

# **Platelets and Neutrophils Cross Talk Mediating Cancer Growth and Metastasis in Urinary Bladder Carcinoma**

*Thesis submitted for partial fulfillment of Master Degree in Clinical and Chemical Pathology*

**By**

**Maha Mahmoud Samy Othman**

*(M.B; B.Ch)*

**Supervisors**

***Prof. Dr. Eman El Mahgoub***

*Professor of Clinical and Chemical Pathology,  
Faculty of Medicine, Cairo University*

***Prof. Dr. Bothina Madkour***

*Professor Researcher of Haematology  
Theodor Bilhrz Research Institute*

*Faculty of Medicine  
Cairo University  
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بسم الله الرحمن الرحيم

## **Dedication**

I would like to dedicate this work to my family specially my ***Father*** and ***Mother*** who supported me through my entire life. I cannot express my gratitude to all they did and still doing to me. Thank you my guarding angels.

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# **List OF CONTENTS**

|  | <b>PAGE</b> |
|--|-------------|
| <b><u>INTRODUCTION</u></b>   |             |
| Scientific Background  | 1           |
| Aim of the work  | 3           |
| <b><u>REVIEW OF LITRETURE:</u></b>   |             |
| <b><u>CHAPTER 1</u></b>  |             |
| <b><i>Polymorphonuclear Leukocytes</i></b>                                 |             |
| Neutrophil Granulocyte   | 5           |
| Neutrophil Development   | 6           |
| Neutrophil Structure   | 8           |
| Neutrophil Functions   | 14          |
| Neutrophil disorders   | 18          |
| <b><u>CHAPTER 2</u></b>  |             |
| <b><i>Platelets</i></b>  |             |
| Platelet Ontogeny  | 20          |
| Platelet Structure   | 22          |
| Platelet Receptors   | 22          |
| Platelet Functions   | 24          |
| Platelet Disorders   | 29          |
| <b><u>CHAPTER 3</u></b>  |             |
| <b><i>P-Selectin &amp; its Ligand P-selectin glycoprotein ligand-1</i></b> |             |
| Structure and cell expression of P-selectin                                | 34          |
| Structure and cell expression of P-selectin glycoprotein ligand-1          | 38          |
| The role of P-selectin in host defense and inflammation                    | 42          |
| The role of P-selectin and PSL-1 in haemostasis and thrombosis             | 44          |
| The role of P-selectin in cancer growth and metastasis                     | 46          |
| <b><u>CHAPTER 4</u></b>  |             |
| <b><i>Bladder Cancer</i></b>   |             |
| Incidence and Epidemiology of bladder cancer                               | 52          |
| Risk Factors of cancer bladder   | 53          |
| Diagnosis of cancer bladder  | 55          |
| Genetic Background   | 57          |
| Pathological classification of cancer bladder                              | 62          |
| Broad lines of treatment of cancer bladder                                 | 72          |
| <b><u>PATIENTS &amp; METHODOLOGY</u></b>                                   | 75          |

|                                      |     |
|--------------------------------------|-----|
| <b><u>RESULTS</u></b>                | 89  |
| <b><u>DISCUSSION</u></b>             | 123 |
| <b><u>SUMMARY AND CONCLUSION</u></b> | 136 |
| <b><u>RECOMMENDATIONS</u></b>        | 140 |
| <b><u>REFERENCES</u></b>             | 141 |
| <b><u>APPENDIX</u></b>               | 165 |

## **List of Tables**

|   | <b>PAGE</b> |
|---|-------------|
| <b>Table (1):</b> Current TMN Staging for Primary Bladder Cancer.   | 71          |
| <b>Table (2):</b> Reagents provided in sCD62 ELIZA kit.   | 84          |
| <b>Table (3):</b> Summary of assay procedure sCD62 ELIZA kit.   | 86          |
| <b>Table (4):</b> Demographic features of the studied groups and control group.   | 89          |
| <b>Table (5):</b> Platelet count ( $\times 10^3/\text{ul}$ ) in different studied groups.                                       | 92          |
| <b>Table (6):</b> Absolute neutrophil count ( $\times 10^3/\text{ul}$ ) in different studied groups.                            | 95          |
| <b>Table (7):</b> Platelet expression of CD62P % by flowcytometry in different studied groups.                                  | 98          |
| <b>Table (8):</b> Neutrophil expression of CD162 (%) by flowcytometry in different studied groups.                              | 101         |
| <b>Table (9):</b> Soluble P-selectin (sP-Selectin) (ng/ml) by ELISA in different studied groups.                                | 108         |
| <b>Table (10):</b> Correlation analysis of different studied parameters in control group.                                       | 111         |
| <b>Table (11):</b> Correlation analysis of different studied parameters in (Locally invasive tumor) group.                      | 114         |
| <b>Table (12):</b> Correlation analysis of different studied parameters in (Invasive tumor with regional LN involvement) group. | 116         |
| <b>Table (13):</b> Correlation analysis of different studied parameters in (Invasive tumor with distant metastasis) group.      | 119         |
| <b>Table (14):</b> Stepwise multiple linear regression analysis.  | 122         |
| <b>Table (15):</b> Haemogram of the Control group.  | 165         |



