EFFECTS OF BORTEZOMIB ON HIF-1 AND HIF-2 TRANSCRIPTIONAL ACTIVITIES

NORAINI BINTI ABD AZIZ

FBSB 2013 39
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NORAINI BINTI ABD AZIZ

MASTER OF SCIENCE
UNIVERSITI PUTRA MALAYSIA

2013
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By

NORAINI BINTI ABD AZIZ

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfillment of the Requirements for the Degree of Master of Science

November 2013
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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfillment of the requirement for the degree of Master of Science

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By

NORAINI BINTI ABD AZIZ

November 2013

Chairman : Norazizah Shafee, PhD

Faculty : Biotechnology and Biomolecular Sciences

Bortezomib is the first proteasomal inhibitor (PI) to be used therapeutically in humans for treating relapse cases of multiple myeloma and mantle cell lymphoma. A proposed mechanism is that it prevents proteasomal degradation of pro-apoptotic proteins, leading to enhance apoptosis. Although the alpha subunit of hypoxia inducible factor 1 (HIF-1α) is not degraded, the heterodimeric HIF-1 fails to transactivate target genes. HIF-1 and HIF-2 are related hypoxia-inducible transcription factors that are important for survival of hypoxic tumor cells. Most reports have focused on the effects of bortezomib on HIF-1 but not HIF-2 transcriptional activities. In the present study, the effect of bortezomib on
HIF-2 activity in cells with different levels of expression of the HIF-1α and HIF-2α subunits, was investigated. Results showed that bortezomib treatment suppressed the transcription and expression of CA9, a HIF-1-specific target gene, but had minimal effects on EPO and GLUT-1, which are the target genes of both HIF-1 and HIF-2. A similar dichotomy of responses was also seen with exogenously-introduced hypoxia response elements of CA9 and EPO. These data led to a conclusion that bortezomib attenuates the transcriptional activity of only HIF-1 but not HIF-2. This novel finding on the lack of inhibitory effect of bortezomib on HIF-2 transcriptional activity will be important in the improvement of design and treatment modalities to enhance the efficacy of this and other proteasomal inhibitor drugs.
KESAN BORTEZOMIB TERHADAP AKTIVITI TRANSKRIPTI HIF-1 DAN HIF-2

Oleh

NORAINI BINTI ABD AZIZ

November 2013

Pengerusi : Norazizah Shafee, PhD

Faculty : Bioteknologi dan Sains Biomolekul

Bortezomib adalah perencat proteosomal yang pertama digunakan secara terapeutik pada manusia bagi merawat kes-kes berulang seperti mieloma berbilang dan limfoma sel mantel. Satu mekanisma yang dicadangkan adalah bortezomib berupaya menghalang degradasi proteosomal protein-protein pro-apoptosis yang menyebabkan peningkatan apoptosis. Walaupun subunit faktor induksi hipoksia 1α (HIF-1α) tidak didegradasi, heterodimer HIF-1 gagal untuk mengaktifkan gen sasaran. HIF-1 and HIF-2 adalah faktor induksi transkripsi hipoksia yang penting untuk kelangsungan hidup sel-sel tumor hipoksia. Banyak laporan terdahulu memberi tumpuan kepada kesan bortezomib terhadap aktiviti transkripsi HIF-1 sahaja, tetapi tidak HIF-2. Dalam kajian ini, kesan
bortezomib terhadap aktiviti HIF-2 di dalam sel-sel yang mempunyai tahap ekspresi HIF-1 dan HIF-2 yang berbeza telah dikaji. Hasil yang diperoleh dalam kajian ini menunjukkan bahawa rawatan menggunakan bortezomib dapat merencatkan transkripsi dan ekspresi CA9, iaitu gen sasaran khusus bagi HIF-1. Walaubagaimanapun, ia mempunyai kesan minimum terhadap EPO dan GLUT-1. Tindak balas yang sama juga telah dilihat dalam unsur respon hipoksia yang dibawa secara eksogen bagi CA9 dan EPO. Data-data ini membawa kepada kesimpulan bahawa bortezomib hanya merencatkan aktiviti transkripsi HIF-1, tetapi tidak HIF-2. Pengetahuan mengenai kekurangan kesan bortezomib terhadap aktiviti transkripsi HIF-2 boleh menjadi penyumbang ke arah strategi untuk peningkatan keberkesan reka bentuk serta kaedah rawatan menggunakan dadah ini atau perencat proteosom yang lain.
ACKNOWLEDGEMENTS

In the name of Allah, The Most Gracious, The Most Merciful.

