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LAPORAN

Call For Paper International Conference on Biology, Sciences and Education

> Padang, 17-18 Oktober 2019 Universitas Negeri Padang

Oleh Febriana Dwi Wahyuni, M.Si

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2019

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PENDAHULUAN

Setiap dosen berkewajiban untuk melaksanakan Tridarma Perguruan Tinggi, yaitu pengajaran, penelitian, dan melakukan pengabdian kepada masyarakat. Kinerja dosen masih banyak berfokus pada pengajaran sehingga mengesampingkan dua kewajiban lainnya yaitu penelitian dan pengabdian kepada masyarakat. Saat ini, dosen mulai dituntut untuk banyak melakukan penelitian dan mempublikasikan hasil penelitiannya. Salah satu media untuk mempublikasikan hasil penelitian adalah melalui kegiatan Call For Paper. Melalui kegiatan ini, para dosen maupun peneliti bisa menyebarluaskan hasil penelitiannya.

TUJUAN

Konferensi ini bertujuan untuk sharing knowledge dari hasil penelitian yang telah dilakukan.

PELAKSANAAN

Waktu dan tempat

Konferensi dilaksanakan pada:

- Hari dan tanggal : Kamis-Jumat, 17-18 Oktober 2019
- □ Waktu : 08.00 WIB selesai
- Lokasi : Universitas Negeri Padang

Peserta Workshop

Mahasiswa, dosen maupun peneliti dari berbagai Perguruan Tinggi di Indonesia dan Luar negeri

Pendaftaran

Peserta yang bisa mendaftar untuk kegiatan Call For Paper International Conference on Biology, Sciences and Education adalah mahasiswa, dosen dan peneliti dari semua Perguruan Tinggi maupun lembaga penelitian di Indonesia dan Luar Negeri. Panitia pelaksana adalah dari Departemen Biologi, Universitas Negeri Padang.

Output dan Outcome

Setelah mengikuti kegiatan ini, diharapkan peserta dapat mentransfer ilmu dari hasil penelitian yang telah dilakukan

Penutup

Demikian Term of Reference ini dibuat sebagai panduan kegiatan. Semoga dapat memberikan informasi bagi pihak-pihak yang terkait.

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CONFERENCE SCHEDULE

DAY 1	17 October 2019				
07.30 - 08.30	Registration				
08.31 - 09.30	Opening Ceremony (ICoBioSE 2019)				
09.30 - 10.00	Coffee Break				
10.01 – 12.00	Keynote Speaker I Prof. Datin Dr Mariani Binti Md Nor University of Malaya Keynote Speaker II Dr. Katsuyuki Eguchi Tokyo Metropolitan University, Tokyo, Japan				
12.00 - 13.15	Lunch				
13.16 - 14.00	Poster Session				
14.01 - 15.00	Parallel Session I				
15.01 - 15.15	Coffee Break				
15.16 - 17.00	Parallel Session II				
17.01 - 19.00	Break				
19.01 - 22.00	Sarasehan di Gubernuran Sumbar* (masih dikonfirmasi)				
DAY 2	18 October 2019				
08.00 - 10.00	Keynote Speaker III Prof. Madya Dr. Haniza Hanim Mohd Zain Universitas Pendidikan Sultan Idris, Tanjung Malim, Malaysia Keynote Speaker IV Prof. Si-Min Lin, Ph.D National Taiwan Normal University, Taipei, Taiwan Keynote Speaker V Dr. Wendy Wang Yanling Lee Kong Chian Natural History Museum, Singapore				
10.00 - 10.15	Parallel Session III				
10.15 - 10.30	Coffee Break				
10.30 - 12.15	Parallel Session IV				
12.15 - 13.30	Praying break + Lunch				
13.30 - 15.45	Parallel Session V				
15.45 – Selesai	Penutupan				



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In Silico Analysis for *Cry*I Gene Amplification from Local Isolate of *Bacillus thuringiensis*

