Journal of Applied Pharmaceutical Science Vol. 10(02), pp 060-066, February, 2020
Available online at http://www.japsonline.com
DOI: 10.7324/JAPS.2020.102010
ISSN 2231-3354

Synthesis of new boron containing compound (CCB-2) based on curcumin structure and its cytotoxic effect against cancer cells

Edy Meiyanto[1,2*], Ratna Asmah Susidarti[1,2], Riris Istighfari Jenie[1,2], Rohmad Yudi Utomo[1,2], Dhania Novitasari[2], Febri Wulandari[2], Mitsunori Kirihata[3]

[1]Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Universitas Gadjah Mada, Yogyakarta 55281, Indonesia.
[2]Cancer Chemoprevention Research Center, Faculty of Pharmacy, Universitas Gadjah Mada, Yogyakarta 55281, Indonesia.
[3]Research Center of BNCT, Osaka Prefecture University, 1-2, Gakuen-cho, Naka-ku, Sakai, Osaka 599-8570, Japan.

ARTICLE INFO

Received on: 01/11/2019
Accepted on: 30/12/2019
Available online: 05/02/2020

Key words:
Boron containing compound CCB-2, curcumin analogue, cytotoxicity, cancer cells.

ABSTRACT

Boron containing compound (BCC) offers the potential further development for therapy against malignant cancers. We successfully synthesized a new compound based on curcumin structure (curcumin analogue) containing boron atoms, namely, CCB-2 and revealed its cytotoxic activities on various cancer cell lines. The compound was simply synthesized based on aldol condensation using acetone and 4-formylphenyl boronic acid resulted a symmetry CCB-2. The compound was then tested for cytotoxic activities in several cell lines. CCB-2 demonstrated cytotoxic effect on MCF-7/HER-2, MCF-7, RAW 264.7, and 4T1 with IC_{50} value of 12 µM, 54 µM, 26 µM, and < 10 µM, respectively, while less toxic in fibroblast cells. This compound performed superior cytotoxic against highly metastatic cancer cell, 4T1. In addition, CCB-2 induced cells accumulation in G2/M phase, but decreased the accumulation of intracellular Reactive oxygen species level in 4T1 cells. All the data suggest that this new compound is promising to be developed as anti-cancer agent rather than for Boron Neutron Capture Therapy-based cancer therapy.

INTRODUCTION

Boron containing compound (BCC) has been developed and explored for cancer therapy emphasizing its self-cytotoxic activity and as boron delivery agent for Boron Neutron Capture Therapy (BNCT). The number of BCC has increased exponentially covering the progression of new synthetic compounds and the method with the low toxicology profile (Trippier and McGuigan, 2010). BNCT is considered as rational therapy against cancer, particularly in localized cancers (Cerecetto and Couto, 2018).

An ideal boron carrier is needed for the success of this therapy, such as high accumulation in tumour tissues and low toxicity (Barth *et al*., 2018b). However, among numerous boron carriers that have been successfully designed, only two compounds that successfully entered clinical trial as boron carrier for BNCT, named sodium borocaptate (BSH) and boronophenylalanine (BPA) (Barth *et al*., 2018a). Therefore, the boron carrier development remains a major challenge to find the more selective and specific target due to the non-selective effect of BPA which highly accumulated in the kidney (Bendel *et al*., 2005; Takahara *et al*., 2015). Cancer Chemoprevention Research Center, Faculty of Pharmacy, Universitas Gadjah Mada, previously had successfully developed a BCC called Pentagamaboronon-0 (PGB-0) (Fig. 1A) which is known to have low cytotoxic activity against breast cancer cells (Kusumastuti *et al*., 2019; Utomo *et al*., 2017). In order to increase the uptake of PGB-0, several modifications have been established by adding the monosaccharide fructose and sorbitol and have been elucidated for its cytotoxic effect on breast cancer cells (Hairunisa *et al*., 2018; Hermawan *et al*., 2019; Qodria *et al*., 2018; Ramadani *et al*., 2018; Susidarti *et al*., 2019).

We then developed CCB-2 (Fig. 1B) a simpler curcumin analogue bearing boronic acid group on the benzene structure. This compound is proposed as the new candidate for anticancer, in spite of boron delivery agent for BNCT. Boron molecule itself was believed to be tolerable in biological system since it was also found in several natural products, and several studies have been reported the activities of boron compound as anticancer (Glynn *et al*., 2015;

*Corresponding Author
Edy Meiyanto, Faculty of Pharmacy, Universitas Gadjah Mada, Yogyakarta 55281, Indonesia. E-mail: edy_meiyanto@ugm.ac.id

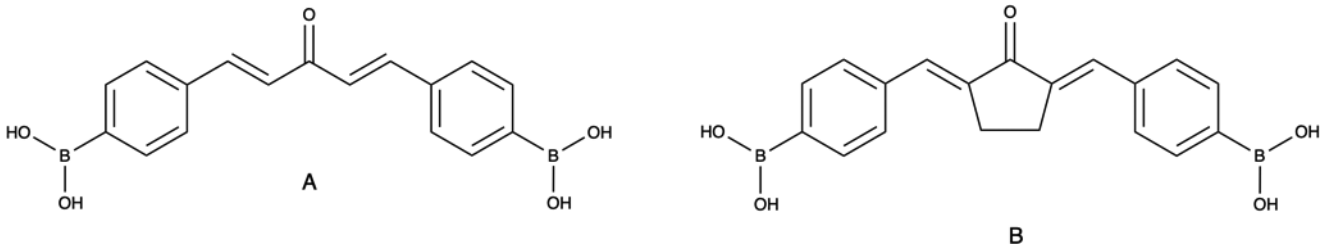


Figure 1. (A) CCB-2 and (B) PGB-0.

