

**THE EFFECTS OF MIR-125B ACCUMULATED IN
BONE MATRIX ON BONE METASTASIS**

**FINAL SCIENTIFIC ASSIGNMENT
INTERNATIONAL DENTAL COURSE**



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APPROVAL SHEET

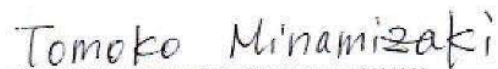
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**THE EFFECTS OF MIR-125B ACCUMULATED IN BONE MATRIX ON
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is my original work and all the data given in this thesis are real and authentic.

If in the future it is proven that I commit plagiarism, I will take full responsibility and accept the consequences.

Thus, this declaration has been made truthfully.

Surabaya, October 25th, 2018



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ABSTRACT

Background: MicroRNAs (miRNAs) are involved in RNA silencing and post-transcriptional regulation of gene expression. Growing evidence indicates that miRNAs play pivotal roles in the skeletal system. Previously we demonstrated that miRNAs accumulated in bone matrix as cargos of matrix vesicles (MVs) secreted from osteoblasts, in which miR-125b inhibited osteoclast formation by targeting the transcription repressor *Prdm1*. Transgenic (Tg) mice overexpressing miR-125b under the control of the human osteocalcin promoter exhibited high bone mass with a decreased number of osteoclasts. Cancer bone metastasis frequently causes osteolytic lesions with increased activity of osteoclasts. In silico studies showed miR-125b may target multiple genes involved in the survival and function of breast and prostate cancer cells that prone to metastasize to bone.

Hypothesis: Taken all together, it is suggested that that miR-125b accumulated in bone matrix may suppress metastatic cancer cell activities in bone.

Methods and Results: qRT-PCR analysis showed that levels of miR-125b in PC-3 (human prostate cancer) cells, MCF7 (human breast cancer) cells, and PY8119 (murine mammary carcinoma) cells were much less than those in mouse MC3T3-E1 osteoblasts and their MVs. MTT or EdU assays revealed a decrease in proliferation of PC-3, MCF7 and PY8119 cells, when treated with MVs or transfected with miR-125b mimic. In addition, MVs and/or miR-125b mimic decreased migration and invasion activities of these cell lines. To confirm the effects of miR-125b in vivo, PY8119 cells were injected intratibially into wild-type (WT) mice and Tg mice. Histological examination suggested that all mice exhibited osteolytic lesions with PY8119 cells for 2 weeks postdosing, while the reduction in osteoclast numbers and PY8119 cell foci were observed in Tg mice vs. WT mice. Micro-CT analysis showed that bone loss associated with PY8119 cell injection was diminished in Tg mice vs. WT mice.

Conclusion: These results suggest that miR-125b accumulated in bone matrix may have an ability to suppress bone metastasis via its dual action as an anti-osteoclastogenesis and tumor suppressor. We have now attempted to identify a direct target of miR-125b in cancer cells.

Keywords: miR-125b, Matrix vesicle, Osteolysis, Breast cancer, Bone metastasis

TABLE OF CONTENT

APPROVAL SHEET	iii
JUDGES	iv
DECLARATION OF ORIGINALITY	v
ACKNOWLEDGEMENT	vi
ABSTRACT	x
TABLE OF CONTENT	xii
CHAPTER 1 INTRODUCTION	1
1.1 Background.....	1
1.2 Research Question.....	2
1.3 Purpose	3
1.4 Research Benefit.....	3
CHAPTER 2 LITERATURE REVIEW	4
2.1 Bone.....	4
2.2 Bone Metastasis.....	6
2.2.1 Breast Cancer Bone Metastasis.....	7
2.3 Matrix Vesicles.....	7
2.4 MicroRNA.....	9
2.4.1 MicroRNA-125b	10
CHAPTER 3 CONCEPTUAL MAPPING AND HYPOTHESIS.....	12
3.1 Conceptual Mapping	12
3.2 Hypothesis	13
CHAPTER 4 RESEARCH METHODOLOGY	13
4.1 Research Type	14
4.2 Research Workflow	14
4.3 Materials and Instruments	15
4.3.1 Materials.....	15
4.3.2 Instruments.....	16
4.4 Methods	17
4.4.1 Animals	17
4.4.2 Cell Culture	18

4.4.3 Reverse Transcription and Genotyping Check	18
4.4.4 Proliferation, Migration and Invasion Assay	17
4.4.5 Intratibial Injection.....	18
4.4.6 Micro-computer Tomography Analysis	19
4.4.7 Histomorphological Analysis.....	19
4.5 Data Analysis.....	19
CHAPTER 5 RESULTS	20
5.1 MiR-125b enriches in MVs and is deregulated in cancer cells.	20
5.2 MVs suppresses migration, invasion and proliferation of cancer cells <i>in vitro</i>	20
5.3 MiR-125b mimic represses migration of cancer cells.	23
5.4 Tg mice overexpressing miR-125b demonstrate high trabecular bone mass.....	24
5.5 MiR-125b diminish bone loses caused by cancer cells progression.	24
CHAPTER 6 DISCUSSION.....	26
CHAPTER 7 FIGURES AND FIGURE LEGENDS	30
REFERENCES	38
ATTACHMENT	41