UNIVERSITI SAINS MALAYSIA
GERAN PENYELIDIKAN UNIVERSITI PENYELIDIKAN
LAPORAN AKHIR

CB1 CANNABINOID RECEPTOR IN BRAIN MESOLIMBIC SYSTEM OF MITRAGYNINE-SENSITISED ALBINA WASTAR RATS AS A CANDIDATE MOLECULAR TERGET IN MITRAGYNA SPECIOSA (KETUM) ADDICTION

PENYELIDIK

DR. MUZAIMI MUSTAPHA

PENYELIDIK BERSAMA

PROF. SHARIF MAHSUFI MANSOR
PROF. JAFRI MALIN ABDULLAH
PROF. HASNAN JAAFAR
NURUL IMAN WAN ISMAIL

2015

F0525/
Fasa 2/201

BORANG FRGS – P3(R)

KEMENTERIAN PENDIDIKAN MALAYSIA

FINAL REPORT
FUNDAMENTAL RESEARCH GRANT SCHEME (FRGS)
Laporan Akhir Skim Geran Penyelidikan Fundamental (FRGS)
Pindaan 2/2013

A

RESEARCH TITLE : **CB1 Cannabinoid Receptor in Brain Mesolimbic System of Mitragynine-Sensitised Albino Swiss mice as a candidate molecular targets in *Mitragyna speciosa* (Ketum) Addiction**
PHASE & YEAR : **2/2010**
START DATE : **1 April 2011**
END DATE : **31 Mac 2013**
EXTENSION PERIOD (DATE) : **30 September 2013**

PROJECT LEADER: DR MUZAIMI MUSTAPHA

PROJECT MEMBERS: 1. Profesor Sharif Mahsufi Mansor
(including GRA) 2. Profesor Jafri Malin Abdullah
3. Profesor Hasnan Jaafar
4. Nurul Iman Wan Ismail

PROJECT ACHIEVEMENT

B

ACHIEVEMENT PERCENTAGE			
Project progress according to milestones achieved up to this period	**0 - 50%**	**51 - 75%**	**76 - 100%**
Percentage (please state #%)			**95%**

RESEARCH OUTPUT		
Number of articles/ manuscripts/ books *(Please attach the First Page of Publication)*	**Indexed Journal**	***Non-Indexed Journal***
	1	
Conference Proceeding *(Please attach the First Page of Publication)*	**International**	***National***
	1	
Intellectual Property *(Please specify)*	**Nil**	

HUMAN CAPITAL DEVELOPMENT

Human Capital	Number				Others (please specify)
	On-going		Graduated		
Citizen	Malaysian	Non Malaysian	Malaysian	Non Malaysian	
PhD Student					
Master Student	1				
Undergraduate Student					
Total	1				

EXPENDITURE

C

Budget Approved *(Peruntukan diluluskan)* :
Amount Spent *(Jumlah Perbelanjaan)* :

Balance *(Baki)* :
Percentage of Amount Spent :
(Peratusan Belanja)

ADDITIONAL RESEARCH ACTIVITY

D

International		
Activity	Date (Month, Year)	Organizer
Oral Presentation	26-28 July, 2013	2013 International Symposium on Biological Engineering and Natural Sciences (ISBENS), Bangkok
National		
Activity	Date (Month, Year)	Organizer
Poster Presentation	22-23 June, 2011	16th National Conference on Medical & Health Sciences, Kota Bharu

PROBLEMS / CONSTRAINTS IF ANY

F

RECOMMENDATION

F

[illegible]