Jordan and Wilson, 2004). In this study, we synthesize CCB-2 with simple procedure and purification based on aldol condensation reaction. Our finding showed that the CCB-2 performed strongest cytotoxic effect toward 4T1 cells, a triple-negative breast cancer model. The cytotoxic effect seemed to correlate with the cell cycle modulation and decreasing of Reactive oxygen species (ROS) intracellular level.

MATERIALS AND METHODS

Materials and instruments

All of the materials and solvents used in the experiments were classified as analytical reagent grade unless otherwise specified. Thin-layer chromatography was conducted on glass plate silica gel 60 F254 (Merck KGaA, Darmstadt, Germany) and then visualized under Ultra Violet (UV) 254 and 366 nm. ^{1}H-Nuclear magnetic resonance (NMR) spectrum were recorded on a JMTC-400/54/SS (500 MHz, JEOL Ltd., Tokyo, Japan) spectrometer. Infra Red (IR) spectra were determined as KBr pellets of the solids on a Fourier Transform Infra Red (FTIR) spectrophotometer (Perkin Elmer, Waltham, MA). The molecular weight of the compound was elucidated using mass spectrometer (Shimadzu, Japan).

General procedure for the synthesis and purification of CCB-2

The CCB-2 compound was synthesized according to the PGB-0 synthetic procedure with slight modification (Susidarti *et al*., 2019) using solvent-free mechanochemical synthesis. The starting material consisted of 4-formylphenyl boronic acid and acetone (Scheme 1). Briefly, 4-formylphenyl boronic acid (2 mmol) was mixed with acetone (1 mmol), then homogenized using stirring. Ten µl of concentrated HCl was added drop wise and then heated at 80°C for 1.5 hours. The crude product showed as brown solid was purified using preparative thin layer chromatography with chloroform:ethyl acetate (9:1) as the mobile phase. The purified compound then elucidated by High Performance Liquid Chromatography (HPLC), UV, FTIR, Mass Spectroscopy, and ^{1}H-NMR.

Cell culture

For the study, we used 4T1, MCF-7, MCF-7/HER-2, RAW 264.7, and NIH-3T3 cells were cultured in a CO_2 incubator (37°C) in Dulbecco's modified Eagle's medium (DMEM) (Gibco, Invitrogen, USA) supplemented with 10% Fetal Bovine Serum (FBS) (Sigma, St. Louis, CA), 150 µg/ml Penicillin,-Streptomycin (Gibco, Invitrogen, USA). The cells were detached for subculture using trypsin-Ethylenediaminetetraacetic acid (EDTA) 0.25% (Gibco, Invitrogen, USA).

Cytotoxicity assay

Cytotoxicity effect of CCB-2 was evaluated using the water-insoluble colorimetric 3-(4,5-Dimethylthiazol-2-yl)-2,5-Diphenyltetrazolium Bromide (MTT) assay (Molecular Probes, Oregon) in accordance with the manufacturer's instructions. The cells were seeded into separate wells of a 96-well plate. On the next day, the cells were treated with CCB-2 with various concentrations for 24 hours. Then, 100 µl of MTT solution with final concentration 0.5 mg/ml was added and incubated for 2–4 hours depend on cells to form formazan crystal. The reaction was then stopped using Sodium dodecyl sulfate 10% with 0.01 N HCl and incubated for overnight in dark condition. The absorbance was measured on a microplate reader (Corona Electric Co., Ltd., Ibaraki-Ken, Japan) according to the manufacturer's instructions. Percentage cell viability was calculated based on the absorbance data to be determined the IC_{50} value.

Cell cycle analysis

The cell cycle distribution after treatment was assessed by flow cytometry. Cells were grown in 6-well plate 24 hours prior from the treatment. The cells were treated with CCB-2 and incubated for 24 hours. The next day, all the medium were discarded in conical, the cells were trypsinized, and centrifuged 2,000 rpm for 3 minutes. The collected cell pellets were then fixed with ethanol for 30 minutes and centrifuged again for 3 minutes. Then, the cells were washed twice with cold Phosphate Buffer Saline (PBS) and centrifuged before resuspended in propidium iodide solution (50 µg/ml in PBS containing 1% triton X-100) with DNase-free RNase A (20 µg/ml) for 15 minutes at 37°C in a dark place. Treated cells were then injected into Accuri C6 flow cytometer (BD Biosciences). The red fluorescence was measured using the FL-1 setting (log mode) after the cell debris was electronically gated out. Twenty thousand events were acquired for subsequent analyzed with in-house program (BD Biosciences).