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Abstract. Bacillus thuringiensis is one of bacterial type that was used as a microbiological control agent for pests and vectors of plant disease. It was because in *B. thuringiensis* there is Cry protein which is specific insect repellent. This Cry Protein is only toxic to certain insects. One of them is CryI protein which is toxic to Lepidoptera insects. Lepidoptera can attack various types of plants. CryI Proteins was encoded by *cryI* gene. The method can be used for *cryI* gene amplification is Polymerase Chain Reaction (PCR). Primer is one important component in PCR. The aim of this research was get a good primer candidate for *cryI* gene amplification from *B. thuringiensis*. In silico analysis for designing *cryI* primer was carried out using several software, such as BLAST for searching sequence *cryI* gene, Bioedit for alignment, and DINAmelt for analyze dimer structure of primer was selected that could represent *cryI* gene, with forward primer 5'-CGGTGAATGCCCTGTTTACT -3' and reverse primer 5'-CGGTCTGGTTGCCTATTGAT -3'. Amplification of the *cryI* gene by PCR *in silico* produces a product with a length about 200 bp.

Keywords: bioinformatics, cry gene, PCR in silico, primer

1. Introduction

One of barrier in the production of a commodity in tropical and humid countries is the attack of plantdisturbing organisms such as insect pests and plant pathogens [1]. Even in certain plants such as rice, pest insects are still a major obstacle and become a serious problem, for example brown plant hopper and stem borer [1]. The control effort that is usually done by farmers is spraying insecticides. The use of insecticides was allegedly caused a negative impact on the environment and caused resistance to target insects. Concerns about the negative impact of the use of insecticides have increased public attention to bioinsecticides as an alternative to reducing pest populations [2].

Bacillus thuringiensis (Bt) is a bacterium that produces crystalline proteins that are toxic to insects and nematodes during sporulation [3]. *B. thuringiensis* bioinsecticides constitute 90-95% of the bioinsecticides commercialized for use by farmers in various countries [2]. The advantages of using this bacterium as a biopesticide include a protein produced by *B. thuringiensis* which is a specific buffer and is called a Cry protein (from the word crystal) or also called as δ -endotoxin. Cry protein is only toxic to certain types of insects and is not toxic to useful insects or to other organisms [3]. One type of Cry protein, CryI, is known to be able kill insects from the Lepidoptera group [4]. Insects of this type Lepidoptera are pests that attack various plants. The development of recombinant DNA technology is now possible to carry out the transformation of cry genes into plant cells. The introduction of cry genes into plant genomes is expected to produce plants capable of expressing endotoxins that can cause pest

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insect death. In this way it is also expected to reduce the use of insecticides that can pollute the environment.

Advances in technology in the field of molecular biology have encouraged scientists to isolate chromosomes or plasmids that contain cry genes. The cry gene is a DNA sequence that codes for the formation of Cry proteins [3]. Several studies have been carried out for cry gene amplification [5,6]. One method that can be used for cry gene amplification is the Polymerase Chain Reaction (PCR) which will be carried out in this study by applying a discipline of bioinformatics.

Bioinformatics is an interdisciplinary field which is broadly defined as a combination of biology (molecular biology) and computing using computer and software assistance [7]. One of the most significant roles of bioinformatics is to design and produce primary sequences. Primary is one component that plays an important role in the PCR process [8]. A good primer is a specific primer. Unspecified primers can cause amplification of other regions in the genome that are not targeted or otherwise there are no amplified genome regions. To get a primer that meets the good primary criteria for amplification, in silico design is carried out [7]. The aim of this study was to analyze the combination of primary pairs used in the PCR process to amplify the *cryI* gene from *B. thuringiensis*. The *cryI* gene that will be amplified is a domain of the total *cryI* gene sequences that already exist in *genebank*.

Materials and Methods

B. thuringiensis isolate

Bacteria culture of *B. thuringiensis* was obtained from the InaCC culture collection with code number B432 and B327

Bacterial DNA extraction

B. thuringiensis DNA extraction method was used the gSYNC DNA Extraction kit from Geneaid.

Target Gene Sequences

Target gene used in this research is *cry*I gene from *Bacillus thuringiensis* (accession number X56144). These sequence were obtained from the NCBI website (National Center for Biotechnology Information, www.ncbi.nlm.nih.gov).

Blast

The gene then was analyzed with Basic Local Alignment Search Tools (BLAST) software with other *B*. *thuringiensis* strains so that identity values from the highest to the lowest were obtained. Gene sequences that have high identity values are used in the multiple alignment method to obtain conserved sequences.

