DOI: 10.21776/ub.jpacr.2022.11.02.646 *J. Pure App. Chem. Res.*, **2022**, 11 (2), 149-165
31 August 2022

Secondary Metabolites of *Nicotiana tabacum* and Their Biological Activities: A Review

Devi Anggraini Putri,[1]* Riyadatus Solihah,[1] Rianur Oktavia,[1] Dwi Aprilia Anggraini,[1] and Sri Fatmawati[2]

[1]Department of Medical Laboratory Technology, Sekolah Tinggi Ilmu Kesehatan Ngudia Husada Madura, Bangkalan, Indonesia
[2]Department of Chemistry, Faculty of Science and Data Analytics, Institut Teknologi Sepuluh Nopember Surabaya, Indonesia

*Corresponding email: devi@stikesnhm.ac.id

Received 10 November 2021; Accepted 31 August 2022

ABSTRACT

Nicotiana tabacum (tobacco) is one of the most commercially farming products in the world. Indonesia is one of the top ten tobacco producing countries in the world since 1990's. Based on literature studies, tobacco has the main secondary metabolites such as cembranoids and flavonoids. They have fine biological activities as antiviral, antimicrobial, antioxidant, anti-HIV, anti-proliferative, anti-inflammatory, anti-parasitic, anti-termite, and cytotoxicity effects. Therefore, *N. tabacum* is potential for further investigation on natural products research. This review aims to provide scientific evidences related to structure activity relationship between secondary metabolites and biological activities of *N. tabacum*.

Key word: *Nicotiana tabacum*, secondary metabolite, bioactivity

INTRODUCTION

Herbal medicines are commonly plant-derived substances that have been used to treat illness traditionally since the ancient period [1]. Because of a huge number of plants in Indonesia, some people have used them as their alternative medicines called *Jamu*. Although there are about 30.000 plant species in Indonesia, only 5000 species have been identified as medicinal plants [2]. For this reason, the discovery of *Jamu* will be a big opportunity for future standardized herbal medicines and phytomedicines research [3], [4], [5]. Scientifically, *Jamu* is very potent for treatment of diseases caused by free radical [6], bacteria [7], virus [8] as well as metabolic disorder [9]. However, scientific evidences of *Jamu* is not development significantly compared traditional Chinese medicine (TCM) and ayurveda from India.

Secondary metabolites and its biological activities from natural plants has an important role as scientific data to find a new drug. Through secondary metabolites, the chemical structures might be known as phytochemical sources to finding new drugs. Furthermore, the report of biological activity is used to know the pharmacological effects and study the structure activity relationship (SAR). In the past decade, our research has been focused on scientific data of herbal medicines based on secondary metabolites and their biological activities. The results showed that many plants are potent to be evaluated their biological activities as well as the chemical constituents [4], [10], [11], [12], [13], [14], [15]. Leading to new drug discovery, the secondary metabolite and its biological activity from other potent plants is urgently needed. In this review, we focus on *Nicotiana tabacum*, one of the most commercially cultured plants

The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

in the world [16]. Furthermore, Indonesia is one of the top ten tobacco producing countries in the world since 1990's [17], [18].

N. tabacum, recognized as tobacco, is plant species of Solanaceae family. Historically, *N. tabacum* was known as medical treatment after nicotine was successfully identified from tobacco leaves. In the nineteenth century, nicotine salicylate was prepared as a treatment for scabies [19]. Moreover, nicotine was also used to treat patient of Parkinson's disease. Although the therapeutic effects of *N. tabacum* have been known since the mid of nineteenth century, only a limited case of studies have been identified on their chemical constituents and pharmacological effects. Based on our literature studies, *N. tabacum* is rich of secondary metabolite compounds such as cembranoids [20], [21], flavonoids [22], [23], [24], alkaloids [25], [26], and terpenoids [27], [28], [29], [30]. Furthermore, either extracts or isolated compounds of *N. tabacum* had been reported its biological activities as antiviral [31], anti-tobacco mosaic virus (anti-TMV) [22], [23], [29], [30], [32], [33], [34], [35], [36], [37], antibacterial [38], [39], [40], antioxidant [41], [42], [43], cytotoxicity effects [44], [45], anticancer [21], [46], antitumor [20], [27], antipoliferative [47], anti-inflammatory [48], anti-HIV [24], antitermite [49], and antiparasitic [25]. Therefore, this review aims to provide scientific evidences of *N. tabacum* for further investigation.

TAXONOMY AND BOTANICAL STUDY OF *N. tabacum*

N. tabacum is widely cultivated around the world on tropical and subtropical regions especially in Indonesia. They spread easily in large area such as in forests, mountains, dry valleys, wetlands, and savannahs as well. Based on its taxonomy's report, *N. tabacum* belongs to Plantae kingdom; order Solanaceae; family Solanaceae; genus Nicotiana [50]. Among the numerous species of Nicotiana genus, only *N. tabacum* and *N. rustica* are of preferred to economic crop. Furthermore, the leaf is the most commercial parts and a dry period is better time for harvesting the leaves. Its leaves are green oval-shaped with a length of 50 cm. The height of *N. tabacum* is about 1.5-2 m. This plant has branching stems and short flowers. Its limbs are white, pink, or reddish colors. The morphological characteristics of *N. tabacum* is presented in **Figure 1**.

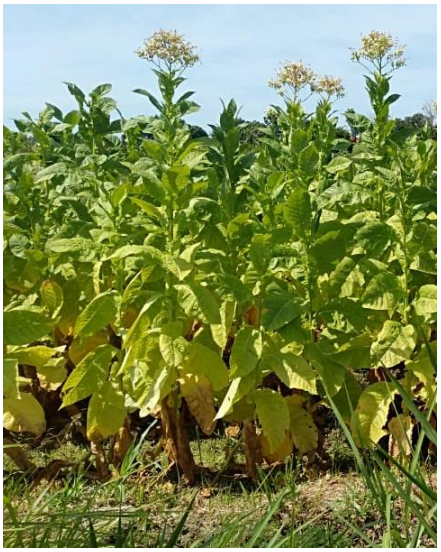


Figure 1. *Nicotiana tabacum* (tobacco)

The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

SECONDARY METABOLITES OF *N. tabacum*

Secondary metabolites are chemical constituents that have basically carbon skeleton structures called compounds that have some biological activity effects [51], [52], [53]. They are commonly produced from plants, bacteria as well as fungi. The secondary metabolites can protect plants against stresses in its ecosystems. So, the secondary metabolites might have some biological activities for drug discovery. The previous research reported that *N. tabacum* is rich of secondary metabolites from phenolics, terpenoids, and alkaloids group. In this review, the secondary metabolites of *N. tabacum* are classified based on its biological activity.