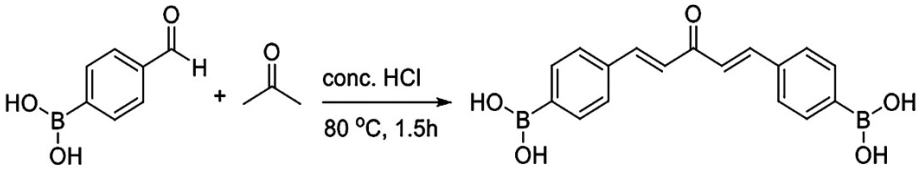
Intracellular ROS level measurement

Cells (5×10^4) were seeded on 24 well plates with DMEM culture medium (Gibco, USA) overnight. Cells were collected by trypsin-EDTA 0.25% (Gibco, USA) and then added with 500 µl 1× supplemented buffer (PBS + FBS 10%). Cells were stained by 25 µM 2′, 7′ -dichlorofluorescein diacetate (DCFDA) (Sigma, Oregon), then incubated at 37°C CO_2 5% for 30 minutes. Cells were treated with Doxorubicin 100 nM as positive control and CCB-2, then were incubated at 37°C CO_2 5% for the next 4 hours. Intracellular ROS level was measured by BD Accuri C6 flow cytometer (BD Bioscience).

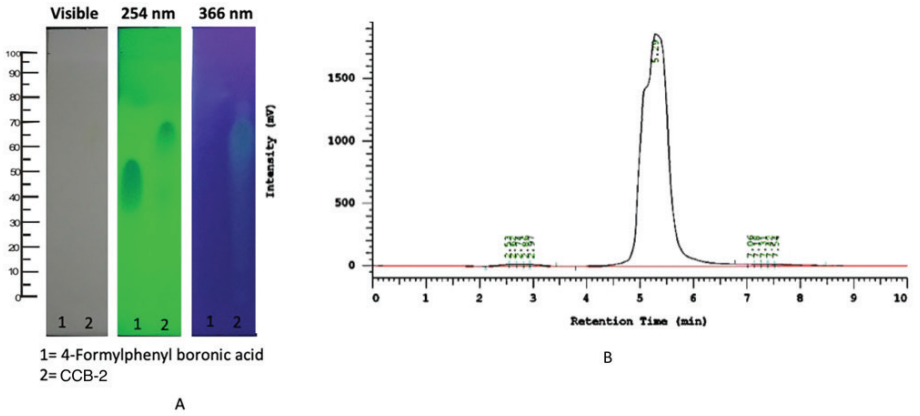
RESULTS

The preparation and synthesis of CCB-2 compound

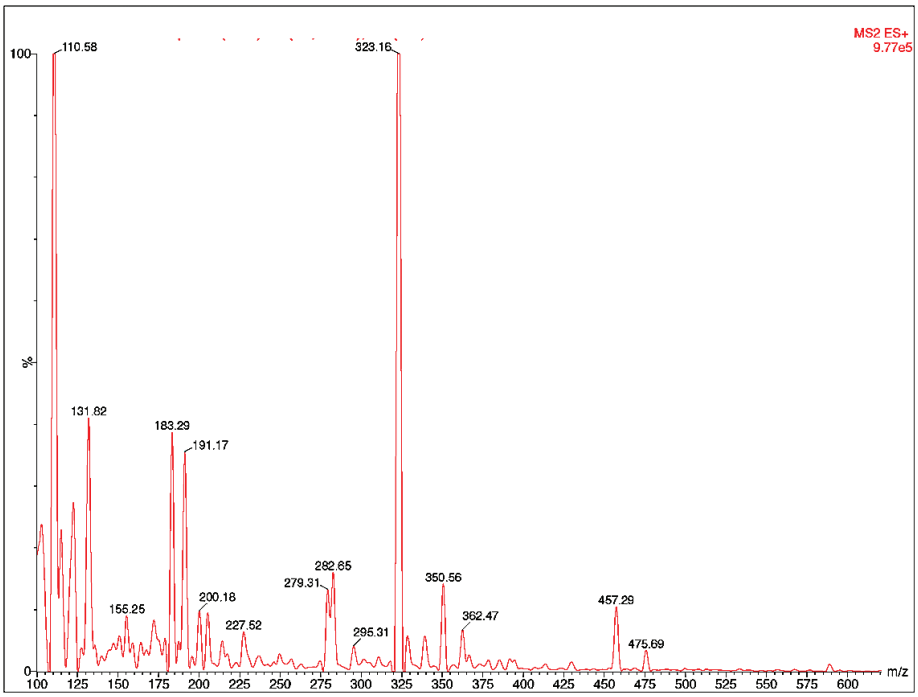
We were successfully synthesized the compound using acetone and 4-formylphenyl boronic acid at room temperature condition and purified the product by preparative Thin Layer Chromatography (TLC) and HPLC (Supplementary Figure 1). The compound of CCB-2 was obtained as a yellow powder. The molecular formula was confirmed by ESI+, which showed base ion peak at *m/z* 323.16, which confirmed with theoretical number for $C_{17}H_{16}B_2O_5$: 322.12 (Supplementary Figure 2). Based on IR analysis (Supplementary Figure 3) was reported as follows: 1,622.678 cm^{-1} (C=O), 1,338.878 cm^{-1} (B-O), 3,337.914 cm^{-1} (−OH), 1,556.264 cm^{-1} (C=C aromatic), 1,637.978 cm^{-1}