G	Emerging new psychoactive substances impose public health implications worldwide, and include plants' phytochemicals such as Mitragynine (MG) from Mitragyna speciosa (or 'Ketum'). Studies have demonstrated the reciprocal interaction between endocannabinoid (ECB) system and opioid systems in the modulation of brain-reward and neurobiological mechanisms underlying drug addiction. To date the neurochemical basis of Ketum abuse potential remains elusive. This research attempted to implicate ECB system in the abuse liabilities of emerging psychoactive plant, Ketum and its opioid-like alkaloid, mitragynine (MG). We had demonstrated the locomotor and behavioural effects of MG-sensitised Swiss albino mice in IntelliCage® using an approach of context-independent sensitisation. Animals were later sacrificed for immunohistochemical localisation of CB1 receptor in the brain with confirmatory analysis using Western blotting of the brain homogenates. Adult Albino Swiss mice were subjected to experimental groups of MG alone; MG + NIDA-41020 (CB1 receptor antagonist); Morphine sulphate; Δ-9-Tetrahydrocannabinol + NIDA-41020; and vehicle. Daily intraperitoneal injection (i.p) of MG (5 to 25 mg/kg; 5mg/kg increment) at a 6-day interval was administered for 30 days, and plus once daily, 20mg NIDA-41020 (oral). Findings to date showed neuroadaptive chronic exposure of morphine, and works are in its final phase for comparison with MG and in the presence of CB1 antagonist. Data analysis to date provide supportive ground for the first time to our knowledge, to indicate the involvement of CB1 receptor as the neural target of Ketum misuse potential. (Two manuscripts in preparation for potential publication in indexed-journals).

Date : **6 Feb 2014**
Tarikh

Project Leader's Signature:
Tandatangan Ketua Projek

COMMENTS, IF ANY/ENDORSEMENT BY RESEARCH MANAGEMENT CENTRE (RMC)
(Komen, sekiranya ada/Pengesahan oleh [illegible])

H ....

....

....

Name:
Nama:

Signature:
Tandatangan:

Date: 13/3/14
Tarikh:

JABATAN BENDAHARI
KUMPULAN WANG PENYELIDIKAN FUNDAMENTAL
PENYATA PERBELANJAAN SEHINGGA 31 DISEMBER 2013

Jumlah Geran	Ketua Projek	DR MUZAIMI MUSTAPHA
Peruntukan (Tahun 1) 2011	Tajuk Projek	CB1 CANNABINOID RECEPTOR IN BRAIN MESOLIMBIC SYSTEM OF MITRAGYNINE-SENSITISED ALBINO WISTAR RATS AS A CANDIDATE MOLECULAR TARGET IN MITRAGYNA SPECIOSA (KETUM) ADDICTION
Peruntukan (Tahun 2) 2012		
	Tempoh	30 BULAN (01/04/2011-30/09/2013)
	No. Akaun	

Kwgan	Akaun	PTJ	Projek	Peruntukan Projek	Perbelanjaan Terkumpul sehingga Tahun lalu	Peruntukan Semasa	Tanggungan Semasa	Bayaran Tahun Semasa	Belanja Tahun Semasa	Baki Projek
203	11000 PPSP		6171132	28,800.00	12,938.18	15,861.82	-	11,835.01	11,835.01	4,026.81
203	14000 PPSP		6171132			-	-		-	-
203	15000 PPSP		6171132	-		-	-		-	-
203	21000 PPSP		6171132	5,500.00	5,045.20	454.80			-	454.80
203	22000 PPSP		6171132	-		-	-		-	-
203	23000 PPSP		6171132			-	-		-	-
203	24000 PPSP		6171132		24.00	(24.00)			-	(24.00)
203	26000 PPSP		6171132			-	-		-	-
203	27000 PPSP		6171132	29,300.00	20,471.92	8,828.08	4,192.00	8,205.28	12,397.28	(3,569.20)
203	28000 PPSP		6171132			-			-	-
203	29000 PPSP		6171132	4,400.00	2,726.50	1,673.50		1,298.20	1,298.20	375.30
203	35000 PPSP		6171132			-			-	-
203	A11102 PPSP		6171132	-	628.95	(628.95)			-	(628.95)
203	A11559 PPSP		6171132	-	-	-			-	-

Neuroscience and Biobehavioral Reviews 37 (2013) 138–151

Neuroscience and Biobehavioral Reviews

Review

From Kratom to mitragynine and its derivatives: Physiological and behavioural effects related to use, abuse, and addiction

Zurina Hassan[a,1], Mustapha Muzaimi[b,1], Visweswaran Navaratnam[a], Nurul H.M. Yusoff[a], Farah W. Suhaimi[a], Rajakumar Vadivelu[a], Balasingam K. Vicknasingam[a], Davide Amato[c], Stephan von Hörsten[d], Nurul I.W. Ismail[b], Nanthini Jayabalan[b], Ammar I. Hazim[a], Sharif M. Mansor[a], Christian P. Müller[c,*]

