THE INHIBITION OF HUMAN IMMUNODEFICIENCY VIRUS-1 INTEGRATION BY DESIGNED ZINC FINGER PROTEIN

SUPACHAI SAKKHACHORNPHOP

DOCTOR OF PHILOSOPHY
IN BIOMEDICAL SCIENCE

THE GRADUATE SCHOOL
CHIANG MAI UNIVERSITY
AUGUST 2011

THE INHIBITION OF HUMAN IMMUNODEFICIENCY VIRUS-1 INTEGRATION BY DESIGNED ZINC FINGER PROTEIN

SUPACHAI SAKKHACHORNPHOP

A THESIS SUBMITTED TO THE GRADUATE SCHOOL IN

PARTIAL FULFILLMENT OF THE REQUIREMENTS

FOR THE DEGREE OF

DOCTOR OF PHILOSOPHY

IN BIOMEDICAL SCIENCE

THE GRADUATE SCHOOL
CHIANG MAI UNIVERSITY
AUGUST 2011

THE INHIBITION OF HUMAN IMMUNODEFICIENCY VIRUS-1 INTEGRATION BY DESIGNED ZINC FINGER PROTEIN

SUPACHAI SAKKHACHORNPHOP

THIS THESIS HAS BEEN APPROVED

TO BE A PARTIAL FULFILLMENT OF THE REQUIREMENTS

FOR THE DEGREE OF DOCTOR OF PHILOSOPHY

IN BIOMEDICAL SCIENCE

EXAMINING COMMITTEE	THESIS ADVISORY COMMITTEE
Kill Person CHAIRPERSON	Hutelin Ogypnin ADVISOR
Prof. Kiat Ruxrungtham, M.D.	Assoc. Prof. Dr. Chatchai Tayapiwatana
But Just MEMBER	
Assoc Prof. Dr. Bruce E. Torbett	Prof. Dr. Carlos F. Barbas, III
Hotelin Oonefall MEMBER	Co-Ve - CO-ADVISOR
Assoc. Prof. Dr. Chatchai Tayapiwatana	Prof. Dr. Watchara Kasinrerk
CV_h_C MEMBER	CO-ADVISOR
Prof. Dr. Watchara Kasinrerk	Prof. Thira Sirisanthana, M.D.
Like frilling MEMBER	

Prof. Thira Sirisanthana, M.D.

ACKNOWLEDGEMENTS

This thesis arose on several years of research that has been done since I came to Assoc. Prof. Dr. Chatchai's Lab. By that time, I have worked with a great number of people whose contribution in assorted ways on this research road. This thesis would not have been possible unless all these special people and funding. It is a pleasure to convey my gratitude to them all in my acknowledgments.

First and foremost, I would like to express my deepest gratitude and sincere appreciation to my thesis advisor, Assoc. Prof. Dr. Chatchai Tayapiwatana for giving me the opportunity to be a Ph.D student in his laboratory. He provided me the supervision, advice, and excellent guidance, valuable suggestions, extraordinary experiences and many encouragements from the very early stage throughout the period of study including to obtain the financial support from the Thailand Research Fund through the Royal Golden Jubilee Ph.D program. His truly scientist intuition has made him as a constant oasis of ideas and passions in science, which exceptionally inspire and enrich my growth and maturation.

It has been an honor to be a member for 6 months in laboratory of Prof. Dr. Carlos F. Barbas, III, my co-advisor at the Skaggs Institute for Chemical Biology and the Departments of Molecular Biology and Chemistry, the Scripps Research Institute, La Jolla, California, USA, for working on zinc finger protein and lentiviral vector technology. I am extremely grateful to thank for his help in providing me with excellent guidance, constructive suggestions and comments, and kind supports throughout my study, and revision of my manuscripts. I'm also thankful for Dr. Beatriz Gonzalez Alonso, Roberta Fuller, Thomas Gaj and Mishelle McClanahan-

Shinn for technical assistance and everybody in this laboratory for their warm friendship and kind help during working in La Jolla.

My deepest gratitude goes to my co-advisor, Prof. Dr. Watchara Kasinrerk, for his valuable suggestions kindly provide some monoclonal antibodies, and equipments throughout my thesis. I would also like to thank members in this laboratory for their friendship, kindness, and contributions for technical supports.

For the continued supports from master degree through Ph.D. program, I would like to express my sincere gratitude to my co-advisor, Professor Thira Sirisanthana, for his kindness, encouragement, valuable suggestions, providing the financial support from the Fogarty AIDS International for Research and Training Program, the Fogarty International Clinical Research Training for Scholars and Fellows Program, Johns Hopkins University, and support the construction and facilities for viral culture experiments at the Research Institute for Health Sciences (RIHES), Chiang Mai University. I am particularly indebted to thanks Mr. Rassamee Keawvichit, Mrs. Kanlaya Wongworapat, and all the staff in clinical laboratory of the RIHES for their kind help and support in viral load assay. I appreciate Mr. Peter Lange, Mrs. Paphawadee Damrongmanee, and all staff of administrative office, and extend thanks a librarian, Ms. Tippawan Kunkaew, for the best service, their friendship.

For the assistance to access the BIACORE 2000TM biosensor, my grateful thanks go to Assoc. Prof. Dr. Prachya Kongtawelert, Faculty of Medicine, Chiang Mai University and thank to Mr. Kanchanok Kodchakorn for technical support.

The Faculty of Allied Health Science, Chulalongkorn University and Rushmore Precision co., Ltd, have been a source to acquire many high quality pictures by a confocal laser scanning microscope LSM 700 (Carl Zeiss MicroImaging, Germany) and ZEN 2010 software.

Warm friendship, suggestions, and kind assistance from Asst. Prof. Dr. Khajornsak Tragoolpua, Asst. Prof. Dr. Songyot Anuchapreeda and sisters of lineage student code: Lect. Dr. Nutjeera Intasai and Asst. Prof. Dr. Sawitree Chiampanichayakul were appreciated in deepest in my mind.

I thank to Mr. Wutigri Nimlamool, Mr. Somphot Saoin, Dr. Sawitree Nangola, and all the staff in Assoc. Prof. Dr. Chatchai's Lab for kind helps, warm friendship. I have appreciated to Mrs. Kongkham Kruawan for her kindness in providing lunch boxes every day. The best services in official document preparations were obtained by kindness and friendship of Mrs. Tuanjai Ponsung. Thank you so much.

This work was supported by the Thailand Research Fund through the Royal Golden Jubilee Ph.D program (grant number PHD/0112/2550), the Fogarty AIDS International for Research and Training Program, Johns Hopkins University, the National Center for Genetic Engineering and Biotechnology (BIOTEC) of the National Science and Technology Development Agency, Thailand, and the U.S. National Institutes of Health grant GM065059 (CFB), and the National Research University Project under Thailand's Office of the Higher Education Commission for partial financial support.

The proof of English usage in this thesis was mainly from the valuable help of Assoc. Prof. Dr. Bruce E. Torbett an invited committee from the Scripps Research Institute, La Jolla, California, USA.

Finally, I deeply express my special thanks to my parents, and my sister Tassanee Sakkhachornphop as a second mom to take care my family throughout my time in USA. Indispensably, I thanks to my wife Pranee Sakkhachornphop, son N' Angpao, and daughter N' Imm for their love, patience, understanding, and encouragement.

Supachai Sakkhachornphop

ลิขสิทธิ์มหาวิทยาลัยเชียงใหม Copyright[©] by Chiang Mai University All rights reserved