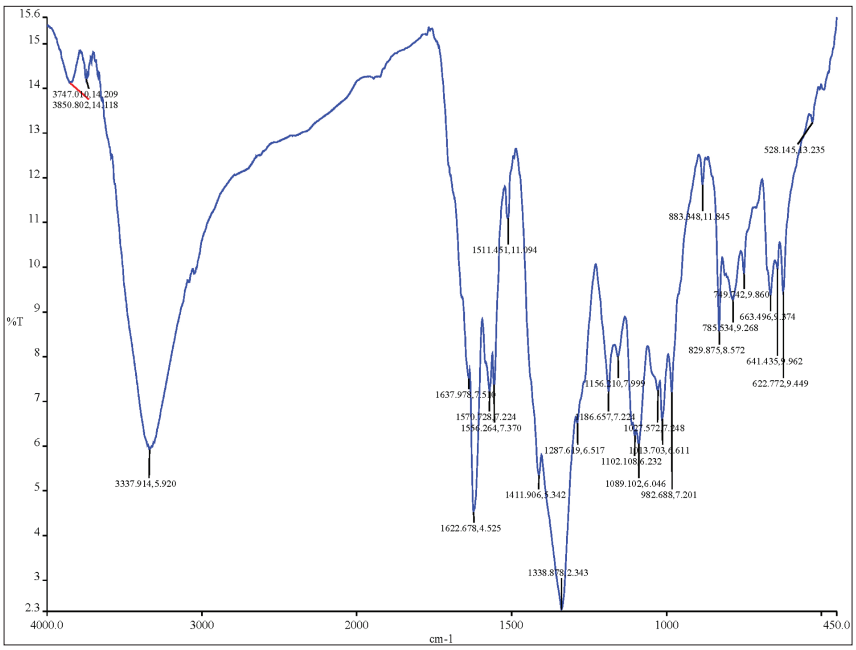
Scheme 1. Synthetic scheme of CCB-2.



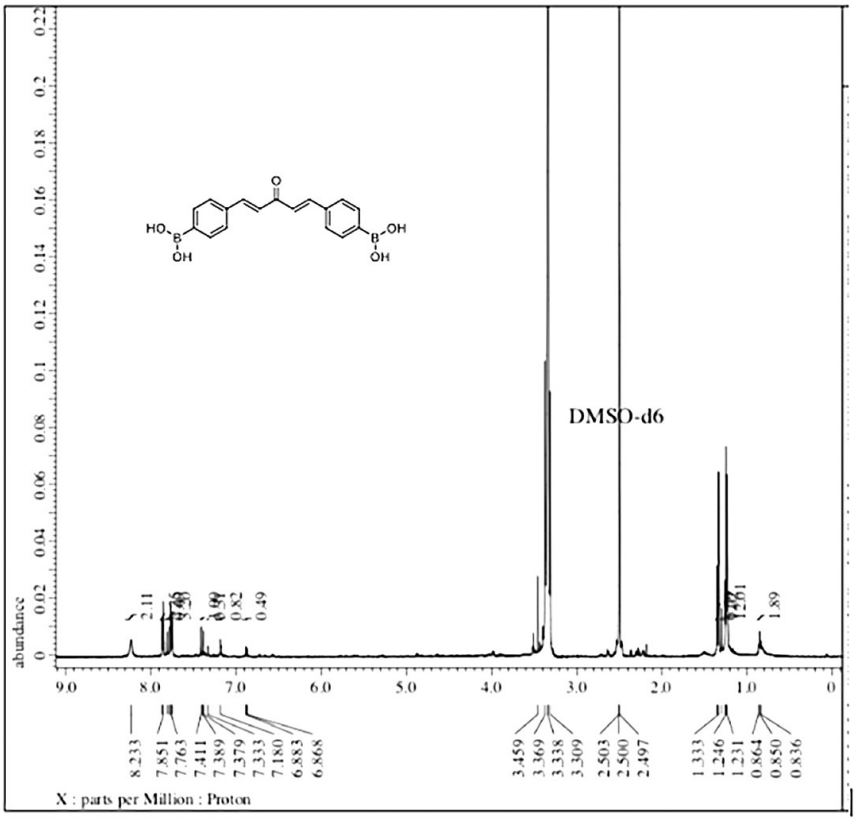
Supplementary Figure 1. (A) TLC profile using chloroform:ethyl acetate (9:1) as solvent separation, and (B) HPLC profile using acetonitrile:water (40:60) as mobile phase, C18 as stationary phase and 254 nm as the wavelength detection of CCB-2.



Supplementary Figure 2. Mass Spectrometry spectra of CCB-2. The molecular weight of CCB-2 was determined using mass spectrometer.



Supplementary Figure 3. IR spectra of CCB-2 were determined as KBr pellets of the solids on a FTIR spectrophotometer.



Supplementary Figure 4. [1]H-NMR spectra of CCB-2. [1]H-NMR spectra were recorded on a JMTC-400/54/SS spectrometer.

(C=C alkene). The NMR analysis provided [1]H-NMR (500 MHz, Dimethyl sulfoxide-d6) profile as follows: δ (ppm): δ7.4 (=CH, d, 2H); δ7.76 (=CH, J=8, dd, 4H); δ7.79 (=CH, d, 2H); δ7.86 (=CH, J=8, dd, 4H); δ 8.23 (-OH, s, 4H) (Supplementary Figure 4). The infrared spectrum markedly confirmed the CCB-2 conjugated C=O group with a strong peak of wave number at 1,622.678 cm^{-1}. Strong peak was also observed at 1,338.878 cm^{-1} representing as B-O bond. The broad and strong peak at 3,337.914 cm^{-1} was characteristic for OH group. The resonance peak at 1,556.264 cm^{-1} and 1,637.978 cm^{-1} represented as C=C aromatic and C=C

alkene, respectively. For the ^{1}H-NMR, we have to concern that CCB-2 possessed symmetrical structure, therefore, it would obtain the equivalent number of protons. The presence of OH group was shown by the singlet at δ 8.23, while two vinylic protons were represented by singlet peak with integration of 2H at δ 7.4. The presence of two doublet signals each with coupling constant of 8 Hz and 4H integration were characteristic of two *p*-di-substituted benzene rings. Based on the NMR data, we confirmed that the synthesized compound was CCB-2. The compound then was subjected to cytotoxic activities on cancer cells.