As presented in **Table 1**, several chemical constituents had been reported their biological activities as antiparasitic, anti-HIV-1, cytotoxicity, anticancer, anti-tobacco mosaic virus, and antitumor. There are three compounds from alkaloids groups as antiparasitic against *R. sanguineus* tick larvae. They are (*S*)-nornicotine **(1)**, (*R*)-anatabine dihydrochloride **(2)**, and (*S*)-anatabine dihydrochloride **(3)**. Among them, compound **(2)** is highest inhibitory activity at concentration of 300 µmol [25]. Cembranoid (1*S*,2*E*,4*R*,6*E*,8*R*,11*S*)-8,11-epoxy-2,6,12(20)-cembratriene-4-ol **(4)** showed moderate anticancer against HepG2 cell line with an IC_{50} of 14.38 µM [20]. There are three compounds that exhibited cytotoxicity against C8166 cells as well as anti-HIV-1 activity. They are nicotphenol (A) **(5)**, (B) **(6)**, and (C) **(7)**. Among them, compound **(5)** showed fine cytotoxicity (CC_{50}) and also anti-HIV-1 (EC_{50}) with value of 239.4 and 1.86 µg/mL, respectively [24].

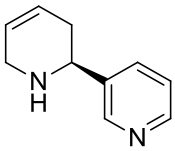
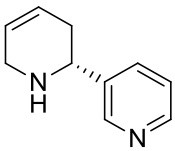
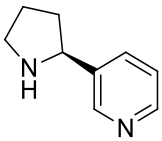
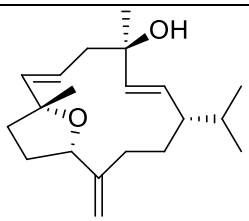
In the past decade, two cembranoids were also reported as antimigratory against prostate cancer cell line namely PC-3M-CT in dose of 10 µM and PC-3 in dose of 50 Mm [21]. They are (1*S*,2*E*,4*R*,6*R*,7*E*,11*E*)-2,7,11-cembratriene-4-*O*-methyl-4,6-diol **(8)** and (1*S*,2*E*,4*R*,6*R*,7*S*,8*S*,11*E*)-7,8-epoxy-2,11-cembradiene-4-*O*-methyl-4,8-diol **(9)**. Recently, most of flavones and sesquiterpenes from *N. tabacum* have been reported as anti-tobacco mosaic virus (anti-TMV) by using the half-leaf method. There are about 20 compounds which have potential as anti-TMV. They are 5,7-dihydroxy-2'-methoxy-6-methyl-flavone **(10)**, 5-hydroxy-2',7-di-methoxy-6-methyl-flavone **(11)** [32], pisatin **(12)**, turbinataphenol A **(13)** [44], methyl-2-hydroxy-4-isopropyl-7-methoxy-6-methylnaphthalene-1-carboxylate **(14)**, methyl-2-hydroxy-6-(hydroxymethyl)-4-isopropyl-7-methoxynaphthalene-1-carboxylate **(15)**, lacinilene C **(16)** [27], nicosesquiterpene A **(17)**, nicosesquiterpene B **(18)** [28], tabasesquiterpene B **(19)**, 3-(6-methoxy-3-oxo-1,3-dihydroisoben-zofuran-5-yl)-3-oxopropyl acetate **(20)**, nicotpanoid A **(21)** [36], 4'8-dihydroxy-6,7-dimethoxyisoflavone **(22)**, 4'-6-dihydroxy-8-methoxycarbonyl-7-methoxyisoflavone **(23)**, 4',7-dimethoxy-8-hydroxymethyl-6-hydroxyisoflavone **(24)** [22], tababiphenyl C **(25)**, tababiphenyl E **(26)** [35], tabaisocoumarin B **(27)** [33], tabamide A **(28)** [34], 14-noreudesmane sesquiterpene **(29)** [29], and licoisoflavone **(30)** [23]. Among 20 compounds, compound **(25)** and **(30)** have fine inhibitory activity with value of 48.4% compared with ningnanmycin as a positive control.

Furthermore, the secondary metabolites of *N. tabacum* also reported their cytotoxicities inhibit five cancer cell lines such as NB4, A549, PC3, SHSY5Y, and MCF7 [27], [44], [45]. There are three compounds exhibited inhibitory activity against NB4 namely compound **(15)**, (-)-dehydrodiconiferyl alcohol **(31)**, and nigrolineabiphenyl A **(32)**. Compound **(31)** showed lowest inhibition with IC_{50} of 5.5 µM. It means the compound **(31)** has a good potential as bioactive substance. Moreover, it might be caused a furan ring and hydroxyl (-OH) group as active sides. For inhibitory activity against A549, there are three compounds potentially such as compound **(13)**, **(15)**, and tababiphenyl I **(33)**. Among them, the compound **(33)** showed lowest inhibition with IC_{50} of 2.8 µM. Furthermore, compound **(16)**, 5-methoxy-22-dimethyl-2H-furo[34-g]-chromen-8(6H)-one **(34)**, and tababiphenyl G **(35)** exhibited inhibitory activity

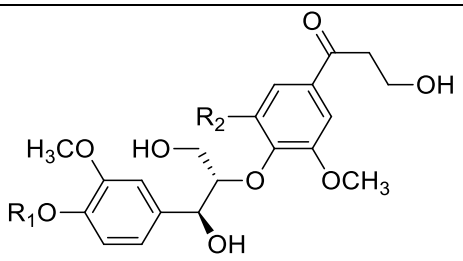
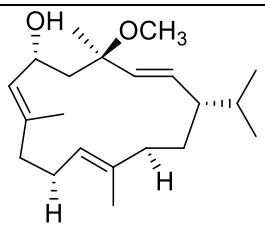
The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

against SHSY5Y. Compound **(35)** has potential as bioactive substance because of the lowest IC_{50} value of 3.8 µM. In addition, there are three compounds potentially inhibitor PC3 namely compound **(13)**, compound **(33)**, and methyl-2-hydroxy-4-isopropyl-7-methoxy-6-methylnaphthalene-1-carboxylate **(36)**. Among them, compound **(33)** exhibited lowest inhibitory activity against PC3 with IC_{50} value of 3.7 µM. For inhibitory activity against MCF7, there are four compounds potentially such as compound **(13)**, **(14)**, **(15)**, and nigrolineabiphenyl B **(37)**. Among them, compound **(13)** showed lowest IC_{50} with value of 4.8 µM. From those reports, compound **(33)** has strongly potential as inhibitor A549 and PC3 as well. It might be affected by the bioactive substituents such as hydroxyl and methoxy groups.

