Journal of Entomology

ISSN 1812-5670

Academic Journals Inc.

www.academicjournals.com

Journal of Entomology 9 (6): 361-367, 2012
ISSN 1812-5670 / DOI: 10.3923/je.2012.361.367

Present Status of Carbamate, Pyrethroid and Phenylpyrazole Insecticide Resistance to German Cockroach, *Blattella germanica* (Dictyoptera: Blattellidae) in Indonesia

[1]Resti Rahayu, [1]Intan Ahmad, [2]Endang Sri Ratna, [1]Marselina I. Tan and [1]Nova Hariani
[1]Bandung Institute of Technology, School of Life Sciences and Technology, Jalan Ganesha No. 10, Bandung, 40132, Indonesia
[2]Department of Plant Protection, Bogor Agricultural University, Darmaga Campus, Bogor 16680, Indonesia

Corresponding Author: Intan Ahmad, Bandung Institute of Technology, School of Life Sciences and Technology, Jalan Ganesha No. 10, Bandung, 40132, Indonesia Fax: (+62) 22 251 5033

ABSTRACT

Six strains of German cockroach, *Blattella germanica*, collected from three major cities in Indonesia which are Jakarta, Bandung and Surabaya were bio-assayed with topical application to monitor insecticide resistance to insecticides propoxur, permethrin and fipronil. In general, compared with VCRU susceptible strain as the standard, all cockroach strains were resistant to all insecticides tested, with Resistance Ratios ranging from 2.11-1013.17 fold. Majority of strains exhibited very high resistance (RR_{50}>50) to permethrin; this study also revealed the existence of extremely high resistance to permethrin (RR_{50} of 1013.17 fold) in HHB-JKT strain. Resistance levels to both propoxur and fipronil is vary (RR_{50} ranging from 2.11-44.72 fold). But, in general, the field strains are still susceptible to fipronil. This study is the second report of German cockroach resistance to insecticides in Indonesia which suggest the importance of continued monitoring of insecticides susceptibility for controlling the German cockroach, *B. germanica*. In addition, the findings can be used in formulating potential strategies for cockroach resistance management.

Key words: *Blattella germanica*, insecticide resistance, permethrin, propoxur, fipronil, Indonesia

INTRODUCTION

The German cockroach, *Blattella germanica* (Dictyoptera: Blattellidae) is unquestionably the most significant urban insect pest. It is known to be the vector of many pathogens that may cause a threat to human health, trigger asthma, vertigo and nausea to humans. Many professional Pest Control Operators (PCOs) as well as home owners have made efforts to control this pest mostly by insecticides. Unfortunately, the intensive and continuous use of insecticides to control this pest has caused the development of resistance of *B. germanica* to many insecticides (Ahmad *et al*., 2009; Nasirian *et al*., 2011). Furthermore, it is believed the German cockroach is able to develop resistance to many types of insecticides, owing to its survival ability, shorter life cycle and ability to digest chemical substances (Buczkowski and Schal, 2001). Resistance in the German cockroach to all the major groups of insecticides viz., organochlorines, organophosphates, carbamates and synthetics pyrethroids is quite common and has been reported in many countries (Hemingway *et al*., 1993; Jensen, 1993; Lee *et al*., 1996; Valles and Yu, 1996; Kristensen *et al*., 2005; Nasirian, 2010). As for Indonesia, the resistance of *B. germanica* to

insecticide is poorly documented and the only available information was published by Ahmad *et al.* (2009), they reported 95 fold resistance of German cockroach collected from Bandung strain to permethrin and suggested that the detoxifying enzyme Mixed Function Oxidases (MFO) played an important role in the development of resistance.

In Indonesia, synthetic insecticide, particularly pyrethroids and to the some extent of organophosphates and carbamates is commonly used by PCOs to control German cockroach. However, according to recent reports from some PCOs, notably from Jakarta, Bandung and Surabaya, as well as complaints from customers, intensive insecticide treatments did not reduce the population of cockroach into the expected level. This information leads to the possibility that cockroaches in some parts of Indonesia may have developed resistance to insecticides.