Cytotoxicity study of CCB-2 compound

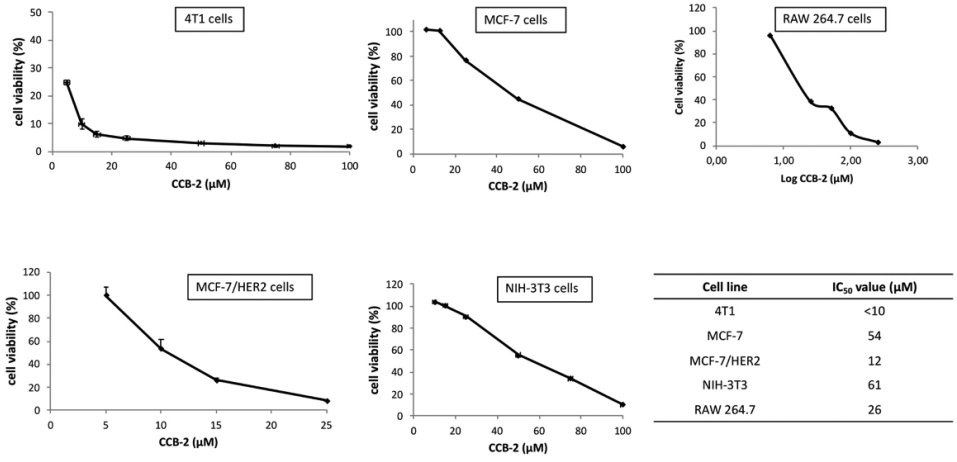
The aim of this study is to explore the anticancer activity of new boron carrier CCB-2 against cancer cells. Our results demonstrated that CCB-2 compound performed cytotoxic effect in dose dependent manner on MCF-7/HER-2, MCF-7, 4T1, and RAW 264.7 cells but performed less cytotoxic effect against NIH-3T3 cells (Fig. 2). Interestingly, CCB-2 exert cytotoxic activity more on 4T1 cells than MCF-7/HER2 since 4T1 is characterized as triple negative breast cancer (TNBC) cells (Sztalmachova *et al*., 2015). The selectivity is an important property of desired chemotherapeutic agents, by the mean a compound should exhibit higher potency against cancer cells compared to normal cells. Mathematically, selectivity is a ratio that measures the window of cytotoxicity in normal cells, with selectivity index (SI) > 3 represents good selectivity of a compound (Mahavorasirikul *et al*., 2010). Hence, we checked the cytotoxic activity of compounds on NIH-3T3 cells (fibroblast cells that represented non-cancerous cell model) and calculate their SI. The compound exhibited SI > 3 in 4T1 cells, suggesting its good selectivity and potency to be developed as a chemotherapeutic agent. We explored further concerned the molecular mechanism of CCB-2 compound on TNBC cells.

The effect of CCB-2 compound to cell cycle progression

Physiological-based cytotoxicity of CCB-2 was elucidated first by cell cycle analysis. Previously, PGB-0 was reported to induce G2/M arrest on 4T1 breast cancer cells (Kusumastuti *et al*., 2019). In this study, we examined the effect of CCB-2 on the cell cycle progression of 4T1 cells. After 24 hours of treatment, cells exposed to Dox rapidly accumulated in the G2/M phase of the cell cycle within 24 hours and subsequently underwent cell death, while the treatment with CCB-2 seemed to increase the population in the G2/M phase and induced cell death (Fig. 3), indicating that the molecular inhibitory actions of these two components differ. Furthermore, CCB-2 likely increased the population in G2/M phase within 24 hours (from 26.6% ± 1.6% at 0 hour to 29.9% ± 0.1% at 24 hour). Therefore, further investigation is needed to understand the molecular-based mechanism of this phenomenon better.

CCB-2 decreased ROS intracellular level

A relatively high level of ROS in cancer cells is the consequence of the active metabolism to generate much more energy for the physiological processes (Liou and Storz, 2010). However, ROS level in the cancer cells must be maintained in the physiologically safe level by expressing more highly of ROS. One of important factors in cancer progression is the intracellular ROS level (Kumari *et al*., 2018). An increase of intracellular ROS induces cell senescence and activates several anti-apoptotic proteins (Meiyanto *et al*., 2019). On the other hand, a decrease of ROS level may potential to develop as antioxidant and counter the side effect of doxorubicin that increases ROS levels in cells. Interestingly, the higher level of intracellular ROS over the threshold will induce cell death in cancer cells (Larasati *et al*., 2018). To find whether CCB-2 treatment correlates with ROS level, we performed oxidized DCFDA staining flow cytometry to measure intracellular ROS level on 4T1 cells. We used doxorubicin as it is been known that this anthracycline drug produces excessive ROS in cancer cells (Yokoyama *et al*., 2017). Our result showed that the treatment of CCB-2 in 4 hours decreased intracellular ROS level in 4T1 cells (Fig. 4). Moreover, in combination with doxorubicin, CCB-2 also reduced the level of intracellular ROS in cancer cells.