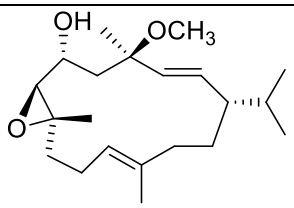
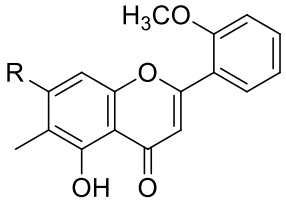
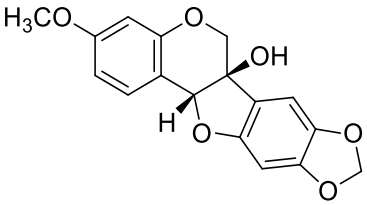
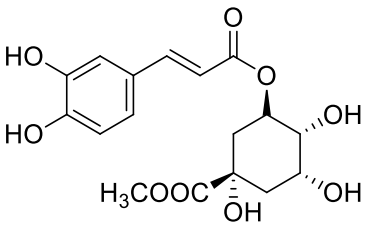
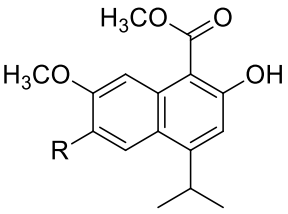
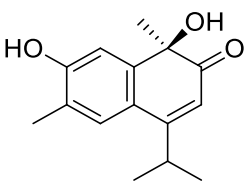
Table 1. Isolated compounds from *N. tabacum* and their biological activities

No	Extracts	Chemical constituents	Biological activities	References
1	The compounds were isolated from methanol extract of *N. tabacum* leaves	 **(1)**  **(2)**  **(3)**	Antiparasitic activity against *R. sanguineus* tick larvae. Compounds **(1)**, **(2),** and **(3)** exhibited good activity with value of 79.8, 88.3, and 81.1 % respectively at concentration of 300 µmol.	[25]
2	The compounds was isolated from dichloromethane extract of *N. tabacum* flowers	 **(4)**	Anticancer activity inhibit a HepG2 cell line. Compound **(4)** showed moderate activity with an IC_{50} of 14.38 µM.	[20]

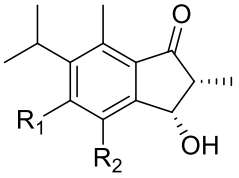
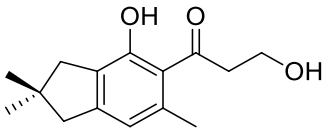
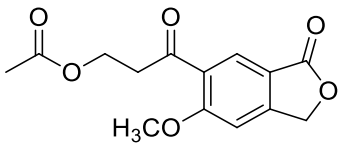
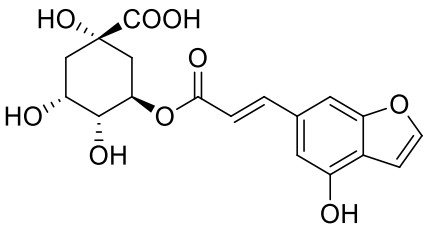
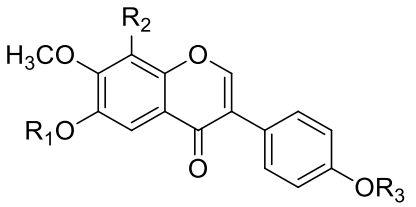
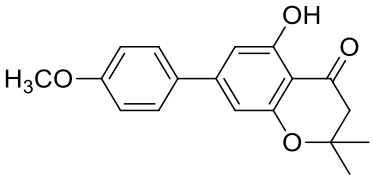
The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

3	The compounds were isolated from leaves methanol extract	 **(5)** R_1=H; R_2=H **(6)** R_1=CH_3; R_2=H **(7)** R_1=H; R_2=OCH_3	Cytotoxicity assay inhibit C8166 cells (CC_{50}) and anti-HIV-1 (EC_{50}). Compounds **(5)**, **(6)**, and **(7)** exhibited activity with CC_{50} values of 239.4; 142.6; and 118.8 µg/mL respectively as well as EC_{50} of 1.86; 2.78; and 2.64 µg/mL respectively. Furthermore, the three compounds also showed moderate cytotoxicities inhibit HL-60, HepG2, KB, and MDA-MB-231 cell lines.	[24]
4	The compounds were isolated from leaves of ethanol extract	 **(8)**	Antimigratory activities against prostate cancer cell line namely PC-3M-CT and PC-3. Compounds **(8)** and **(9)** showed anti-migratory	[21]

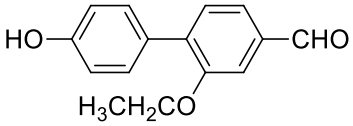
The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

		 **(9)**	activity with dose of 10 µM against PC-3M-CT and dose of 50 µM against PC-3.	
5	a. Compounds **(10)**, **(11)**, and **(27)** were isolated from roots and stems of acetone extract b. Compounds **(12)** and **(13)** were isolated from roots of acetone extract c. Compounds **(14)**, **(15)**, **(16)**, **(20)**, and **(21)** were isolated from stems of acetone extract d. Compounds **(17)**, **(18)**, **(19)**, **(22)**, **(23)**, **(24)**, **(28)**, and **(29)** were isolated from leaves of acetone extract e. Compounds **(25)** and **(26)** were isolated from acetone-water extract f. Compound **(30)** was isolated from	 **(10)** R=OH **(11)** R=OCH_3  **(12)**  **(13)**  **(14)** R=CH_3 **(15)** R=CH_3OH 	The anti-TMV activity of these compounds by using half-leaf method: -Compound **(10)**: 34.2% -Compound **(11)**: 33.4% -Compound **(12)**: 35.1% -Compound **(13)**: 33.2% -Compound **(14)**: 33.6% -Compound **(15)**: 35.8% -Compound **(16)**: 36.7% -Compound **(17)**: 36.7% -Compound **(18)**: 45.6% -Compound **(19)**: 35.2% -Compound **(20)**: 34.6% -Compound **(21)**: 35.4% -Compound **(22)**: 25.2% -Compound **(23)**: 22.6% -Compound **(24)**: 27.4%	[22, 23, 27, 28, 29, 30, 32, 33, 34, 35, 36, 44]