First and foremost, I thank Allah S.W.T for His blessing and giving me the strength to complete this research.

My deepest gratitude to my supervisors, Assoc. Prof. Dr. Norazizah Shafee, Prof. Dr. Eric J. Stanbridge, and Dr. Abhimanyu A/L Veerakumarasivam for their most invaluable guidance, endless encouragement, help and patience throughout this project and for the critical review in the completion of this thesis.

I wish to express my deepest appreciation to all the members of Laboratory of Virology 143 who have shared their knowledge with me and made my time in the laboratory most enjoyable.

Special thanks to those who have contributed directly or indirectly to my research and thesis.

Last but not least, my endless gratitude and thanks to my parents, my only sister and brother in law for being the greatest support and for giving me the strength needed to achieve all my aims and dreams. Without them, I would never have been able to achieve so much.
I certify that a Thesis Examination Committee has met on 7 November 2013 to conduct the final examination of Noraini binti Abd Aziz on her thesis entitled "Effects of Bortezomib on HIF-1 and HIF-2 Transcriptional Activities" in accordance with the Universities and University Colleges Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U.(A) 106] 15 March 1998. The Committee recommends that the student be awarded the Master of Science.

Members of the Thesis Examination Committee were as follows:

**Janna Ong Abdullah, PhD**  
Associate Professor  
Faculty of Biotechnology and Biomolecular Sciences  
Universiti Putra Malaysia  
(Chairman)

**Syahrilnizam Abdullah, D.Phil**  
Associate Professor  
Faculty of Medicine and Health Sciences  
Universiti Putra Malaysia  
(Internal Examiner)

**Muhajir Hamid, PhD**  
Associate Professor  
Faculty of Biotechnology and Biomolecular Sciences  
Universiti Putra Malaysia  
(Internal Examiner)

**Chua Kek Heng, PhD**  
Professor  
Faculty of Medicine  
Universiti Malaya  
(External Examiner)

______________________________
NORITAH OMAR, PhD  
Associate Professor and Deputy Dean  
School of Graduate Studies  
Universiti Putra Malaysia

Date: 21 January 2013
This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfillment of the requirement for the degree of Master of Science. The members of the Supervisory Committee were as follows:

**Norazizah Shafee, PhD**  
Associate Professor  
Faculty of Biotechnology and Biomolecular Sciences  
Universiti Putra Malaysia  
(Chairman)

**Eric J. Stranbridge, PhD**  
Distinguished Professor  
School of Medicine  
University of California, Irvine, USA  
(Member)

**Abhimanyu A/l Veerakumarasivam, PhD**  
Senior Lecturer  
Faculty of Medicine and Health Sciences  
Universiti Putra Malaysia  
(Member)

__________________________________________

**BUJANG BIN KIM HUAT, PhD**  
Professor and Dean  
School of Graduate Studies  
Universiti Putra Malaysia  
Date:
DECLARATION

I declare that the thesis is my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously, and is not concurrently submitted for any other degree at Universiti Putra Malaysia or at any other institutions.

_________________________
NORAINI BINTI ABD AZIZ

Date: 7 November 2013
TABLE OF CONTENTS

ABSTRACT ii
ABSTRAK iv
ACKNOWLEDGEMENTS vi
APPROVAL vii
DECLARATION ix
LIST OF TABLE xiii
LIST OF FIGURES xiv
LIST OF APPENDICES xvi
LIST OF ABBREVIATIONS xvii