Cell line	IC_{50} value (μM)
4T1	<10
MCF-7	54
MCF-7/HER2	12
NIH-3T3	61
RAW 264.7	26

Figure 2. Cytotoxic effect of CCB-2 on several cells. Cells were treated by CCB-2 for 24 hours and quantified for its viability using MTT reagent as described in method. The percentage of viability cells is converted from the absorbance and determined for the IC_{50} value. The data was presented as average ± standard error (SE) (n = 3).

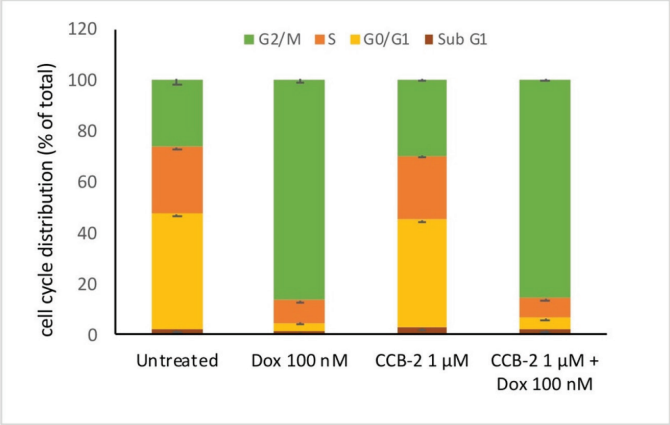


Figure 3. Cell cycle modulation of CCB-2 in 4T1 cells. The 4T1 cells (5×10^4 cells/well) were treated by CCB-2 in single and combination with Dox for 24 hours. The cells then were stained by using Propidium Iodide and subjected to a flow cytometer. The data was presented as average ± standard deviation ($n = 3$).

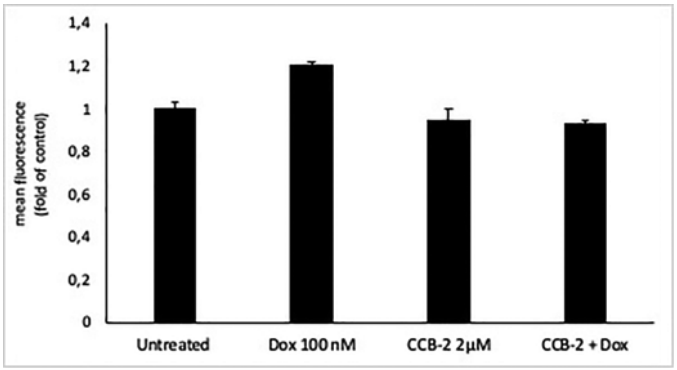


Figure 4. CCB-2 reduced the intracellular ROS level on 4T1 cells. The 4T1 cells (5×10^4 cells/well) were treated with CCB-2 in single and combination with doxorubicin (dox) for 4 hours then were subjected to ROS detection analysis by flow cytometer. The data was presented as the average ± SE ($n = 3$).

DISCUSSION

This new approach for synthesis for CCB-2 as BCC led to the effective and efficient production. The reaction steps are also relatively short and simple compared to the synthesis of BPA which required five steps reaction from 4-formylphenyl boronic acid (Harada *et al*., 2018) and PGB-0 (Susidarti *et al*., 2019) as boron-carrier for BNCT. Moreover, our compound CCB-2 constituted from relatively inexpensive and available starting materials which made it simpler to synthesize with low cost for production. Although there were some obstacle during the synthesis of CCB-2, such as the boroxin was formed during synthesis that caused low yield of the target compound, we overcome the problems by acquiring some protection for boronic acid function group by adding pinacol. The protected boronic acid function group allowed the compound to be more stable during reaction and protected during hydrolysis, and hopefully could get higher percentage yield of target compound (Occhiato *et al*., 2005; Willemse *et al*., 2017).

In regard of its cytotoxic activity, CCB-2 demonstrated its cytotoxic effect against cancer cells, with less toxicity in fibroblast cells. Interestingly, the highest cytotoxic effect of CCB-2 was found on 4T1 cells as the representative for TNBC. Furthermore, CCB-2 performed high selectivity onto cancer cells, indicating that the compound has potential usefulness in the treatment of cancer with minimal side effects on normal body cells (Hafidh *et al*., 2012; Koch *et al*., 2005; Mahavorasirikul *et al*., 2010). Previous study using PGB-0 demonstrated cytotoxic effect against TNBC 4T1 cells with IC_{50} value of 294 µM (Kusumastuti *et al*., 2019), and this current result provides higher possibility of CCB-2 to be developed as an anticancer agent. The cytotoxic effect from CCB-2 is suggested partly due to the presence of boronic acid as strong Lewis acid, yet this functional group is responsible to interact with amino acids which correlated with its activity as anticancer (Trippier and McGuigan, 2010). In addition, as any other curcumin-like structure, the CCB-2 also seem to disturb the cell cycle progression in G2/M phase, which possibly contribute to its antiproliferative activity of CCB-2 against cancer cells (Meiyanto *et al*., 2018; 2019). The well-known BCCs Bortezomib has been widely used for multiple myeloma by reversibly inhibit chymotrypsin-like activity at the 26S proteasome, leading to the activation of signaling cascades resulted in cell cycle arrest and apoptosis (Blade *et al*., 2005).