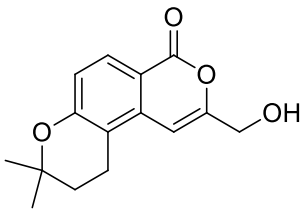
The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

stems and roots of methanol extract	**(16)**  **(17)** R_1=CH_3; R_2=H **(18)** R_1=H; R_2=CH_3  **(19)**  **(20)**  **(21)**  **(22)** R_1=CH_3; R_2=OH; R_3=H **(23)** R_1=H; R_2=$COOCH_3$; R_3=H **(24)** R_1=H; R_2=CH_2OH; R_3=CH_3  **(25)**	-Compound **(25)**: 48.4% -Compound **(26)**: 32.1% -Compound **(27)**: 33.8% -Compound **(28)**: 38.6% -Compound **(29)**: 46.7% -Compound **(30)**: 48.4%

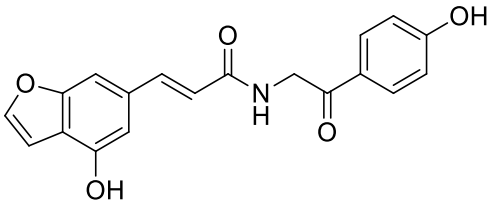
The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733



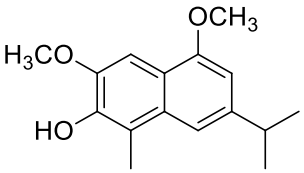
(26)



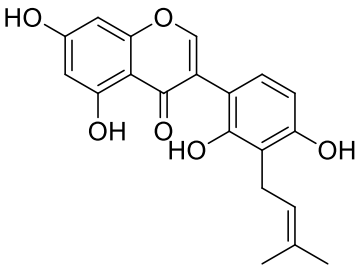
(27)



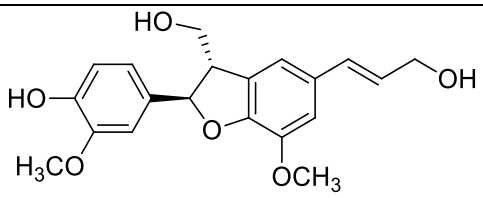
(28)



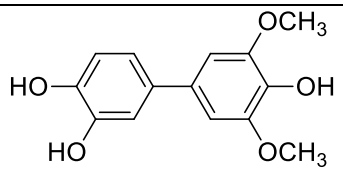
(29)



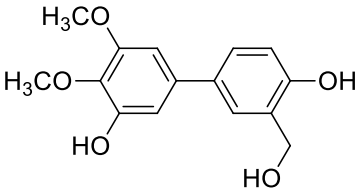
(30)

6	The compounds were isolated from roots, stems, and leaves of acetone extracts	 **(31)**	The cytotoxicities of these compounds against five human tumor cell lines: a. compounds	[27, 44, 45]

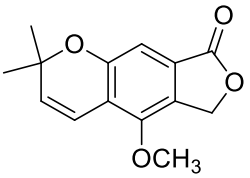
The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733



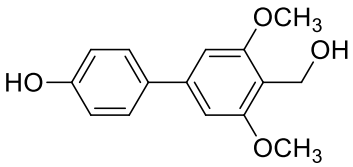
(32)



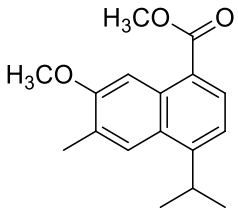
(33)



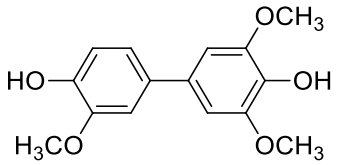
(34)



(35)



(36)



(37)

(15), **(31)**, and **(32)** exhibited inhibition against NB4 with IC_{50} of 5.7; 5.5; and 6.8 μM.

b. compounds **(13)**, **(15)**, and **(33)** exhibited inhibition against A549 with IC_{50} of 3.8; 8.2; and 2.8 μM.

c. compounds **(16)**, **(34)**, and **(35)** exhibited inhibitory activities against SHSY5Y with IC_{50} of 8.2; 6.8; and 3.8 μM.

d. compounds **(13)**, **(33)**, and **(36)** exhibited inhibitory activities against PC3 with IC_{50} of 5.2; 3.7; and 6.7 μM.

The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

		e. compounds **(13)**, **(14)**, **(15)**, and **(37)** exhibited inhibition against MCF7 with IC_{50} values of 4.8; 7.9; 7.8; and 6.2 μM.

BIOLOGICAL ACTIVITIES OF *N. tabacum*

Several plants, bacteria, and fungi has been reported their biological activities. Biological activity is an organism's ability to fight free radicals, viruses, and pathogenic bacteria that will damage the metabolic system of living things. Furthermore, this biological activity can be determined through bioassay technique based on both *in vitro* and *in vivo* results. According to several literatures, both extracts and isolated compounds of *N. tabacum* have been reported their bioactivities such as antiviral, antimicrobial, antioxidant, anti-HIV, anti-proliferative, anti-inflammatory, anti-parasitic, anti-termite, and cytotoxicity effects.

4.1 Anti-tobacco mosaic virus (Anti-TMV)

TMV is a positive-stranded RNA virus that enters the host cell. TMV is a species of the genus *Tobamovirus* and infects *N. tabacum* and also other species in the Solonaceae family including tomato, potato, pepper, and tobacco [54]. Therefore, virus infected edible natural products can affect abnormality human body that include stunting and other malnutrition diseases. Based on that, inhibitors for TMV are urgently needed to prevent the spreading virus. The virus inhibitors called as anti-TMV are compound-identified that was isolated from *N. tabacum* extract. There are about 20 compounds that have been reported as anti-TMV. Isolated compound from leaves acetone extract, the new 14-noreudesmane sesquiterpene **(29)** showed lower inhibitory concentration with IC_{50} value of 25.8 μM than a standard ningnanmycin (IC_{50} 38.2 μM) by using the half-leaf method [29]. Hence, the compound **(29)** has potential as an inhibitor TMV and also followed by other compounds including biphenyl **(25)** [35] and isoflavone **(30)** from methanol extract of *N. tabacum* stems and roots [23].