[a] Centre for Drug Research, Universiti Sains Malaysia, 11800 Minden, Penang, Malaysia
[b] Department of Neurosciences, School of Medical Sciences, Universiti Sains Malaysia, Health Campus, 16150 Kubang Kerian, Kelantan, Malaysia
[c] Department of Psychiatry and Psychotherapy, Friedrich-Alexander-University Erlangen-Nuremberg, Schwabachanlage 6, 91054 Erlangen, Germany
[d] Franz-Penzoldt Center, Experimental Therapy, Friedrich-Alexander-University Erlangen-Nuremberg, Germany

ARTICLE INFO

Article history:
Received 6 August 2012
Received in revised form 8 October 2012
Accepted 22 November 2012

Keywords:
Kratom
Ketum
Mitragyna speciosa
Mitragynine
7-Hydroxymitragynine
Analgesia
Abuse
Addiction

ABSTRACT

Kratom (or Ketum) is a psychoactive plant preparation used in Southeast Asia. It is derived from the plant *Mitragyna speciosa* Korth. Kratom as well as its main alkaloid, mitragynine, currently spreads around the world. Thus, addiction potential and adverse health consequences are becoming an important issue for health authorities. Here we reviewed the available evidence and identified future research needs. It was found that mitragynine and *M. speciosa* preparations are systematically consumed with rather well defined instrumentalization goals, e.g. to enhance tolerance for hard work or as a substitute in the self-treatment of opiate addiction. There is also evidence from experimental animal models supporting analgesic, muscle relaxant, anti-inflammatory as well as strong anorectic effects. In humans, regular consumption may escalate, lead to tolerance and may yield aversive withdrawal effects. Mitragynine and its derivatives' actions in the central nervous system involve μ-opioid receptors, neuronal Ca^{2+} channels and descending monoaminergic projections. Altogether, available data currently suggest both, a therapeutic as well as an abuse potential.

Contents

* Corresponding author at: Section of Addiction Medicine, Department of Psychiatry and Psychotherapy, Friedrich-Alexander-University Erlangen-Nuremberg, Schwabachanlage 6, Erlangen 91054, Germany. Tel.: +49 0 9131 85 36806.
E-mail address: Christian.Mueller@uk-erlangen.de (C.P. Müller).
[1] These authors contributed equally.

0149-7634/$ - see front matter

Title: Use of the IntelliCage system to assess the behavioural effects in Swiss albino mice following chronic morphine treatment

Authors: Nurul Iman Wan Ismail, NanthiniJayabalan, Muzaimi Mustapha.

Affiliations: Department of Neurosciences, School of Medical Sciences, Universiti Sains Malaysia, Health Campus, 16150 Kubang Kerian, Kelantan.

ABSTRACT

Background/Objective: Morphine, being the most efficacious and widely prescribed opioid, is subjects to abuse liability. Adaptive changes in neurons are a hallmark of chronic morphine treatment and are related to altered behaviours associated with morphine dependence and withdrawal syndrome. Studies had shown the possibility of learning and memory interference with chronic morphine exposure. However, multi-dimensional study of animal models of morphine addiction remains scarce. To address this issue experimentally, we performed a longitudinal study using the IntelliCage automated learning system, which allows the detailed analysis of mouse behaviour in social groups.

Methods: Male Swiss albino mice were randomly assigned to either the control (n=6) or morphine-treated group (n=6). Mice were subjected to a 28-day regimen with morphine sulphate (5 mg/kg, s.c.). Control animals received an equal volume of saline (1 ml/kg) and were administered intraperitoneally (i.p.). The IntelliCage were used as the setting for the context-independent behavioural sensitisation to observe exploratory behaviours and cognitive performances of the animals.Protocols covering circadian exploration, motivation for natural reward with/without air-puff punishment, place learning and reversal place learning were used for longitudinal characterisation of behaviour.

Results: Differences in exploratory activities were significant where the morphine group showed behavioural inflexibility and shifted circadian rhythms compared with their baseline activity and the control group (P<0.05). Overall, both groups showed significant increment in lick preference at sucrose reward corner (P<0.001), where the morphine group had significantly higher preference for sucrose corner than control group (P<0.05). Both groups showed significant reduction in the number of visits to the sucrose reward corner when it was associated with an air-puff punishment(P<0.001). However, no significance difference was observed between morphine and control group in their resistance to punishment(P>0.05), as measured by the reduction in sucrose consumption despite air-puff punishment. In the place