Surprisingly, the treatment using CCB-2 decreased intracellular ROS level in cancer cells instead, which indicated that the cell cycle inhibition by CCB-2 is somehow not associated with the over the threshold of intracellular ROS level. Unlike the lead compound, curcumin showed to elevate intracellular ROS level through the inhibition of several ROS metabolic enzymes in cancer cells (Larasati *et al*., 2018), and this result also supported with the latest study by Nakamae *et al*. (2019) that curcumin and its derivatives controlled ROS upregulation in tumor growth inhibition. The recent finding has been suggested that another curcumin-like structure, named Pentagamavunon (PGV)-1 revealed to specifically inhibited cell cycle progression in prometaphase stage and raised intracellular ROS level in cancer cells (Lestari *et al*., 2019). Hence, it becomes an interesting finding as CCB-2 may also possible targeted in microtubule just like curcumin and PGV-1 but not ROS metabolism. Taken together, CCB-2 is likely preferred to be developed as solely anti-cancer agent but still has the potential to be used for BNCT-based therapy. It needs to be elucidated further to know the molecular mechanism, markedly in cell cycle modulation target in order to develop new chemotherapeutic agent for cancer treatment against highly metastatic cancer.

CONCLUSION

In summary, the preparation method for CCB-2 synthesis was successfully applied. CCB-2 remains curcumin analogue with the simple structure show cytotoxic activity toward cancer cells, including TNBC cells. Therefore, CCB-2 is potential to be developed as BCC for anticancer therapy against malignant tumor.

ACKNOWLEDGEMENT

The authors express our gratitude to Flagship Basic Research for University year 2019 from Ministry of Research, Technology and Higher Education, Republic of Indonesia who funded this study with contract number of 2518/UN1.DITLIT/DIT-LIT/LT/2019.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interests.

ETHICAL ISSUES

Not Applicable.

REFERENCES

Barth RF, Mi P, Yang W. Boron delivery agents for neutron capture therapy of cancer. Cancer Commun, 2018a; 38(1):35.

Barth RF, Zhang Z, Liu T. A realistic appraisal of boron neutron capture therapy as a cancer treatment modality. Cancer Commun, 2018b; 38(1):36.

Bendel P, Margalit R, Salomon Y. Optimized 1H MRS and MRSI methods for the in vivo detection of boronophenylalanine. Magn Reson Med, 2005; 53(5):1166–71.

Blade J, Cibeira MT, Rosiñol L. Bortezomib: a valuable new antineoplastic strategy in multiple myeloma. Acta Oncol, 2005; 44(5): 440–8.

Cerecetto H, Cuoto M. Medicinal chemistry of boron-bearing compounds for BNCT- Glioma treatment: current challenges and perspectives. Glioma - Contemporary Diagnostic and Therapeutic Approaches, 2018. [ONLINE] Available via https://www.intechopen.com/books/glioma-contemporary-diagnostic-and-therapeutic-approaches/medicinal-chemistry-of-boron-bearing-compounds-for-bnct-glioma-treatment-current-challenges-and-pers (Accsessed 17 October 2019).

Glynn SJ, Gaffney KJ, Sainz MA, Louie SG, Petasis NA. Molecular characterization of the boron adducts of the proteasome inhibitor Bortezomib with epigallocatechin-3-gallate and related polyphenols. Org Biomol Chem, 2015; 13(13):3887–99.

Hafidh RR, Abdulamir AS, Bakar FA, Jalilian FA, Abas F, Sekawi Z. Novel molecular, cytotoxical, and immunological study on promising and selective anticancer activity of Mung bean sprouts. BMC Complement Altern Med, 2012; 12(1):208.

Hairunisa I, Utomo RY, Ertanto Y, Jenie RI, Meiyanto E. Pentagamaboronon-0 fructose inhibited migration and overexpression of matrix metalloproteinases 9 on MCF-7/HER2 breast cancer cells. Indones J Cancer Chemoprevent, 2018; 9(3):134–42.

Harada S, Kajihara R, Muramoto R, Jutabha P, Anzai N, Nemoto T. Catalytic asymmetric synthesis of α-methyl-p-boronophenylalanine. Bioorg Med Chem Lett, 2018; 28(10):1915–8.

Hermawan A, Susidarti RA, Ramadani RD, Qodria L, Utomo RY, Ishimura M, Hattori Y, Ohta Y, Kirihata M, Meiyanto E. Cellular uptake evaluation of pentagamaboronon-0 (PGB-0) for boron neutron capture therapy (BNCT) against breast cancer cells. Invest New Drugs, 2019; 37(6):1292–9.

Jordan MA, Wilson L. Microtubules as a target for anticancer drugs. Nat Rev Cancer, 2004; 4(4):253–65.

Koch A, Tamez P, Pezzuto J, Soejarto D. Evaluation of plants used for antimalarial treatment by the Maasai of Kenya. J Ethnopharmacol, 2005; 101(1–3):95–9.

Kumari S, Badana AK, Malla R. Reactive oxygen species: a key constituent in cancer survival. Biomark Insights, 2018; 13:1–9.

Kusumastuti R, Utomo RY, Khumaira A, Putri H, Jenie RI, Meiyanto E. Pentagamaboronon-0 increased cytotoxicity of and inhibited metastasis induction by doxorubicin in breast cancer cells. J App Pharm Sci, 2019; 9(06):43–51.