Multiple Alignments

The determination of sustainable areas is determined using the Bioedit program. The sequence of bases obtained is aligned with the ClustalW Program and then the one which has the most similarity in its constituent bases is similar to 90-100%. As a basis for tracking primary attachments [9].

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Primary Design

Primary DNA determination analysis was performed with Primer3 software, and manually selected the most sustainable areas by considering the area is the cipher sequence, with general requirements, including the number of nucleotides, GC content and there is no possibility of complementarity between bases in one primary chain or between one primer with another.

Analysis of Primary Dimer

Sometimes the primers that are designed can recognize the sequence of themselves, bind to one another to form a structure called a dimer. This can be a problem because primers will tend to stick together, not with the target gene and this can reduce DNA concentration. For this reason, it is necessary to do a dimer prediction analysis using available software to predict the presence of dimers in primary candidates, namely DINAmelt (http://unafold.rna.albany.edu/?q=DINAMelt) made by NM Markham and Michael Zucker of Rensselaer Polytechnic Institute [10,11]. From the results of this analysis using DINAmelt, it can be seen whether the dimers are formed in the primer, how much the formation of the G-C bonds, and the presence of the 3 'end of the complement.

Analysis of Restriction Sites in the cryI Gene

To find out the restriction sites contained in the *cry*I gene, an analysis was performed using *Snapgene* software. The purpose of knowing the restriction site is so that genes can be cut with one of the desired endonuclease restriction enzymes.

Amplification of the *cry*I gene by PCR

Amplification of the *cry*I gene using specific primer cry1 forward 5'-CGGTGAATGCCCTGTTTACT-3' and reverse 5'-CGGTCTGGTTGCCTATTGAT-3' with a total volume of 25 μ l containing 1 μ l DNA genome, 12.5 μ l PCR HS redmix master mix (Bioline), 1 μ l for each primer and 9.5 ddH₂O. PCR amplification was carried out as many as 35 cycles using Sensoquest. For one cycle consists of 3 stages, namely denaturation, attachment, and elongation. The predenaturation stage is carried out for 3 minutes at a temperature of 95°C once, the denaturation stage is 1 minute, the primary attachment stage is at 50-60°C for 1 minute, the extension of the DNA chain at a temperature of 72°C for 1 minute. In the last cycle, the chain was extended longer at 72°C for 5 minutes.

PCR Electrophoresis

PCR results were migrated into 1% agarose gel under 100 volt 30 minute conditions. 1 kb DNA marker is used as a marker. Gel staining used fluorosafe DNA stain which is directly added to the agarose gel. Gels containing DNA fragments were visualized using UV Trans Illuminators and documented using the Digibox Camera Documentation System gel.

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Result and Discussion a. Result *cry*I gene Sequences

Based on the search results of the *cry*I gene sequences in genebank with accession number X56144, *cry*I gene sequences were obtained from *Bacillus thuringiensis* consisting of 3516 bp (figure 1.)

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	cryI gene; crystal protein; insecticidal protein; protoxin. Bacillus thuringiensis		Find in this Sequence
ORGANISM	Bacillus thuringiensis		
	Bacteria; Firmicutes; Bacilli; Bacillales; Bacillaceae; Bacillus; Bacillus cereus group.		Related information
AUTHORS	1 Bosse,M., Masson,L. and Brousseau,R.		Protein
TITLE	Nucleotide sequence of a novel crystal protein gene isolated from Bacillus thuringiensis subspecies kenyae		PubMed
JOURNAL	Nucleic Acids Res. 18 (24), 7443 (1990)		Taxonomy
FERENCE	2 <u>2259636</u> 2		Full text in PMC
	Brousseau,R. Direct Submission		
	Submitted (17-OCT-1990) Brousseau R., Biotechnology Research Institute, 6100 Royalmount Avenue, Montreal, Quebec H4P 2R2, Canada		Recent activity
FERENCE	3 (bases 1 to 3516)		Turn Off
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JOURNAL	Submitted (24-MAR-1991) Bro∷sseau R., DNA and Gene Synthesis, Biothechnical Research Institute, 6100 Royalmount Ave., Montreal,		Bacillus thuringiensis strain BT HMM C
	Quebec H4P-2R2, Canada		crystal toxin protein (crylli) gene, No
DMMENT EATURES	See also X53985, which is 100% homologous to this updated sequence. Location/Qualifiers		Q bacillus thuringiensis crylli gene (35)
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	TGGLRINARFNQFRRELTISVLDIISFFRNYDSRLYPIPTSSQLTREVYTDPVINITD YRVGPSFENIENSAIRSPHLMDFLNNLTIDTDLIRGVHYNAGHRVTSHFTGSSQVITT		
	PQYGITANAEPRRTIAPSTFPGLNLFYRTLSNPFFRRSENITPTLGINVVQGVGFIQP		
	NNAEVLYRSRGTVDSLNELPIDGENSLVGYSHRLSHVTLTRSLYNTNITSLPTFVWTH HSATNTNTINPDIITQIPLVKGFRLGGGTSVIKGPGFTGGDILRRNTIGEFVSLQVNI		
	NSPITQRYRLRFRYASSRDARITVAIGGQIRVDMTLEKTMEIGESLTSRTFSYTMFSN PFSFRANPDIIRIAEELPIRGGELYIDKIELILADATFEEEYDLERAQKAVNALFTST		
	NQLGLKTDVTDYHIDQVSNLVECLSDEFCLDEKRELSEKVKHAKRLSDERNLLQDPNF		
	RGINRQPDRGWRGSTDITIQGGDDVFKENYVTLPGTFDECYPTYLYQKIDESKLKAYT RYELRGYIEDSQDLEIYLIRYNAKHETVNVPGTGSLWPLSAQSPIGKCGEPNRCAPHL		