Therefore, knowing that the information on the resistance of *B. germanica* to insecticides from Indonesia is still limited; this report describes the present resistance status of *B. germanica* collected from Jakarta, Bandung and Surabaya to carbamate, pyrethroids and phenylpyrazole.

MATERIAL AND METHODS

Laboratory experiment

Cockroaches: There were seven strains of *B. germanica* used in this experiment (Table 1). The VCRU (Vector Control Research Unit) strain is a standard susceptible strain, originally obtained from the Vector Control Research Unit School of Biological Sciences Universiti Sains Malaysia. The field strains were collected from Jakarta, Bandung and Surabaya.

All cockroaches were reared without selection pressure in the Laboratory of Entomology, School of Life Sciences and Technology, Bandung Institute of Technology, Indonesia. Rearing and experimental conditions were; temperature of 22-28°C, relative humidity of 52-95% and photoperiod of 12:12 and was provided with water and mixture of fresh minced potatoes, oat meal and bread *ad libitum*. Rearing methods by Noland *et al.* (1949) and modification by Ahmad and Suliyat (2011) were applied. For experimental purposes, only male cockroaches were used.

Insecticides: The insecticides used in this study were technical grade i.e., propoxur 92.25%, permethrin 92.00% and fipronil 86.9% [Active Ingredients (AI)]. Both propoxur and permethrin were provided by PT. Inti Everspring, Indonesia, whilst fipronil was provided by PT. Bayer Indonesia. Prior to the tests, these insecticides were diluted with analytical grade acetone.

Insecticide bioassays: Topical tests to detect and quantify the resistance were carried out. The topical tests in which male adults *B. germanica* were treated with insecticide concentrations followed the methods as described by Lee *et al.* (1996). One microliter insecticide solution was applied topically on the first abdominal sternite of each carbon dioxide-anesthetized *B. germanica*

Table 1: Source of cockroaches, *B. germanica*

Strain	Collection site	Collection year	Origin
VCRU (Susceptible)	VCRU laboratory	2007	Penang
GFA-JKT	Food court	2007	Jakarta
GFB-JKT	Food court	2008	Jakarta
HHB-JKT	Restaurant	2007	Jakarta
KRSA-BDG	Restaurant	2007	Bandung
KRSB-BDG	Restaurant	2008	Bandung
SBY-SBY	Kitchen	2007	Surabaya

using a microsyringe (Valles and Yu, 1996). After treatment, cockroaches were kept in a 1 L volume of plastic jar, fed on slices of fresh peeled potatoes and provided with water. Mortality was observed at 72 h after treatment.

Resistance ratio: The Resistance Ratio (RR) was obtained by comparing the Lethal Dose $(LD)_{50}$ and of each field strain to the LD_{50} of susceptible strain (Scott *et al*., 1990; Lee *et al*., 1996).

Statistical analysis: The data were analyzed by probit analysis using POLO-PC software program (Leora Software, 2004) to determine the LD_{50} and LD_{90}.

RESULT AND DISCUSSION

Our findings provide evidence that in general, *B. germanica* strains collected from Jakarta, Bandung and Surabaya have developed resistance to the three major groups of insecticides as compared to a susceptible laboratory strain (VCRU) (Table 2-4). Using the classification resistance

Table 2: Toxicity of propoxur applied topically to adult cockroaches

Strain[a]	LD_{50} (µg insect^{-1})	RR at LD_{50}	LD_{90} (µg insect^{-1})	Slope±SE
VCRU	0.08	-	0.29	2.33±0.65
GFA-JKT	0.32	4.00	9.07	0.09±0.18
GFB-JKT	0.32	4.00	0.94	2.70±0.56
SBY-SBY	0.98	12.25	31.52	0.85±0,36
KRSA-BDG	0.96	12.00	7.22	1.47±0.34
KRSB-BDG	0.17	2.13	0.87	1.80±0.42
HHB-JKT	1.35	16.88	7.03	1.79±0.35

[a]Strain name/each collection, RR: Resistance ratio = LD_{50} resistant strain/LD_{50} susceptible strain

