



UNIVERSITI PUTRA MALAYSIA

**MATRIX METALLOPROTEINASE-9 AND TISSUE INHIBITORS OF
METALLOPROTEINASE -1 AND -2 AS POTENTIAL NOVEL
BIOMARKERS IN GESTATIONAL HYPERTENSION**

JABRULLAH AB. HAMID

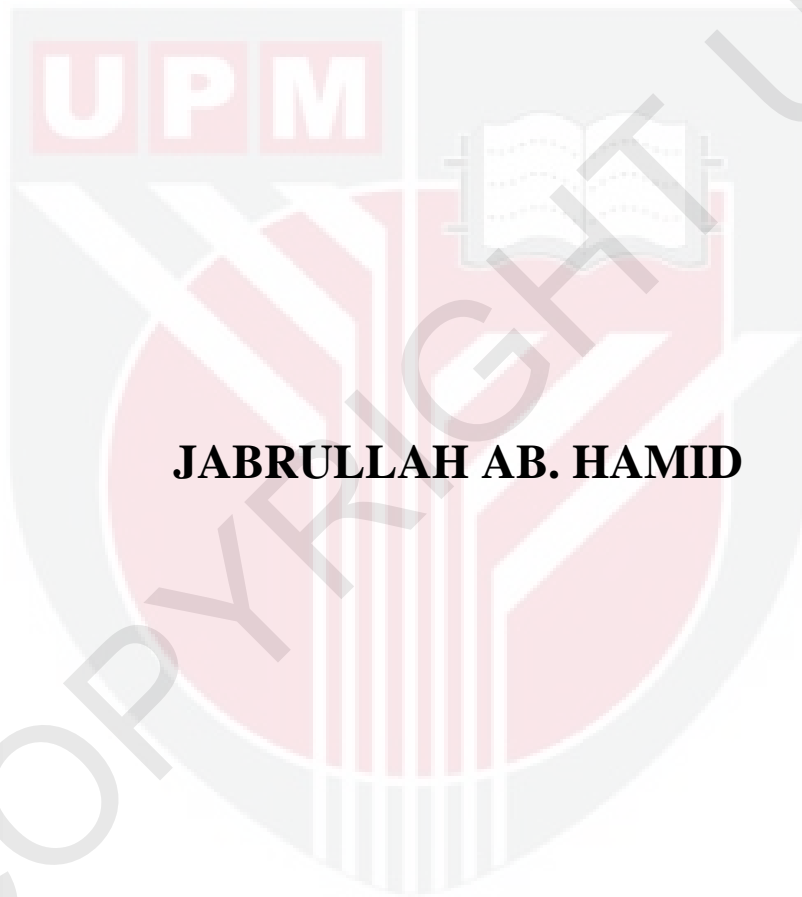
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By

JABRULLAH AB. HAMID

**Thesis Submitted to the School of Graduates Studies, Universiti Putra Malaysia,
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November 2010

Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirements for the degree of Master of Science

MATRIX METALLOPROTEINASE-9 AND TISSUE INHIBITORS OF METALLOPROTEINASE-1 AND -2 AS POTENTIAL NOVEL BIOMARKERS IN GESTATIONAL HYPERTENSION

By

JABRULLAH AB HAMID

November 2010

Chairman: Norhafizah Mohtarrudin, MBBS, M (Path)

Faculty: Faculty of Medicine and Health Sciences

Gestational hypertension (GH) is a common disorder during pregnancy, which could progress to preeclampsia (PE) and even causing various subsequent maternal and perinatal complications. A cluster of enzymes called matrix metalloproteinase (MMP) and tissue inhibitors of metalloproteinase (TIMP) are thought to be involved in the pathophysiology of the disease. The purpose of this study was to determine the circulating (serum) levels of the MMP-9, TIMP-1 and TIMP-2 in GH patients and compare with normotensive pregnant women as well as to correlate the pregnancy outcomes, such as preterm delivery, poor progress, foetal distress, mode of delivery, low birth weight, small for gestational age, low apgar score, meconium-stained liquor and neonatal intensive care unit admission. A total of 118 patients were randomly selected in this unmatched case-control study. Subjects include GH women (n=54) and controls (n=64) were healthy normotensive pregnant women, selected from those who have attended the outpatient clinics at General Hospital Kuala Lumpur and