Figure 1. The cryI gene Bacillus thuringiensis source from NCBI

Basic Local Alignment Search Tools (BLAST)

The BLAST process was carried out in *Bacillus thuringiensis cry*I gene as a determinant strain with other *B. thuringiensis* strains. *B. thuringiensis cry*I gene for insecticidal crystal protein as a determinant because it has a long DNA sequence. BLAST results obtained an identity value of 100%-96% cryI gene against several strains of *B. thuringiensis*.

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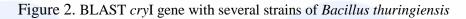
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Job Title	emb X56144.1	Filter Results
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B. thuringiens	is cryl gene for insecticidal cry	stal protein			6493	6493	100%	0.0	100.00%	<u>X56144.1</u>
Bacillus thurin	igiensis BtX I gene for crystal p	protein			6493	6493	100%	0.0	100.00%	<u>X53985.1</u>
Bacillus thurin	igiensis Cry032 (cry032) gene,	complete cds			6482	6482	100%	0.0	99.94%	AF20253
Bacillus thurin	igiensis (cryIE(a)) gene, compl	ete CDS			6482	6482	100%	0.0	99.94%	M73252.1
Bacillus thurin	Bacillus thuringiensis strain BRC-XQ12 insecticidal crystal protein Cry1Ea11 (cry1Ea11) gene. partial cds						99%	0.0	99.94%	JQ65245
Bacillus thurin	Bacillus thuringiensis strain BR64 Cry1Ea10 (cry1Ea10) gene, complete cds							0.0	99.89%	HQ43531
Bacillus thurin	igiensis strain S6 Cry1E-like pr	otein gene, partial cds			6469	6469	99%	0.0	99.91%	HQ43978
Bacillus thurin	igiensis strain HZM2 Cry1Ea g	ene, complete cds			6466	6466	100%	0.0	99.86%	EU24442
Bacillus thurin	igiensis isolate JC190 insectici	dal delta endotoxin Cry	IEa (cryIEa) gene, complete cds	2	6466	6466	100%	0.0	99.86%	AY89413
Bacillus thurin	igiensis protoxin Cry1Ea4 (cry1	1Ea4) gene, complete o	:ds		6460	6460	100%	0.0	99.83%	<u>U94323.1</u>
Bacillus thurin	igiensis serovar tolworthi plasn	nid pKK2 DNA, complete	<u>te genome, strain: Pasteur Instit</u>	ute Standard strain	6139	6139	99%	0.0	98.21%	AP01486
Bacillus thuringiensis strain FB Cry1Ea (cry1Ea) gene, partial cds						6130	99%	0.0	98.18%	JN22610
Bacillus thurin	igiensis strain V4 Cry1Ea gene	<u>partial cds</u>			6111	6111	99%	0.0	98.09%	KF60155
Bacillus thurin	igiensis delta-endotoxin Cry1-A	<u>A32 (cry1-A32) gene, p</u>	artial cds		3616	3616	62%	0.0	96.36%	AY99393
Bacillus thurin	igiensis serovar galleriae strain	HD-29 plasmid pBMB	426 complete sequence		2929	5427	61%	0.0	90.81%	CP01009