4.2 Antimicrobial

Antimicrobial activity evaluation of *N. tabacum* against bacteria, fungi, and oomycete has been reported. The antimicrobial activity was investigated by using *in silico* and *in vitro* technique. The result showed that the terpenoid extract of leaf tobacco is potent to inhibit several pathogens such as *X. campestris*, *E. carotovora*, *E. amylovora*, *Ps. tabaci*, *Ps. glycinea*, *Cladosporium* sp, *A. alternate*, *R. solani*, *F. graminearum*, *F. oxysportum*, *B. oryzae*, *C. gloesporioides*, *S. oryzae*, and *P. infestans* [38]. Furthermore, antibacterial activity of ethyl

The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

acetate extract from *N. tabacum* leaves had been reported by using agar well diffusion assay. The result reported that ethyl acetate extract can inhibit several pathogen bacteria such as *P. aeruginosa*, *K. pneumonia*, *S. aureus*, clinical *S. aureus*, *S. enterica*, *Micrococcus* sp, *P. mirabilis*, biofilm forming *Klebsiella* sp, biofilm forming *E. coli*. Among the pathogens, the ethyl acetate extract showed inhibitory activity against *S. aureus* significantly with area inhibition of 159.9±11.31 mm^2 [39]. In addition, antibacterial activity against six bacteria of *N. tabacum* leaves by using agar well diffusion method was also reported. The methanol extract of *N. tabacum* leaves exhibited antibacterial activity against *B. subtilis*, *C. pyogenes*, *P. aeruginosa*, *S. marcescens*, *S. dysenteriae*, and *S. aureus* with streptomycin as a standard. Moreover, the methanol extract presented strong antibacterial activity against *S. dysenteriae* with MIC value of 1.56 mg/mL [40].

4.3 Antioxidant

Antioxidant agent has an important role to attack free radicals in the human cell. In the long term, those free radicals inside will cause several chronic diseases such as cancer and diabetes [55], [56]. Based on literature study, *N. tabacum* is recommended as antioxidant agent. Antioxidant activity of aqueous and methanol extracts from stems was evaluated by enzymatic and non-enzymatic biochemical assay including superoxide dismutase, catalase, glutathione content, and glutathione *s*-transferase activities [41]. Antioxidant activity of several extracts from *N. tabacum* roots including aquades, methanol, acetone, and hexane extracts has been evaluated by DPPH method with trolox as a standard [47]. Furthermore, antioxidant activity of polyphenol extract from tobacco leaves also has been reported. The polyphenol extract was prepared by using 80% ethanol. The result showed that the polyphenol extract exhibited higher scavenging activities of DPPH, hydroxyl as well as superoxide radicals with IC_{50} values of 5.02, 49.6, and 44.0 μg/mL respectively compared with vitamin C as a standard [43]. Recently, there are several reports about isolation and expression of the key enzymes that catalyze polyphenol biosynthesis from *N. tabacum*. The result indicated that a new chalcone (NtCHS6) and flavonol (NtHDG2) synthase genes were abundantly expressed in *N. tabacum* leaves [57], [58]. Therefore, the polyphenol extract performed a good antioxidant activity.

4.4 Anti-HIV-1

An immunodeficiency disease is caused by human immunodeficiency virus type 1 (HIV-1) [59]. New nicotphenols **(5-7)** isolated from 70% aqueous ethanol of leaves has an anti-HIV-1 activity. The anti-HIV-1 was investigation by the cytopathic inhibitory activity of HIV-1 expressed with EC_{50} value. Among nicotphenols, nicotphenol **(5)** has stronger inhibition with EC_{50} of 1.86 μg/mL compared with azidothymidine as a standard [24].

4.5 Anti-poliferative

Anti-proliferative investigation of extracts derived (aqueous, methanol, acetone, and hexane) from *N. tabacum* roots has been reported. The anti-proliferative effect was evaluated on HeLa cervical adenocarcinoma with doxorubicin as a positive control. Among extracts derived, acetone extract showed the highest anti-proliferative with viability value of 14% at dose of 0.625 mg/mL [47].

4.6 Anti-inflammatory

The study of anti-inflammatory potential from *N. tabacum* had been reported. The phytosterols of *N. tabacum* was evaluated through selective inhibition of cyclooxygenase-1 and cyclooxygenase-2. Based on this research, phytosterols are potential inhibitors of

The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

cyclooxygenase-2 because they are non-toxic and hepatoprotective. In addition, they can regenerate parietal cells well [48].

4.7 Antiparasitic

Pesticides are commonly correlated as antiparasitic to improve the quality of agricultural food [21]. On the other hand, antiparasitic pesticides (APs), such as emamectin benzoate (EB), cypermethrin (CP), and deltamethrin (DE), have been used to treat parasitic diseases [61, 62]. Furthermore, the three APs were also used by the salmon industry [63], [64]. Recently, antiparasitic activity of methanol extract from *N. tabacum* leaves had been evaluated. The bioassay system was done against parasites, namely *Ctenocephalides felis*, *Lucilia cuprina*, *Caenorhabditis elegans*, *Rhipicephalus sanguineus*, and *Ixodes ricinus* [25].

4.8 Anti-termite

Anti-termite activity of ethanol extract from *N. tabacum* stems and roots had been reported. The anti-termite activity was evaluated against *Coptotermes curvignathus* by cellulose pads method. The result performed that the extract possessed a good activity at concentration of 5% [49].

4.9 Cytotoxicity

The cytotoxicity of isolated compounds from *N. tabacum* had been reported against several cancer and tumor cells by using MTT assay with taxol as a standard. The cytotoxicity assay was performed against acute promyelocytic leukemia cells (NB4), neuroblastoma cells (SHSY5Y), lung adenocarcinoma epithelial cells (A549), breast adenocarcinoma cell (MCF7), prostate cancer cells (PC3), liver cancer cells (HepG2), and colon cancer cells (HCT-116). The assay result showed that biphenyl **(33)** has highest cytotoxicity against A549 and PC3 with IC_{50} of 2.8 and 3.7 μM respectively. And also biphenyl **(35)** exhibited cytotoxicity against SHSY5Y with IC_{50} of 3.8 μM. Both of biphenyls **(33** and **35)** were isolated from 70% aqueous acetone of *N. tabacum* leaves [45]. Furthermore, cembranoid **(4)** isolated from dichloromethane extract of *N. tabacum* flowers had performed moderate cytotoxicity against HepG2 with IC_{50} of 14.38 μM [20].