Larasati YA, Yoneda-Kato N, Nakamae I, Yokoyama T, Meiyanto E, Kato J. Curcumin targets multiple enzymes involved in the ROS metabolic pathway to suppress tumor cell growth. Sci Rep, 2018; 8(1):2039.

Lestari B, Nakamae I, Yoneda-Kato N, Morimoto T, Kanaya S, Yokoyama T, Shionyu M, Shirai T, Meiyanto E, Kato JY. Pentagamavunon-1 (PGV-1) inhibits ROS metabolic enzymes and suppresses tumor cell growth by inducing M phase (prometaphase) arrest and cell senescence. Sci Rep, 2019; 9(1):1–12.

Liou G-Y, Storz P. Reactive oxygen species in cancer. Free Radic Res, 2010; 44(5):479–96.

Mahavorasirikul W, Viyanant V, Chaijaroenkul W, Itharat A, Na-Bangchang K. Cytotoxic activity of Thai medicinal plants against human cholangiocarcinoma, laryngeal and hepatocarcinoma cells in vitro. BMC Complement Altern Med, 2010; 10(55):1–8.

Meiyanto E, Putri H, Larasati YA, Utomo RY, Jenie RI, Ikawati M, Lestari B, Yoneda-Kato N, Nakamae I, Kawaichi M, Kato JY. Anti-Proliferative and anti-metastatic potential of curcumin analogue, pentagamavunon-1 (PGV-1), toward highly metastatic breast cancer cells in correlation with ROS generation. Adv Pharm Bull, 2019; 9(3):445–52.

Meiyanto E, Septisetyani EP, Larasati YA, Kawaichi M. Curcumin analog pentagamavunon-1 (PGV-1) sensitizes Widr cells to 5-Fluorouracil through inhibition of NF-κB activation. Asian Pac J Cancer Prev, 2018; 19(1):49–56.

Nakamae I, Morimoto T, Shima H, Shionyu M, Fujiki H, Yoneda-Kato N, Yokoyama T, Kanaya S, Kakiuchi K, Shirai T, Meiyanto E. Curcumin derivatives verify the essentiality of ROS upregulation in tumor suppression. Molecules, 2019; 24(22):4067.

Occhiato EG, Lo Galbo F, Guarna A. Preparation and Suzuki−Miyaura coupling reactions of tetrahydropyridine-2-boronic acid pinacol esters. J Org Chem, 2005; 70(18):7324–30.

Qodria L, Hairunisa I, Utomo RY, Hermawan A, Meiyanto E. Anti-metastatic activity of curcumin analog pentagamaboronon-0-sorbitol against HER2-overexpressed MCF-7 breast cancer cells. Indones J Cancer Chemoprevent, 2018; 9(3):118–25.

Ramadani RD, Utomo RY, Hermawan A, Meiyanto E. Curcumin analog pentagamaboronon-0-sorbitol inhibits cell migration activity of triple negative breast cancer cell line. Indones J Cancer Chemoprevent, 2018; 9(3):126–33.

Susidarti RA, Utomo RY, Qodria L, Ramadani RD, Ohta Y, Hattori Y, Kirihata M, Meiyanto E. Preparation of pentagamaboronon-0 and its fructose and sorbitol complexes as boron carrier for boron neutron capture therapy (BNCT) application. Res Pharm Sci, 2019; 14(4):286–92.

Sztalmachova M, Gumulec J, Raudenska M, Polanska H, Holubova M, Balvan J, Hudcova K, Knopfova L, Kizek R, Adam V, Babula P. Molecular response of 4T1-induced mouse mammary tumours and healthy tissues to zinc treatment. Int J Oncol, 2015; 46(4):1810–18.

Takahara K, Inamoto T, Minami K, Yoshikawa Y, Takai T, Ibuki N, Hirano H, Nomi H, Kawabata S, Kiyama S, Miyatake SI. The anti-proliferative effect of boron neutron capture therapy in a prostate cancer xenograft model. PloS One, 2015; 10(9):e0136981.

Trippier PC, McGuigan C. Boronic acids in medicinal chemistry: anticancer, antibacterial and antiviral applications. Med Chem Comm, 2010; 1(3):183–98.

Utomo RY, Putri H, Pudjono P, Susidarti RA, Jenie RI, Meiyanto E. Synthesis and cytotoxic activity of 2,5-Bis(4-Boronic acid) benzylidine cyclopentanone on HER-2 overexpressed-cancer cells. Indones J Pharm, 2017; 28(2):74.

Willemse T, Schepens W, van Vlijmen HW, Maes BU, Ballet S. The Suzuki–Miyaura cross-coupling as a versatile tool for peptide diversification and cyclization. Catalysts, 2017; 7(3):74.

Yokoyama C, Sueyoshi Y, Ema M, Mori Y, Takaishi K, Hisatomi H. Induction of oxidative stress by anticancer drugs in the presence and absence of cells. Oncol Lett, 2017; 14(5):6066–70.

How to cite this article:
Meiyanto E, Susidarti RA, Jenie RI, Utomo RY, Novitasari D, Wulandari F, Kirihata M. Synthesis of new boron containing compound (CCB-2) based on curcumin structure and its cytotoxic effect against cancer cells. J Appl Pharm Sci, 2020; 10(02):060–066.