Figure 3. Analysis of the cryI gene identity with the cryI gene from another Bacillus thuringiensis strain

Multiple Alignment

From several strains of BLAST results that have 100-96% DNA sequence identity values, multiple alignment is performed, so that several species with DNA sequences of *cry*I genes that have parallel DNA sequences are obtained (Figure 4). Sequences selected parallel to the CDS region that corresponds to the primary DNA sequence that has been designed with Primer3 software.

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Q435318.1 EGAA	TAATCAGAATCAATGC TAATCAGAATCAATGC	STGCCTTATAAT STGCCTTATAAT	IGTTTAAATA. IGTTTAAATA	ATCCTGAAAA ATCCTGAAAA		PTAGATATTGAP PTAGATATTGAP							SCAACT SCAACT
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	TAATCAGAATCAATGC												

Figure 4. The multiple alignment process of *cry*I gene from *B. thuringiensis* with other strain that have 100-96% DNA sequence identity using Bioedit software

DNA primer *cry*I gene

The design of the primary DNA of the *cry*I gene from Bacillus thuringiensis uses primer3 software that has passed the BLAST process and multiple alignment (Table 1).

	Table 1. cry1 Pr	inter design from <i>B. inuringlensis</i>			
	Primer (<i>cry</i> I) Sequence (5'-3')		Tm (⁰ C)	GC%	Self 3'
		Oniversitas			complementarity
-	Forward	CGGTGAATGCCCTGTTTACT	60	50	1
	Reverse	CGGTCTGGTTGCCTATTGAT	60	50	2

Table 1. cry1 Primer design from B. thuringiensis

Dimer Structure in Primer Candidate

The results of the analysis with DINAmelt software, it can be seen that the primers that have been designed only have 2 G-C bond. This is advantageous because it can increase the concentration of PCR products and the possibility for the primer bind to each other is very small.

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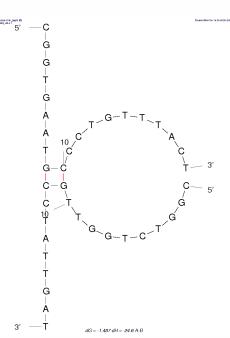




Figure 5. This figure shows DINAmelt analysis results to predict dimer structure on primer pair previously designed

Map of cryI gene

The results of the snapgene analysis showed that the amplified *cry*I gene had 8 restriction sites. This information can be used as a basis for cloning.

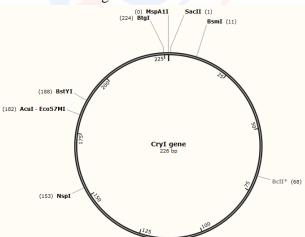


Figure 6. Mapping Restriction Enzymes in *cry*I Genes with Snapgene software

PCR Electrophoresis

DNA visualization results that have been amplified with cry1 forward and cry1 reverse primers show DNA bands with a size of about 200 bp.

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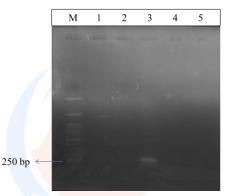


Figure 7. Amplification of *cry*I gene

b. Discussion

Primary design is the first step that determines the success of DNA amplification by PCR method [12]. Criteria that need to be considered in the selection of primers include the length of primer, temperature melting (Tm), GC content and bonds at the end of 3 '. Good primers range from 18-30 base pairs. Primers which have a length more than 30 base pairs will cause the primary attachment to be unspecified. The second characteristic to consider in primary selection is Tm. A good primer has a Tm difference of around 5^oC. This is intended to prevent a decrease in the amplification process. The percentage between bases G and C also needs to be considered because the content of the number of bases G and C can affect the Tm of a primary [13]. A good primary has a percentage of G and C around 40-60%. Another criteria for good primers is having a low self 3' complementarity so that there is no attachment between primer pairs and forms a structure called a hairpin [14].