Serdang Hospital during May 2008 to January 2009 and fulfilled the specified selection criteria. All the patients were followed up until delivery, all desired patients' information and the outcomes of the pregnancies were gathered from medical records. The levels of each biomarker were measured by using specific Enzyme-Link Immunosorbent Assay (ELISA) technique and the MMP/TIMPs ratios were calculated manually. MMP-9 was found slightly higher in the GH group ($p=0.085$). In the subgroup analysis based on parity, MMP-9 and MMP-9/TIMP-1 ratio were slightly higher in nulliparous GH group ($p=0.064$ and $p=0.079$ respectively), and TIMP-1 was slight higher in multiparous GH group ($p=0.062$). However all these findings do not reach statistical significance at $p=0.05$. A weak positive correlation (statistically significant) was found between maternal age and TIMP-1 in GH group ($r=0.278$, $p=0.047$). Similarly, a weak positive correlation (statistically significant) was found between gestational age and TIMP-2 in control group ($r=0.318$, $p=0.010$). There is no correlation found between MMP and TIMPs with blood pressure levels. Pre-eclampsia developed in 8 (15%) GH women. The rate of low birth weight ($p=0.040$), small for gestational age / intra-uterine growth restriction ($p=0.018$), meconium-stained liquor ($p=0.018$) and Neonatal Intensive Care Unit admission ($p=0.001$) were significantly increased in GH group. Although the rate of poor progress, caesarean sections, instrumental deliveries, low APGAR score and neonatal jaundice were increased in GH, they were not statistically significant ($p>0.05$). Preterm delivery rate was not significantly different between GH and normotensive pregnancy. Even though lacking in statistical significance, a higher level of MMP-9 was found in the GH group. Similarly comparisons made

based on parity were noted to be lower, approaching statistical significance, especially in the nulliparous subgroup. This could also be a sign of the increased risk of PE and GH in nulliparous pregnancy. The different patterns of MMP-9 and TIMP-1 and -2 alterations between nulliparous and multiparous women could also suggest different underlying pathophysiological changes between the two subgroups. The analysis comparing pregnancy outcomes between the two groups showed increased risk for various adverse outcomes as well as maternal and perinatal complications among GH women.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Master Sains

**MATRIX METALLOPROTEINASE-9 DAN TISU PENGHALANG
METALLOPROTEINASE-1 DAN -2 SEBAGAI BIOMARKER YANG
BERPOTENSI SEMASA HIPERTENSI GESTASI**

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Penyakit hipertensi gestasi (GH) adalah suatu penyakit yang biasa di kalangan wanita mengandung, yang mana ianya boleh menyebabkan pre-eklampsia (PE) dan juga pelbagai komplikasi kepada ibu dan anak yang akan dilahirkan. Satu kluster enzim dipanggil Matrix Metalloproteinase (MMP) dan Tisu Penghalang Metalloproteinase (TIMP) dikatakan terlibat di dalam proses pato-fisiologi penyakit terbabit. Kajian ini adalah untuk mengkaji paras kandungan MMP-9, TIMP-1 and TIMP-2 di dalam serum, dikalangan pesakit GH untuk perbandingan dengan wanita mengandung yang sihat serta untuk mengaitkannya dengan hasil (*outcome*) kehamilan seperti tidak cukup bulan (*preterm*), tidak cukup perkembangan (*poor progress*), distress janin (*foetal distress*), kaedah kelahiran (*mode of delivery*), rendah berat lahir (*low birth weight*), kecil untuk umur gestasi (*small for gestational age*), rendah skor apgar (*low apgar score*), lumuran cecair mekonium (*meconium-stained liquor*), penyakit kuning neonatal (*neonatal jaundice*) dan kemasukan ke unit rawatan rapi neonatal

(*admission to NICU*). Sebanyak 118 pesakit dipilih secara rawak di dalam kajian kes-kawalan (tanpa perbandingan) ini. Kes adalah wanita GH (n=54); dan kawalan adalah wanita hamil yang sihat (n=64), dipilih melalui kedatangan ke klinik pesakit luar di 2 hospital (Hospital Besar Kuala Lumpur dan Hospital Serdang) semasa Mei 2008 hingga Januari 2009 dan memenuhi kriteria pemilihan. Semua perkembangan pesakit akan diikuti hingga proses kelahiran, dan semua maklumat yang dikehendaki didapati melalui rekod perubatan. Paras kandungan setiap penanda-bio (*biomarker*) disukat dengan menggunakan teknik *Enzyme-Link Immunosorbent Assay (ELISA)* yang spesifik, dan nisbah MMP/TIMP kemudiannya dihitung secara manual. MMP-9 didapati lebih tinggi sedikit di dalam kumpulan GH ($p=0.085$). Di dalam analisis sub-kumpulan atas dasar pariti, MMP-9 dan nisbah MMP-9/TIMP-1 didapati lebih tinggi sedikit di dalam kumpulan nuliparus GH ($p=0.064$ dan $p=0.079$ masing-masing), dan TIMP-1 lebih tinggi sedikit di dalam kumpulan multiparus GH ($p=0.062$). Positif korelasi yang lemah didapati di antara umur maternal dengan TIMP-1 di dalam kumpulan GH ($r=0.278$, $p=0.047$). Positif korelasi yang lemah juga didapati di antara umur gestasi dengan TIMP-2 di dalam kumpulan kawalan ($r=0.318$, $p=0.010$). Tiada korelasi didapati di antara MMP dan semua TIMP dengan paras tekanan darah. PE terjadi di dalam 8 (15%) pesakit GH. Kadar rendah berat kelahiran ($p=0.040$), kecil untuk umur gestasi / perkembangan intra-uterine terbatas (*intra-uterine growth restriction*) ($p=0.018$), lumuran cecair mekonium ($p=0.018$) dan kemasukan ke Unit Rawatan Rapi Neonatal (NICU) ($p=0.001$) meningkat secara signifikan di dalam kumpulan GH. Kadar tidak cukup perkembangan (*poor progress*), pembedahan kelahiran *caesarean (caesarean section)*, kelahiran dengan bantuan instrumen,