Table 3: Toxicity of permethrin applied topically to adult cockroaches

Strain[a]	LD_{50} (µg insect^{-1})	RR at LD_{50}	LD_{90} (µg insect^{-1})	Slope±SE
VCRU	0.06	-	0.11	4.92±0.81
GFA-JKT	18.94	315.67	134.46	1.51±0.22
GFB-JKT	0.17	2.83	1.76	1.27±0.37
SBY-SBY	24.33	405.50	73.63	2.67±0.25
KRSA-BDG	4.27	71.17	49.42	1.21±0.29
KRSB-BDG	0.41	6.83	2.39	1.69±0.37
HHB-JKT	60.79	1013.17	241.69	2.14±0.43

[a]Strain name/each collection, RR: Resistance ratio = LD_{50} resistant strain/LD_{50} susceptible strain

Table 4: Toxicity of fipronil applied topically to adult cockroaches

Strain[a]	LD_{50} (ng insect^{-1})	RR at LD_{50}	LD_{90} (ng insect^{-1})	Slope±SE
VCRU	0.76	-	2.65	2.36±0.62
GFA-JKT	33.91	44.72	85.06	3.21±0.90
GFB-JKT	2.50	3.30	4.00	4.42±1.03
SBY-SBY	2.97	3.91	7.98	2.98±0.61
KRSA-BDG	1.70	2.24	3.10	4.93±0.89
KRSB-BDG	1.60	2.11	3.20	4.15±0.88
HHB-JKT	24.10	31.78	349.50	1.10±0.40

[a]Strain name/each collection, RR: Resistance ratio = LD_{50} resistant strain/LD_{50} susceptible strain

ratio developed by Lee and Lee (2004) we found that the resistance levels of the field collected strains were low to high (2.13-16.88 fold) for the propoxur, low to very high (2.83-1013.17 fold) for the permethrin and low to high for the fipronil (2.11-44.872 fold). Knowing the practice of regular spraying with permethrin and propoxur to control *B. germanica* were commonly carried out by the PCOs in Indonesia, this finding is not really surprising, except for the extremely high resistance to the permethrin, i.e., 1013.17 fold. However, the occurrence of fipronil resistance in two of the six strains was really surprising, because the use of fipronil to control cockroach had not been widely used in Indonesia and only available in bait formulations (Ahmad and Suliyat, 2011).

Resistance levels to propoxur: Three out of six strains had resistance ratios >10 (high resistance) to propoxur, i.e., HHB-JKT (16.88 fold) followed by SBY-SBY (12.25 fold) and KRSA-BDG (12.00 fold). The remaining strains (KRSB-BDG, GFB-JKT and GFA-JKT), had low resistance levels, ranging from 2.13-4.00 fold (Table 2). The slope in the field strain varied from 0.09±0.18-2.70±0.56, only the GFB-JKT strain had similar slope with the VCRU, the laboratory susceptible strain. This finding suggests that the majority of the field strains had more heterozygous individuals for resistance. Interestingly, the levels of cockroach resistance to propoxur reported in these experiments were generally lower than those reported from other countries. The highest resistance of *B. germanica* to propoxur found in this experiment is lower compared to the ones reported in Malaysia, England and Taiwan; 91.6 fold (Lee *et al*., 1996), 46 fold (Hemingway *et al*., 1993) and 62.50 fold (Pai *et al*., 2005), respectively. However, it is still higher compared to the ones reported in United States and Singapore; 16.6 fold (Scharf *et al*., 1996; Valles and Yu, 1996) and 21.5 fold (Chai and Lee, 2010), respectively. This finding explains the situation in Indonesia; due to concern of propoxur toxicity to humans, propoxur has not been commonly used by the PCOs in Indonesia to control the cockroach. Although interestingly, a few household aerosols, registered to control mosquito, cockroach and house fly contain a combination of propoxur (carbamates) and transfluthrin (pyrethroids) as the active ingredients. In contrast, in peninsular Malaysia resistance to propoxur was prevalent due to the use of propoxur as the primary insecticide to control German cockroach (Chai and Lee, 2010).