Dimers are structures formed between primer pairs, where they are united because they have a complementary basis. This process occurs at the appropriate attachment temperature, usually at low temperatures. By looking at the stages of the PCR process, it can be seen that the primary attachment to the printed DNA occurs at optimal annealing temperatures. This process can occur simultaneously with the formation of dimers. The problem that may arise is that primers have a tendency to stick to one another, and not stick to printed DNA. If this dimer bond is too strong, it will interfere with the DNA extension process and will result in low DNA concentration. From the analysis using DINAmelt, it can be seen that the primer forms a dimer with two GC bonds. The bond between bases G and C is a strong bond because it consists of 3 hydrogen bonds. That will make the primer easier to put together.

In silico analysis is an important computational prediction in primary design. Primers must also be tested through a series of optimizations in the laboratory. Primary candidate optimization involves optimization in annealing temperature (Ta) using PCR gradient and optimization of primary concentration. In addition to primers, optimization of the PCR reaction is also carried out to check the minimum detection and quantification of nucleic acids in the reaction, and this requires work in the laboratory to produce a good PCR test.

Conclusion

The primary designed can amplified the cryI gene with a size of about 200 base pairs. Annealing temperature optimization is needed to get a more specific amplicon.

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No. : 10 /ICoBioSE/2019 Attachment : -Subject : Letter of Acceptance

Dear Febriana Dwi Wahyuni, M.Si,

We are pleased to inform you that this following abstract :

Title : IN SILICO ANALYSIS FOR AMPLIFICATION CRYI GENE FROM BACILLUS THURINGIENSIS LOCAL ISOLATE

Author(s) : Febriana Dwi Wahyuni, Cindy Fransisca, Selvi Erna Pratiwi, Henny Saraswati Reg. ID : Bio00126

Has been reviewed and accepted to be presented in International Conference on Biology, Sciences and Education (ICoBioSE) to be held on October 17th – 18th,2019 at Universitas Negeri Padang, Padang City, West Sumatera, Indonesia.

- You are kindly requested to make payment amount IDR 1.500.000 for local presenter or USD 105 for international presenter, and IDR 900.000 for local student presenter or USD 63 for international student presenter no later than October 11th 2019. The payment should be transferred to Bank Negara Indonesia 1946 (BNI 46) with account number 0668110902 and account name RPL 010 BLU UNP UTK DKE. Please confirm your payment through your account by sending the scan of receipt. For student conference, please attach the scan of student ID'card.
- 2. The paper should be submitted no longer than October 2nd ,2019. All full papers will be reviewed and selected to be submitted to IOP Publisher indexed by Scopus or Jurnal Biodjati (Sinta 2). Please kindly follow the guidelines for paper's structure available on our website.
- 3. The selected papers to be published will be charged additional fee of IDR 900.000 or USD 93 for each paper and should be transferred to Bank Negara Indonesia 1946 (BNI 46) with account number 0534437975 and accound name Dezi Handayani, S.Si. It can be also paid to committee directly on the day of the conference.

We look forward to meet you in International Conference on Biology, Science and Education (ICoBioSE). Your faithfully,



Dr. Yuni Ahda, M.Si General Chair



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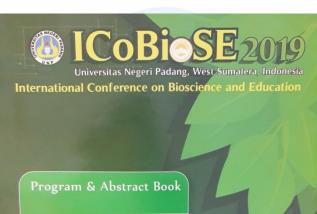
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Empowering Biological Research and Education Leads the Implementation of 4.0th Industrial Revolution Era



