor-κB and proteasome by dithiocarbamate complexes with metals. Current Pharmaceutical Design 2007; 13(30): 3155–3167.
17. Aykul S, Martinez-Hackert E. Determination of half-maximal inhibitory concentration using biosensor-based protein interaction analysis. Analytical biochemistry 2016; 508: 97-103.
18. Sebaugh JL. Guidelines for accurate EC50/IC50 estimation. Pharmaceutical statistics 2011; 10(2): 128-134.
19. Besser JM, Wang N, Dwyer FJ, Mayer FL, Ingersoll CG. Assessing contaminant sensitivity of endangered and threatened aquatic species: Part I. Acute toxicity of five chemicals. Archives of Environmental Contamination and Toxicology 2005: 155-165.
20. Pellerito C, Nagy L, Pellerito L, Szorcsik A. Biological activity studies on organotin (IV) n+ complexes and parent compounds. Journal of Organometallic Chemistry 2006; 691(8); 1733-1747.
21. Basri DF, Nagapan TS, Latif ES. Toxicological Effect of Canarium odontophyllum Extract Against Acute Lymphoblastic Leukemia CCL-119 Cell Line. International Journal of Scientific Research in Environmental Science and Toxicology 2018; 3(1): 1–8.
22. Rashidi M, Seghatoleslam A, Namavari M, Amiri A, Fahmidehkar MA, et al. Selective Cytotoxicity and apoptosis-induction of Cyrtopodion scabrum extract against digestive cancer cell lines. International Journal of Cancer Management 2017; 10(5): 1-7.
23. Pillai V, Patel SK, Buch L, Singh VK. Applied Organometallic Chemistry 2018; 32(12): 1-12.
24. Varela-Ramirez A, Costanzo M, Carrasco YP, Pannell KH, Aguilera RJ. Cytotoxic effects of two organotin compounds and their mode of inflicting cell death on four mammalian cancer cells. Biology and Toxicology 2011: 159-168.
25. Tiekink ERT. Tin dithiocarbamates: applications and structures. Applied Organometallic Chemistry 2008; 22(9): 533–550.
26. Awang N, Yousof NSAM, Rajab NF, Kamaludin NF. In vitro cytotoxic activity of new triphenyltin (IV) alkyl-isopropyl-di-thiocarbamate compounds on human acute T-lymphoblastic cell line. Journal of Applied Pharmaceutical Science 2015; 5(1), 007-011.