rendah nilai APGAR dan sakit kuning neonatal didapati meningkat di dalam GH tetapi tidak mencapai statistik signifikan ($p>0.05$). Kadar kelahiran awal penggal didapati tidak berbeza di antara kumpulan GH dan kawalan. Walaupun kekurangan nilai signifikan secara statistiknya, paras MMP-9 yang lebih tinggi didapati di kalangan kumpulan GH. Analisis mencatatkan nilai p yang lebih rendah, menghampiri nilai statistik signifikan, terutamanya di dalam sub-kumpulan nuliparus. Ini boleh menjadi petanda kepada peningkatan risiko menghadapi PE dan GH di kalangan kehamilan nuliparus. Corak perubahan MMP dan TIMP yang berbeza di antara wanita nuliparus dan multiparus juga boleh bermaksud kepada perubahan patofisiologi yang berbeza di antara kedua-dua sub-kumpulan. Analisis perbandingan mengenai hasil kehamilan di antara kedua-dua kumpulan menunjukkan peningkatan risiko serta beberapa komplikasi kepada ibu dan anak di kalangan wanita GH.

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I certify that a Thesis Examination Committee has met on 12 November 2010 to conduct the final examination of Jabrullah bin Ab. Hamid on his thesis entitled “Matrix Metalloproteinase-9 and Tissue Inhibitors of Metalloproteinases-1 and -2 as Potential Novel Biomarkers in Gestational Hypertension” in accordance with the Universities and Universiti College 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.

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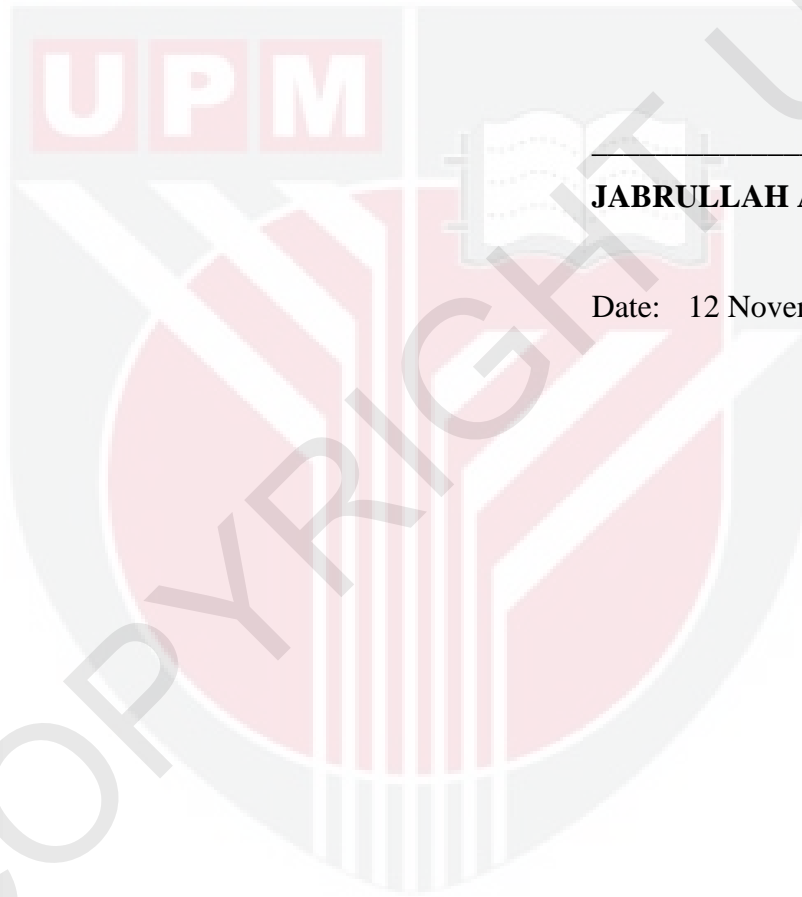
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DECLARATION

I declare that the thesis is my original work except for the 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 local institutions.



JABRULLAH AB. HAMID

Date: 12 November 2010

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