Resistance levels to permethrin: The LD_{50} and RR_{50} values for permethrin tested against all strains are shown in Table 3. All *B. germanica* field strain were resistant to permethrin and most of them were considered highly resistance with RR_{50}>50. The HHB-JKT strain exhibited the highest resistance to permethrin (1013.17 fold) followed by SBY-SBY *strain* (405.50 fold) and GFA-JKT (315.67 fold). Based on the previous report by Ahmad *et al*. (2009), a cockroach strain from Bandung had a RR_{50} of 95 for permethrin. It was really surprising to have found such a high resistance to permethrin (1013.17 fold) from HHB-JKT strain. This value of resistance, to the best of our knowledge, is apparently the highest in the world. Report of cockroaches resistance with RR_{50}>50 fold to permethrin is very rare. Some cases of cockroaches resistances to permethrin with RR_{50}>50 fold are Malaysian strain (280 fold) (Lee and Lee, 2004) and Korean strain 109.8 fold (Chang *et al*., 2010). In Iran, for example, despite continued use of permethrin to control German cockroach for almost 30 years, the resistance ratios of seven field collected strains were relatively low, ranged from 5.26-23.7 fold (Limoee *et al*., 2006). The finding of RR_{50}>1000 fold in this study is exceptional; hence, it is necessary to make new criteria in classifying resistance level, i.e., extremely high resistance for RR>1000. However, interestingly, the exception was found on GFB-JKT and KRSB-BDG strains which had RR_{50}<10, i.e., 2.83 and 6.83 fold, respectively. The

slope in the field strain varied from 1.21±0.29 to 2.67±0.25 which is lower than the VCRU strain (4.92±0.81). This finding indicates that field strains had more heterozygous individuals for resistance, whilst the VCRU strain had more homozygous individuals for susceptible.

The fact that majority of Indonesian strains had very high resistance to permethrin could be explained by the following reasons; first, because permethrin has been used in Indonesia to control various public health pests, including cockroach for more than 25 years. Second, because most of the field technicians of the PCO's did not have good understanding about insect resistance phenomena to insecticide and how to manage the resistance insects. This condition is supported by the recent survey carried out by Ahmad and Suliyat (2011) in 18 kitchen restaurants around Bandung. They reported that despite every other week of cypermethrin spray by PCO over a period of two years, the survey found that all 18 kitchens had moderate to heavy infestations with the number of cockroaches ranging from 41-243. Even though control failures was rather obvious and resistance seemed to be the problem; for reason that pyrethroids have low toxicity to humans as compare with other insecticides, PCO kept doing the same thing except changing one brand of pyrethroids insecticides with another pyrethroids.

Resistance levels to fipronil: The LD_{50} and RR_{50} values for fipronil tested against all strains are shown in Table 4. Based on the LD_{50} values among fipronil, propoxur (Table 2) and permethrin (Table 3), it is shown that fipronil was the most toxic insecticide with an LD_{50} of 0.76 ng insect^{-1} on VCRU strain. This LD_{50} value shows that fipronil is 79.1 fold and 105.5 fold more toxic than propoxur and permethrin, respectively. The findings of the present study is in agreement with those reported by Scott and Wen (1997) who found that fipronil was 100 fold more toxic to the susceptible cockroach as compared to bendiocarb (carbamate) and chlorpyrifos (organophosphate). Furthermore Nasirian *et al.* (2006) reported that fipronil was 2233 fold more toxic than permethrin if used topically to the susceptible cockroach. Interestingly, pattern of the slope which measures the heterogeneity of the test population in fipronil is different than those in propoxur and permethrin. In this experiment we find that the slope in the field strain varied from 1.10±0.40 to 4.93±0.89, whereas, the slope for the VCRU was 2.36±0.62. Based on the slopes, the finding suggests that the majority of the field strains, i.e., GFB-JKT, KRSA-BDG and KRSB-BDG had more susceptible homozygous individuals for fipronil.

