UNIVERSITI TEKNOLOGI MARA

COMPUTATIONAL ANALYSIS ON MISSENSE MUTATION OF ORNITHINE TRANSCARBAMYLASE (OTCase) ENZYME

ERNIE ZURAIDA BINTI ALI

Thesis submitted in fulfillment
of the requirements for the degree of
Doctor of Philosophy
(Pharmacoinformatics)

Faculty of Pharmacy

February 2020

AUTHOR'S DECLARATION

I declare that the work in this thesis was carried out in accordance with the regulations of Universiti Teknologi MARA. It is original and is the results of my own work, unless otherwise indicated or acknowledged as referenced work. This thesis has not been submitted to any other academic institution or non-academic institution for any degree or qualification.

I, hereby, acknowledge that I have been supplied with the Academic Rules and Regulations for Post Graduate, Universiti Teknologi MARA, regulating the conduct of my study and research.

Name of Student	:	Ernie Zuraida Binti Ali
Student I.D. No.	:	2014440844
Programme	:	Doctor of Philosophy – PH951
Faculty	:	Pharmacy
Thesis	:	Computational Analysis on Missense Mutation of Ornithine Transcarbamylase (OTCase) Enzyme
Signature of Student	:	.....
Date	:	February 2020

ABSTRACT

Missense mutations occur close to the binding pocket of ornithine transcarbamylase (OTCase) enzyme associated with a severe form of ornithine transcarbamylase deficiency (OTCD), an X-linked disorder. These mutations affect the functions of OTCase enzyme in the body that can lead to acute clinical symptoms. Early diagnosis and intervention is crucial for effective decision on managing patients and reducing mortality rate. However, the current diagnosis and biochemical analyses being used are still time consuming, labour intensive and costly. Therefore, in this work, three (3) stages of computational approaches (mutation prediction servers, molecular docking, molecular dynamics simulations) are explored and implemented to predict the severity of the missense mutations in OTCD. The findings are based on two novels, Q171H and N199H, and 30 other known missense mutations located in the OTCase ligand binding pocket. The novel mutations were obtained from the nucleotide sequencing results from two OTCD Malaysian patients, while the other 30 known mutations were collected from HGMD and 1000G browser databases. The consensus prediction of five mutation prediction servers concluded all 32 mutations are disease-causing. Further study using docking of PALO ligand, an analog of N-phosphonacetyl-L-ornithine resulted in binding of the ligand to the mutant OTCase structures with slightly differently conformation when compared to the wild type structure. In the final stage, MD simulations of the mutant complexes (Q171H-PALO, N199H-PALO, R92P-PALO and H168R-PALO) confirmed the conformational instability due to the disruption of intermolecular interactions between PALO and OTCase. The missense mutations disrupted hydrogen bond interactions with the neighbouring residues (S267, M268, D263, C303, R330 and E326), including residues involved in the catalytic mechanism (C303, D263 and R330). In addition, the role of SMG loop as a second ligand recognition site was observed during the simulations. Application of computational approaches successfully revealed the mutations potentially disturb catalytic efficiency contributing to the occurrence of various OTCD symptoms. In conclusion, computational approaches can potentially be used as preliminary screening and rapid diagnosis for new mutation as it can provide the fast result, save time and cost.

ACKNOWLEDGEMENT

In the name of Allah, the Most Loving and the Most Merciful. I would like to thank and praise the Allah for strength, health and passion. He has lighting my way and directing me through each and every success to complete this thesis.

I would like to thank and express my full appreciation to my supervisor, Dr Siti Azma Jusoh, for her encouragement, guidance, patience and constructive critics during my research work. I thank to Dr Yuslina Zakaria for her understanding, motivational supports and believing me to stay on the course.

I must acknowledge Institute for Medical Research and Ministry of Health for the awarded study leaves. Special thanks to my lovely friend, Zalikha Ibrahim for her assistance and willingness to share her knowledge. Thanks also goes to my friends, Nur Azira Ismail and Mohd Naim Fadhli for being such a good support team.

I would like to express my sincere gratitude, thanks and love to my father, Hj Ali bin Alias, my late mother, Hjh Salnah bt Ali and my mother-in-law, Che'e Buang bt Haron for their continuous encouragement during my period of study. Thanks also to all my sisters and brothers for their support and care all these years.

Finally, I would like to express my appreciation, thanks and love to my husband, Associate Professor Dr Mohd Amran Mohd Radzi for his unconditional love, understanding, care, supplications and endurance throughout the most difficult periods of this work. To his goes a great part of my gratitude.

I believed without those supports, I would not finish this thesis. I also believed that this journey is not only about me, but also about us. Alhamdulillah.

TABLE OF CONTENT