CHAPTER

1 INTRODUCTION 1

2 LITERATURE REVIEW 4
  2.1 Hypoxia 4
  2.2 Adaptation to hypoxia 5
  2.3 Hypoxia-inducible factor 6
    2.3.1 HIF-1α 7
    2.3.2 HIF-2α 10
  2.4 Regulation of HIF transcriptional activity 10
  2.5 Distinct functions of HIF-1α and HIF-2α 13
  2.6 HIF target genes and their functions in cells 14
    2.6.1 Glucose metabolism 14
    2.6.2 Erythropoiesis 15
    2.6.3 pH control 15
  2.7 The role of HIF in cancer 16
    2.7.1 HIF in renal cancer cells 16
    2.7.2 HIF in breast cancer cells 17
  2.8 HIF as a target molecule in cancer therapy 18
  2.9 Ubiquitin proteasome pathway 18
    2.9.1 Proteasome inhibitiior: Bortezomib 21
### 3 MATERIALS AND METHODS

<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>3.1 Materials</td>
<td>23</td>
</tr>
<tr>
<td>3.1.1 Chemical and reagents</td>
<td>23</td>
</tr>
<tr>
<td>3.1.2 Instruments</td>
<td>23</td>
</tr>
<tr>
<td>3.1.3 Source of plasmids and cell lines</td>
<td>25</td>
</tr>
<tr>
<td>3.1.4 Proteasomal inhibitor</td>
<td>25</td>
</tr>
<tr>
<td>3.2 Maintenance of cell culture</td>
<td>26</td>
</tr>
<tr>
<td>3.2.1 Reconstitution of cells from liquid nitrogen storage</td>
<td>26</td>
</tr>
<tr>
<td>3.2.2 Culturing of cells</td>
<td>26</td>
</tr>
<tr>
<td>3.2.3 Cryopreservation of cells</td>
<td>27</td>
</tr>
<tr>
<td>3.2.4 Mycoplasma test</td>
<td>28</td>
</tr>
<tr>
<td>3.3 Bortezomib treatment</td>
<td>28</td>
</tr>
<tr>
<td>3.4 Cell viability assay</td>
<td>29</td>
</tr>
<tr>
<td>3.5 Preparation of cell lysate</td>
<td>29</td>
</tr>
<tr>
<td>3.5.1 Protein extraction</td>
<td>29</td>
</tr>
<tr>
<td>3.5.2 Determination of protein concentration</td>
<td>30</td>
</tr>
<tr>
<td>3.5.3 SDS-PAGE and Western blotting</td>
<td>32</td>
</tr>
<tr>
<td>3.5.4 Immunoblotting and immunodetection</td>
<td>33</td>
</tr>
<tr>
<td>3.6 Preparation and analysis of RNA samples</td>
<td>34</td>
</tr>
<tr>
<td>3.6.1 RNA extraction</td>
<td>34</td>
</tr>
<tr>
<td>3.6.2 Confirmation of RNA integrity</td>
<td>36</td>
</tr>
<tr>
<td>3.6.3 Primers for reverse transcriptase-polymerase chain reaction (RT-PCR)</td>
<td>37</td>
</tr>
<tr>
<td>3.6.4 RT-PCR</td>
<td>37</td>
</tr>
<tr>
<td>3.7 Preparation of reporter plasmids</td>
<td>39</td>
</tr>
<tr>
<td>3.7.1 Transformation of reporter plasmids</td>
<td>39</td>
</tr>
<tr>
<td>3.7.2 Small scale plasmid extraction</td>
<td>40</td>
</tr>
<tr>
<td>3.7.3 Restriction enzyme digestion of plasmids</td>
<td>41</td>
</tr>
<tr>
<td>3.7.4 Agarose gel electrophoresis</td>
<td>42</td>
</tr>
<tr>
<td>3.7.5 Large scale plasmid extraction</td>
<td>42</td>
</tr>
<tr>
<td>3.8 Transient transfection</td>
<td>44</td>
</tr>
<tr>
<td>3.8.1 Transfection optimization for firefly and Renilla luciferase expression</td>
<td>44</td>
</tr>
<tr>
<td>3.8.2 Transfection using Lipofectamie 2000</td>
<td>44</td>
</tr>
<tr>
<td>3.8.3 Luciferase reporter assay</td>
<td>46</td>
</tr>
<tr>
<td>3.9 Data quantitation and statistical analyses</td>
<td>47</td>
</tr>
</tbody>
</table>

### 4 RESULTS AND DISCUSSION

<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>4.1 Mycoplasma test</td>
<td>48</td>
</tr>
<tr>
<td>4.2 Effects of bortezomib on cancer cells</td>
<td>50</td>
</tr>
</tbody>
</table>