Even though the majority of field strains showed low resistance to fipronil (<5), two out of six strains, i.e., GFA-JKT and HHB-JKT exhibited high resistance to fipronil (>10 to = 50). There is a possibility that the high resistance levels found in GFA-JKT and HHB-JKT strains is due to the cross-resistance between similar modes of actions of the previously applied insecticides. This possibility is based on earlier reports which showed that cyclodienes resistant insects were more tolerant to fipronil (Scott and Wen, 1997; Kristensen *et al.*, 2005). For example, German cockroach strains Zo960302 and Ga021001 from Europe were 1270 and 2030 fold resistant to dieldrin (cyclodienes) and 15 and 14 fold resistant to fipronil. Even though when the experiments took place, fipronil had never been used in Europe to control cockroach and the use of dieldrin (cyclodienes) to control cockroach was stopped 20 years earlier (Kristensen *et al.*, 2005). Furthermore the cross resistance of German cockroach is not only between fipronil and cyclodienes but also with pyrethroids as reported by Wei *et al.* (2001) that the Apyr-R strain had 480 fold resistance to deltamethrin and two-fold resistance to fipronil.

This finding in the present study is rather surprising and alarming as well. First, it is surprising because fipronil usage as the active ingredient of gel bait being applied to control cockroach in

Indonesia is quite recently (about 5 years). In addition to that, due to their expensive price, the use of gel baits to control cockroaches has not been widely used. Second, it is alarming because fipronil is considered as the best insecticide (in bait formulations) available to control the insecticide resistant cockroach. Ahmad and Suliyat (2011), reported that laboratory-made bait containing 0.03% fipronil was very effective in controlling the population of pyrethroids resistant German cockroach found in 18 kitchens, >90% reduction was achieved after 2 weeks of treatment and 100% reduction was achieved after 5 weeks of treatment, with the initial number of cockroach per kitchen ranging from 41-243.

In conclusion, present findings show that all *B. germanica* strains collected from Jakarta, Bandung and Surabaya were resistant to the three groups of insecticides (multiple resistance to propoxur, permethrin and fipronil) with resistance ratios ranging from 2.11-1013.17 fold. Since majority of *B. germanica* had developed a very high resistance levels to permethrin and one strain had developed an extremely high resistance level to permethrin (RR>1000) it is imperative to take preventive steps. A permethrin halt for a certain of period should be carried out to avoid further control failure in the field. For the control of *B. germanica*, propoxur and especially fipronil are suitable replacement for permethrin, except for those strains which had developed very high resistance to the compounds. Our findings also suggest that for informed decisions of which insecticide(s) is to be used, monitoring of insecticide susceptibility needs to be continued with more samples from all over Indonesia. Nonetheless, unless alternative control strategies is available and PCOs have adequate knowledge on how to manage insect resistance, we have reasons to worry that it is only a matter of time before resistance to any new insecticides is developed. Although this is not surprising seen from the ecological and evolutionary perspectives, as insect will develop resistance to any insecticide applied as a means of survival.

ACKNOWLEDGMENTS

We thank PT. Inti Everspring Indonesia and PT. Bayer Indonesia for providing us with the insecticides used in this experiment, Prof. Chow Yang Lee for VCRU strain (susceptible strain) and to Hilda Lionata for English correction. This study was partially funded by DIPA ITB (2008-2009) to Intan Ahmad.

REFERENCES

Ahmad, I. and Suliyat, 2011. Development of fipronil gel bait against German cockroaches, *Blattella germanica* (Dictyoptera: Blattellidae): Laboratory and field performance in Bandung, Indonesia. J. Entomol., 8: 288-294.

Ahmad, I., Sriwahjuningsih, S. Astari, R.E. Putra and A.D. Permana, 2009. Monitoring pyrethroid resistance in field collected *Blattella germanica* Linn. (Dictyoptera: Blattellidae) in Indonesia. Entomol. Res., 39: 114-118.

Buczkowski, G. and C. Schal, 2001. Emetophagy: Fipronil-induced regurgitation of bait and its dissemination from German cockroach adult to nymphs. Pesticide Biochem. Physiol., 71: 147-155.

Chai, R.Y. and C.Y. Lee, 2010. Insecticide resistance profiles and synergism in field populations of the German cockroach (Dictyoptera: Blattellidae) from Singapore. J. Econ. Entomol., 103: 460-471.

Chang, K.S., E.H. Sin, J.S. Jung, C. Park and Y.J. Ahn, 2010. Monitoring for insecticide resistance in field-collected populations of *Blattella germanica* (Blattaria: Blattellidae). J. Asia-Pac. Entomol., 13: 309-312.