CONCLUSION

N. tabacum (tobacco) has the main secondary metabolites such as cembranoids and flavonoids. They have selective inhibition as antiviral, antimicrobial, antioxidant, anti-HIV, anti-proliferative, anti-inflammatory, anti-parasitic, anti-termite, anticancer, and antitumor. Therefore, as the most commercially agricultural products in the world, chemical constituents and their pharmacological effects of *N. tabacum* are potential for further investigation.

ACKNOWLEDGMENT

We thank to Fatmawati, S., Ph.D for support and helpful discussions on this research. This research was funded by a grant in 2021 No 068/E4.1/AK.04.PT/2021 from Ministry of Research and Technology, National Research and Innovation Agency, Indonesia.

REFERENCES

[1] J. C. Tilburt and T. J. Kaptchuk, Herbal medicine research and global health: an ethical analysis, *Bull. World Health Organ. B World Health Organ*, **2008**, 86(8), 594-599.

The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

[2] Elfahmi, H. J. Woerdenbag, O. Kayser, Jamu: Indonesian traditional herbal medicine towards rational phytopharmacological use, *J. Herb. Med.*, **2014**, 4(2), 51-73.

[3] S. Fatmawati, Yuliana, A. S. Purnomo, M. F. A. Bakar, Chemical constituents, usage and pharmacological activity of *Cassia alata*, *Heliyon*, **2020**, 6(7), 04396.

[4] D. A. Putri and S. Fatmawati, Metabolit Sekunder dari *Muntingia calabura* dan Bioaktivitasnya, *ALCHEMY*, **2019**, 15(1), 57-78.

[5] Yuliana and S. Fatmawati, Senyawa Metabolit Sekunder dan Aspek Farmakologi *Alocasia macrorrhizos*, *Akta Kimia Indonesia*, **2018**, 3(1), 141-158.

[6] P. A. Maulida, D. A. Putri, S. Fatmawati, Free Radical Scavenging Activity of *Chromolaena odorata* L. Leaves, *IPTEK The Journal for Technology and Science*, **2019**, 30(3), 73-75.

[7] L. Khumaidah, A. S. Purnomo, S. Fatmawati, Antimicrobial Activity of *Sonneratia ovata* Backer, *HAYATI Journal of Biosciences*, **2019**, 26(4), 152.

[8] H. Kainama, S. Fatmawati, M. Santoso, P. Kakisina, T. Ersam, *In vitro* and *In vivo* Antiplasmodial of Stem Bark Extract of *Garcinia husor*, *HAYATI Journal of Biosciences*, **2019**, 26(2), 81.

[9] S. Fatmawati and K. Shimizu, Anti-oxidant and Aldose Reductase Inhibitory Activity of *Piper betle* Extracts, *Proc. Pak. Acad. Sci.: B*, **2019**, 56(3), 75-82.

[10] T. Y. K. Lulan, S. Fatmawati, M. Santoso and T. Ersam, α-VINIFERIN as a potential antidiabetic and antiplasmodial extracted from *Dipterocarpus littoralis*, *Heliyon*, **2020**, 6(5), 04102.

[11] E. R. Sukandar, S. Kaennakam, K. Rassamee, P. Siripong, S. Fatmawati, T. Ersam and S. Tip-pyang, Xanthones and biphenyls from the stems of *Garcinia cylindrocarpa* and their cytotoxicity, *Fitoterapia*, **2018**, 130, 112-117.

[12] T. Ersam, S. Fatmawati and D. N. Fauzia, New Prenylated Stilbenes and Antioxidant Activitites of *Cajanus cajan* (L.) Millsp. (Pigeon pea), *Indones. J. Chem.*, **2016**, 16(2), 151-155.

[13] S. Fatmawati, T. Ersam and K. Shimizu, The inhibitory activity of aldose reductase *in vitro* by constituents of *Garcinia mangostana* Linn, *Phytomedicine*, **2014**, 22, 49-51.

[14] S. Fatmawati, K. Shimizu and R. Kondo, Structure-activity relationships of ganoderma acids from *Ganoderma lucidum* as aldose reductase, *Bioorg. Med. Chem. Lett*, **2011**, 21, 7295-7297.

[15] S. Fatmawati, K. Shimizu and R. Kondo, Ganoderic acid Df, a new triterpenoid with aldose reductase inhibitory activity from the fruiting body of *Ganoderma lucidum*, *Fitoterapia*, **2010**, 81, 1033-1036.

[16] WHO, *Tobacco & Health in the Developing World*, The European Commission in collaboration with the World Health Organization and the World Bank, Brussels, **2003**

[17] M. Rachmat, Development of National Tobacco Economy: Developed Country Policy and Lesson Learned for Indonesia, *Analisis Kebijakan Pertanian*, **2010**, 8 (1), 67-83.

[18] M. Rachmat and S. Nuryanti, The Dynamic of World Tobacco Agribusiness and Its Implications for Indonesia, *Forum Penelitian Agro Ekonomi*, **2009**, 27(2), 73-91.

[19] A. Charlton, Medicinal uses of tobacco in history, *J. R. Soc. Med.*, **2004**, 97, 292-296.

The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

[20] X. F. He, X. D. Hou, X. Ren, K. Guo, X. Z. Li, Z. Q. Yan, Y. M. Du, Z. F. Zhang and B. Qin, Two new cembranic diterpenoids from the flowers of *Nicotiana tabacum* L., *Phytochem. Lett.*, **2016**, 15, 238-244.

[21] H. N. Baraka, M. A. Khanfar, J. C. Williams, E. M. El-Giar and K. A. El-Sayed, Bioactive Natural, Biocatalytic, and Semisynthetic Tobacco Cembranoids, *Planta Medica*, **2011**, 77, 467-476 https://doi.org/10.1055/s-0030-1250478

[22] L. Li, Q. P. Shen, C. B. Liu, Y. Wang, J. J. Yao, T. Zhang, F. M. Zhang, P. He, X. X. Shi, Z. H. Liu, M. M. Miao and G. Y. Yang, Isoflavones from the leaves of *Nicotiana tabacum* and their anti-tobacco mosaic virus activities, *Phytochem. Lett.*, **2015**, 13, 156-159.