PARALLEL SESSION SCHEDULE

Thursday, October 17, 2019

NODIVERSITY Nod : Fitra Arya Dwi Nugraha, M.Si Ruang : BIO 1		BIOFUNCTION Mod : Dr. Rijal Satria, Ph.D Ruang : BIO 2				
	Reg. ID	Nama	Reg. ID	Nama		
No	Reg. ID	Humu	Sesi I			
1		Syazwan Saidin		Syafruddin Ilyas		
2	BI000145	Dr. Suwarno, M.SI	Bio00234	Dr. Dewi Imelda Roesma, M.Si		
3	BI000046	Mareta Widiya, M.Pd.Si	BIO00172	Dr. Haliatur Rahma, S.Si., MP		
4	BIO00133	Fatimah, S.Si	BiO00199	Dr. Hasmiwati, M.Kes		
5	BI000054	Dharma Ferry, M.Pd	BI000010	Dr. Syamsurizal		
6	BIO00164	Retno Prihatini	Bio00003	Dr. Dwi Hilda Putri, M. Biomed.		
			Sesi II			
1	Bio60115	Dra. Zuchrotus Salamah, M.Si		Dr. Djong Hon Tjong, M.Si		
2	Bio00160	Prof. Zainus Salimin	Bio00126	Febriana Dwi Wahyuni, M.Si		
3	Bio00059	Khairissa Trisliani Asmara, S.Si	Bio00131	Indri Puspita Sari, S.Si.		
4	Bio00060	Wika Mardhiyah	Bio00165	Armaita. S,KM. M.Si		
5	Bio00146	Nabela Fikriyya, S.Pd		Dasumiati		
6	Bio00117	Drs. Hadi Sasongko, M.si.	Bio00005	Dr. Violita., S.Si., M.Si		



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IN SILICOANALYSIS FOR AMPLIFICATION CRYI GENE FROM BACILLUS THURINGIENSIS LOCAL ISOLATE

FebrianaDwiWahyuni*, HennySaraswati Department of Biotechnology, Faculty of Health, UniversitasEsaUnggul, Indonesia febriana@esaunggul.ac.id

ABSTRACT

Bacillus thuringiensis is one of bacterial type that was used as a microbiological control agent for pests and vectors of plant disease. It was because in B. thuringiensisthere is Cry protein which is specific insect repellent. This Cry Protein is only toxic to certain insects. One of them is Cryl protein which is toxic to Lepidoptera insects. Lepidoptera can attack various types of plants. Cryl Proteins was encoded by cryl gene. The method can be used for cryl gene amplification is Polymerase Chain Reaction (PCR). Primer is one important component in PCR. The aim of this research was get a good primer candidate for cryl gene amplification from B. thuringiensis. In silico analysis for designing cryl primer was carried out using several software, such as BLAST for searching sequence cryl gene, Bioedit for alignment, and DINAmelt for analyze dimer structure of primer. Ten primer candidates were successfully obtained based on the result of the primer BLAST from NCBI. One primer was selected that could represent cryl gene, with forward primer 5'-GTACCGATTCCCTATGGCCG-3' and reverse primer 5'-GTACCGGTGTGACACGAAGGA-3'. Amplitication of the cryl gene by PCR in silico produces a product with a length about 770 bp.

Keywords: Bioinformatics, Cry gene, PCR in silico, Primer

Bio00136

Bio00134

UTILAZATION OF C-CINNAMALKALIX [4] RESORSINARENA OF SYNTHESIS AS ABSORBENT MATERIAL FOR DANGEROUS DYES OF RHODAMIN B

Sri Benti Etika[®], Edi Nasra

ABSTRACT

This study aims to determine the optimum conditions of Rhodamin B using C-Cinnamal Calix [4] Resonsinarena (CCCR) absorbent synthesized by UV-VIS Spectrophotometer. CCCR is synthesized by reacting cinnamaldehyde with resorcinol through an electrophilic substitution reaction. Cinnamaldehyde was obtained from isolation using cinnamon oil as much as 7.036 grams with a yield of 14.072% from 50 g cinnamon oil and CSKR produced as much as 5.6775 g brownish red. Cinnamaldehyde and CSKR were characterized using FTIR and UV-VIS. The results of the UV-VIS spectrum and FTIR data by the CSKR from isolated cinnamaldehyde showed the same results as pure cinnamaldehyde. The results of CSKR characterization using UV-VIS have absorption at a wavelength of 336 nm with an absorbance of 3,6685 Au (0,36685A) which indicates that there is a double bond conjugated to cinnamaldehyde and CSKR. The results of CSKR characterization using FTIR showed a peak of 3806.68 cm-1 indicating the presence of an O-H group. Furthermore, there is absorption at the romatic ring. Furthermore, absorption at the fingerprint wave number 1610.56 cm-1 which indicates the presence of C-C groups. At wave number 1448.90 cm-1 shows the aromatic ring. Furthermore, absorption at the fingerprint wave number 692.31 cm-1 - 836.05 cm-1 indicates the presence of (CH2). The CSKR melting point is 358° C.

Keywords: Absorbent, CCCR, Rhodamin, UV-VIS Spectrophotometer

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DOKUMENTASI ACARA

