Hemingway, J., G.J. Small and A.G. Monro, 1993. Possible mechanisms of organophosphorus and carbamate insecticide Resistance in German cockroaches (Dictyoptera: Blattelidae). Novel point Mutations are associated with knockdown areas. Econ. Entomol., 86: 1623-1630.

Jensen, K.M.V., 1993. Insecticide resistance in *Blattella germanica* (L.) (Dictyoptera: Blattellidae) from food pro-ducing establishments in Denmark. Proceedings of the First International Conference on Insect Pests in the Urban Environment. 1993, BPCC Wheatons, Exeter, UK., pp: 135.

Kristensen, M, K.K. Hansen and K.M.V. Jensen, 2005. Cross-resistance between dieldrin and fipronil in German cockroach (Dictyoptera: Blatellidae). J. Econ. Entomol., 98: 1305-1310.

Lee, C.Y., H.H. Yap, N.L. Chong and R.S.T. Lee, 1996. Insecticide resistance and synergism in field collected German cockroaches (Dictyoptera: Blattellidae) in Peninsular Malaysia. Bull. Entomol. Res., 86: 675-682.

Lee, L.C. and C.Y. Lee, 2004. Insecticide resistance profiles and possible underlying mechanisms in German cockroaches, *Blattella germanica* L. (Dictyoptera: Blattellidae) from Peninsular Malaysia. Med. Entomol. Zool., 55: 77-93.

Leora Software, 2004. Polo-PC: Probit and logit analysis. LeOra Software, California.

Limoee, M., H. Ladonni, A.A. Enayati, H. Vatandoost and M. Aboulhasani, 2006. Detection of pyrethroid resistance and cross resistance to DDT in seven field-collected strains of the German cockroach, *Blattella germanica* (L.) (Dictyoptera: Blattellidae). J. Boil. Sci., 6: 382-387.

Nasirian, H., 2010. An overview of german cockroach, *Blattella germanica*, studies conducted in Iran. Pak. J. Biol. Sci., 13: 1077-1084.

Nasirian, H., H. Ladonni, M. Aboulhassani and M. Limoee, 2011. Susceptibility of field populations of *Blattella germanica* (Blattaria: Blattellidae) to spinosad. Pak. J. Biol. Sci., 14: 862-868.

Nasirian, H., H. Ladonni, M. Shayeghi, H. Vatandoost and M.R. Yaghoobi-Ershadi *et al.*, 2006. Comparison of permethrin and fipronil toxicity against German cockroach (Dictyoptera: Blattellidae) strains. Iran. J. Publ. Health, 35: 63-67.

Noland, J.L., J.H. Lilly and C.A. Bauman, 1949. A laboratory method for rearing cockroaches and its application to dietary studies on the German roach. Ann. Ent. Soc. Am., 42: 63-70.

Pai, H.H., S.C. Wu and E.L. Hsu, 2005. Insecticide resistance in German cockroach (*Blattella germanica*) from hospitals and households in Taiwan. Int. J. Environ. Health Res., 15: 33-40.

Scharf, M.E., J. Hemingway, B.L. Reid, G.J. Small and G.W. Bennett, 1996. Toxicological and biochemical characterization of insecticide resistance in a field-collected strain of *Blattella germanica* (Dictyoptera: Blattellidae). J. Econ. Entomol., 89: 322-331.

Scott, J.G. and Z. Wen, 1997. Toxicity of fipronil to susceptible and resistant strains of German cockroaches (Dictyoptera: Blattellidae) and house flies (Diptera: Muscidae). J. Econ. Entomol., 90: 1152-1156.

Scott, J.G., D.G. Cochran and B.D. Siegfried, 1990. Insecticide toxicity, synergism and resistance in the German cockroach (Dictyoptera: Blattellidae). J. Econ. Entomol., 83: 1698-1703.

Valles, S.M. and S.J. Yu, 1996. Detection and biochemical characterization of insecticide resistance in the German cockroach (Dictyoptera: Blattellidae). J. Econ. Entomol., 89: 21-26.

Wei, Y., A.G. Appel, W.J. Moar and N. Liu, 2001. Pyrethroid resistance and cross-resistance in the German cockroach, *Blattella germanica* (L.). Pest. Manage. Sci., 57: 1055-1059.