[23] Z. Chen, J. Tan, G. Yang, M. Miao, Y. Chen and T. Li, Isoflavanones from the roots and stems of *Nicotiana tabacum* and their anti-tobaco mosaic virus activities, *Phytochem. Lett.*, **2012**, 5, 233-235.

[24] Y. K. Chen, X. S. Li, G. Y. Yang, Z. Y. Chen, Q. F. Hu and M. M. Miao, Phenolic compounds from *Nicotiana tabacum* and their biological activities, *J. Asian Nat, Prod. Res.*, **2012**, 14(5), 450-456.

[25] S. S. Weber, K. P. Kaminski, J. L. Perret, P. Leroy, A. Mazurov, M. C. Peitsch, N. V. Ivanov and J. Hoeng, Antiparasitic properties of leaf extracts derived from selected Nicotiana species and *Nicotiana tabacum* varieties, *FCT*, **2019**, 132, 110660.

[26] X. Sui, H. Zhang, Z. Song, Y. Gao, W. Li, M. Li, L. Zhao, Y. Li and B. Wang, Ethylene response factor NtERF91 positively regulates alkaloid accumulations in tobacco (*Nicotiana tabacum* L.), *BBRC*, **2019**, 517, 164-171.

[27] P. S. Yang, S. Y. Tang, C. B. Liu, L. Ye, F. M. Zhang, P. He, Z. H. Liu, Y. K. Chen, M. M. Miao, Q. P. Shen and J. Q. Wang, Three new sesquiterpenes from the stems of *Nicotiana tabacum* and their bioactivities, *J. Asian Nat. Prod. Res.*, **2017**, 21(2), 109-116.

[28] Q. P. Shen, X. M. Xu, L. Li, W. Zhao, N. J. Xiang, G. Y. Yang, Y. K. Chen, M. M. Miao, C. B. Liu and Z. H. Liu, Sesquiterpenes from the leaves of *Nicotiana tabacum* and their anti-tobacco mosaic virus activity, *CCL*, **2016**. 30(22). 2545-2550.

[29] S. Z. Shang, W. Zhao, J. G. Tang, J. X. Pu, D. L. Zhu, L. Yang, H. D. Sun, G. Y. Yang and Y. K. Chen, 14-Noreudesmane sesquiterpenes from leaves of *Nicotiana tabacum* and their antiviral activity, *Phytochem. Lett.*, **2016**, 17, 173-176.

[30] S. Z. Shang, W. Zhao, J. G. Tang, X. M. Xu, H. D. Sun, J. X. Pu, Z. H. Liu, M. M. Miao, Y. K. Chen and G. Y. Yang, Antiviral sesquiterpenes from leaves of *Nicotiana tabacum*, *Fitoterapia*, **2015**. 108. 1-4.

[31] L. Bortesi, M. Rossato, F. Schuster, N. Reven, J. Stadlmann, L. Avesani, A. Falorni, F. Bazzoni, R. Bock, S. Schillberg and M. Pezzotti, Viral and murine interleukin-10 are correctly processed and retain their biological activity when produced in tobacco, *BMC Biotechnol.*, **2009**, 9(22), 1-13.

[32] S. Shang, J. Shi, J. Tang, J. Jiang, W. Zhao, X. Zheng, P. Lei, J. Han, C. Wang, D. Yuan, G. Yang, Y. Chen and M. Miao, New isolates from leaves of *Nicotiana tabacum* and their biological activities, *Nat. Prod. Res.*, **2018**, 1-8.

The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

[33] S. Z. Shang, W. X. Xu, L. Li, J. G. Tang, W. Zhao, P. Lei, M. M. Miao, H. D. Sun, J. X. Pu, Y. K. Chen and G. Y. Yang, Antiviral isocoumarins from the roots and stems of *Nicotiana tabacum*, *Phytochem. Lett.*, **2015**, 11, 53-56.

[34] S. Z. Shang, Y. X. Duan, X. Zhang, J. X. Pu, H. D. Sun, Z. Y. Chen, M. M. Miao, G. Y. Yang and Y. K. Chen, Phenolic amides from the leaves of *Nicotiana tabacum* and their anti-tobacco mosaic virus activities, *Phytochem. Lett.*, **2014**, 9, 184-187.

[35] S. Z. Shang, W. X. Xu, P. Lei, W. Zhao, J. G. Tang, M. M. Miao, H. D. Sun, J. X. Pu, Y. K. Chen and G. Y. Yang, Biphenyls from *Nicotiana tabacum* and their anti-tobacco mosaic virus, *Fitoterapia*, **2014**, 99, 35-39.

[36] G. H. Kong, Y. P. Wu, J. L. Shi, N. J. Xiang, L. X. Liu, G. R. Yang, Y. K. Li, X. P. Lu, Q. Liu and Q. F. Hu, Anti-tobacco mosaic virus phenylpropanoids from the stems of *Nicotiana tabacum*, *Phytochem. Lett.*, **2015**, 14, 230-233.

[37] L. Yuan, W. Huang, C. Zhang, N. Xiang, C. Liu, G. Yang, Y. Chen, M. Miao and Y. Ma, Antiviral flavones from the leaves of *Nicotiana tabacum*, *Phytochem. Lett.*, **2015**. 12. 75-78.

[38] Y. Capdesuner, J. G. Brizuela, H. P. Mock, K. V. Hernandez, M. H. Torre and C. E. S. Toca, Accessing to the *Nicotiana tabacum* leaf antimicrobial activity: *In-silico* and *in-vitro* investigations, *Plant Physiol. Biochem.*, **2019**, 139, 591-599.

[39] G. Ameya, A. Manilal and B. Merdekios, *In vitro* antibacterial activity and phytochemical analysis of *Nicotiana tabacum* L. extracted in different organic solvents, *The Open Microbiology Journal*, **2017**, 11, 352-359.

[40] D. A. Akinpelu and E. M. Obuotor, Antibacterial activity of *Nicotiana tabacum* leaves, *Fitotertapia*, **2000**, 71, 199-200.

[41] Y. Sharma, D. Dua, A. Nagar and N. S. Srivastava, Antibacterial activity, phytochemical screening and antioxiadant activity of stem of *Nicotiana tabacum*, *Int. J. Pharm. Sci.*, **2016**, 7(3), 1156-1167 https://doi.org/10.13040/IJPSR.0975-8232.7(3).1156-67

[42] L. G. S. Alderete, E. Agostini and M. I. Medina, Antioxidant response of tobacco (*Nicotiana tabacum*) hairy roots after phenol treatment, *Plant Physiol. Biochem.*, **2011**, 49, 1020-1028.

[43] H. Wang, M. Zhao, B. Yang, Y. Jiang and G. Rao, Identification of polyphenols in tobacco leaf and their antioxidant and antimicrobial activities, *Food Chem.*, **2008**, 107, 1399-1406.

[44] P. He, P. Yang, S. Tang, L. Ye, C. Liu, Q. Shen, D. Tang, F. Zhang, Z. Liu, Y. Chen, S. Yao, N. Xiang and Z. Huang, Three new isobenzofurans from the roots of *Nicotiana tabacum* and their bioactivities, *Nat. Prod. Res.*, **2017**, 31(23), 2730-2736.

[45] M. Zhou, K. Zhou, J. Lou, Y. D. Wang, W. Dong, G. P. Li, Z. Y. Jian, D. Gang, H. Y. Yang, X. M. Li and Q. F. Hu, New biphenyl derivatives from the leaves of *Nicotiana tabacum* and their cytotoxic activity, *Phytochem. Lett.*, **2015**, 14, 226-229.

[46] M. C. Pathak, Study on secondary metabolites produced from callus cultures of *Nicotiana tabacum* by plant tissue culture techniques, *J. Biotechnol.*, **2019**, 305, 12-32.

[47] S. A. Lahham, R. Sbieh, N. Jaradat, M. Almasri, A. Mosa, A. Hamayel and F. Hammad, Antioxidant, antimicrobial and cytotoxic properties of four different extracts derived from the roots of *Nicotiana tabacum* L., *Eur. J. Integr. Med.*, **2020**, 33, 101039.

The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

[48] O. A. Akinloye, D. I. Akinloye, S. B. Onigbinde and D. S. Metibemu, Phytosterols demonstrate selective inhibition of COX-2: *In-vivo* and *in-silico* studies of *Nicotiana tabacum*, *Bioorg. Chem.*, **2020**, 102, 104037, 1-13.

[49] D. N. Sholehah, Uji Aktifitas Anti Rayap Tembakau dan Salak Madura, *Agrovigor*, **2011**, 4(1), 38-41.

[50] CABI, *CABI Invasive Spesies Compendium*, https://www.cabi.org/isc/datasheet/36326#toreferences. 1 October 2020.

[51] M. Chomel, M. G. Larcheveque, C. Fernandez, C. Gallet, A. DesRochers, D. Pare, B. G. Jackson and V. Baldy, Plant secondary metabolites: a key driver of litter decomposition and soil nutrient cycling, *J. Ecol.*, **2016**, 104, 1527-1541.

[52] S. Pagare, M. Bhatia, N. Tripathi, S. Pagare and Y. K. Bansal, Secondary Metabolites of Plants and their Role: Overview, *Current Trends in Biotechnology and Pharmacy*, **2015**, 9(3), 293-304.

[53] D. S. Seigler, *Plant Secondary Metabolism*, **1998**, Springer, Boston.

[54] A. Prabahar, S. Swaminathan, A. Loganathan and R. Jegadeesan, Identification of Novel Inhibitors for Tobacco Mosaic Virus Infection in Solonaceae Plants, *Hindawi*, **2015**, 198214, 1-9.

[55] D. A. Putri and S. Fatmawati, A New Flavanone as a Potent Antioxidant Isolated from *Chromolaena odorata* L. Leaves, *Evidence-Based Complementary and Alternative Medicine*, **2019**, 1453612, 1-12.

[56] D. A. Putri, A. Ulfi, A. S. Purnomo and S. Fatmawati, Antioxidant and antimicrobial activities of *Ananas comosus* peel extracts, *Mal. J. Fund. Appl. Sci.*, **2018**, 14(2), 307-311.

[57] C. Shuai, Z. Yin-chao, P. Xu-hao, L. Yi-ting, W. Feng-yan, C. Mo-ju, Y. Ai-guo and P. Guang-tang, Isolation and expression analysis of NtCHS6, a new chalcone synthase gene from Nicotiana tabacum, *J. Integr. Agric.*, **2017**, 16(7), 1443-1450.

[58] Z. Wang, S. Wang, Y. Xiao, Z. Li, M. Wu, X. Xie, H. Li, W. Mu, F. Li, P. Liu, R. Wang and J. Yang, Functional characterization of a HD-ZIP IV transcription factor NtHDG2 in regulating flavonols biosynthesis in *Nicotiana tabacum*, *Plant Physiol. Biochem.*, **2020**, 146, 259-268.

[59] X. Lu and Z. Chen, The development of anti-HIV-1 drugs, *Yao Xue Xue Bao*, **2010**, 45(2), 165-176.

[60] V. Freed, Environmental Dynamics of Pesticides 1 ed., **1975**, Springer US, New York.

[61] L. Burridge, J. S. Weis, F. Cabello, J. Pizarro and K. Bostick, Chemical use in salmon aquaculture: a review of current practices and possible environmental effects, *Aquaculture*, **2010**, 306, 7-23.

[62] F. Turkan, M. C. Harbi, A. Akgun, F. Gulbagca and F. Sen, Toxicological Effects of Some Antiparasitic Drugs on From Equine Liver Glutathione S-Transferase Enzyme Activity, *J. Pharm. Biomed. Anal.*, **2020**, 180, 113048.

[63] F. Tucca, M. Diaz-Jaramillo, G. Cruz, J. Silva, E. Bay-Schmith, G. Chiang and R. Barra, Toxic Effects of Antiparasitic Pesticides Used by the Salmon Industry in the Marine Amphipod Monocorophium insidiosum, *Arch Environ Contam Toxicol*, **2014**, 67, 139-148.

The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733

[64] F. Tucca, H. Moya, K. Pozo, F. Borghini, S. Focardi and R. Barra, Occurrence of antiparasitic pesticides in sediments near salmon farms in the northern Chilean Patagonia, *Mar. Pollut. Bull.*, **2017**, 115(1-2), 465-468.

The journal homepage www.jpacr.ub.ac.id
p-ISSN : 2302 – 4690 | e-ISSN : 2541 – 0733