|   |     |
|---|-----|
| <b>Table (16):</b> Haemogram of the (Locally invasive tumor) group.                                       | 166 |
| <b>Table (17):</b> Haemogram of the (Invasive tumor with regional LN involvement) group.                  | 167 |
| <b>Table (18):</b> Haemogram of the (Invasive tumor with distant metastasis) group.                       | 168 |
| <b>Table (19):</b> liver and kidney functions of the Control group.                                       | 169 |
| <b>Table (20):</b> liver and kidney functions of the (Locally invasive tumor) group.                      | 169 |
| <b>Table (21):</b> liver and kidney functions of the (Invasive tumor with regional LN involvement) group. | 170 |
| <b>Table (22):</b> liver and kidney functions of the (Invasive tumor with distant metastasis) group.      | 170 |
| <b>Table (23):</b> TNM staging, histopathology and grading of tumor in different studied groups.          | 171 |

## **List of Figures**

|   | <b>PAGE</b> |
|---|-------------|
| <b>Fig (1):</b> A diagram of different kinds of blood cells and their relationship to each other.                 | 4           |
| <b>Fig (2):</b> A neutrophil with a segmented nucleus.  | 6           |
| <b>Fig (3):</b> Different stages of neutrophil granulocyte development.   | 8           |
| <b>Fig (4):</b> Neutrophil and Its Surface Molecules.   | 12          |
| <b>Fig (5):</b> A diagram of different steps of neutrophil function.  | 17          |
| <b>Fig (6):</b> Normal thrombopoiesis.  | 21          |
| <b>Fig (7):</b> Major platelet receptor-ligand interactions.  | 23          |
| <b>Fig (8):</b> A diagram illustrating steps of platelet activation.  | 26          |
| <b>Fig (9):</b> Platelet-independent and platelet-dependent recruitment of PMN.                                   | 28          |
| <b>Fig (10):</b> The domain structure of the selectin family of cell adhesion molecules.                          | 36          |
| <b>Fig (11):</b> The signaling molecules and their pathways involved in P-selectin exocytosis in platelets.       | 37          |
| <b>Fig (12):</b> The domain structure of PSGL-1.  | 40          |
| <b>Fig (13):</b> The interaction of P-selectin expressed on stimulated endothelial cells and activated platelets. | 43          |
| <b>Fig (14):</b> Platelet-endothelium adhesion.   | 45          |
| <b>Fig (15):</b> Interplay of inflammation and hemostasis.  | 46          |
| <b>Fig (16):</b> Receptors involved in the rolling of leukocytes and tumor cells.                                 | 48          |

|   |     |
|---|-----|
| <b>Fig (17):</b> Mechanisms of haemostatic system activation by tumor cells.  | 50  |
| <b>Fig (18):</b> Important genetic and epigenetic defects that characterize the divergent pathways of urothelial tumorigenesis.         | 61  |
| <b>Fig (19):</b> Principal genetic pathways in the development of bladder urothelial tumors.  | 63  |
| <b>Fig (20):</b> Extent of primary bladder cancer.  | 70  |
| <b>Fig (21):</b> Standard curve of P-selectin.  | 87  |
| <b>Fig (22):</b> Pie chart showing percentage of different histopathological pictures of bladder biopsies.                              | 90  |
| <b>Fig (23):</b> Mean values of Platelet count in different studied groups.   | 93  |
| <b>Fig (24):</b> Mean values of absolute neutrophil count in different studied groups.  | 96  |
| <b>Fig (25):</b> Mean values of P-selectin (CD62P) (%) in different studied groups.   | 99  |
| <b>Fig (26):</b> Mean values of Neutrophil expression of P-selectin glycoprotein ligand-1(PGL-1; CD162) (%) indifferent studied groups. | 102 |
| <b>Fig (27):</b> Flow chart showing platelet expression of CD62P % from a subject in the control group.                                 | 103 |
| <b>Fig (28):</b> Flow chart showing neutrophil expression of CD162 % from the same subject in the control group.                        | 103 |
| <b>Fig (29):</b> Flow chart showing platelet expression of CD62P % from a subject in the (Locally invasive tumor) group.                | 104 |
| <b>Fig (30):</b> Flow chart showing neutrophil expression of CD162 % from the same subject in the (Locally invasive tumor) group.       | 104 |

|  |     |
|--|-----|
| <b>Fig (31):</b> Flow chart showing platelet expression of CD62P % from a subject in the (Invasive tumor with regional LN involvement) group.          | 105 |
| <b>Fig (32):</b> Flow chart showing neutrophil expression of CD162 % from the same subject in the (Invasive tumor with regional LN involvement) group. | 105 |
| <b>Fig (33):</b> Flow chart showing platelet expression of CD62P % from a subject in the (Invasive tumor with distant metastasis) group.               | 106 |
| <b>Fig (34):</b> Flow chart showing neutrophil expression of CD162 % from the same subject in the (Invasive tumor with distant metastasis) group.      | 106 |
| <b>Fig (35):</b> Mean values of soluble P-selectin (sP-Selectin) (ng/ml) in different studied groups.  | 109 |
| <b>Fig (36):</b> Correlation between sP-selectin and P-selectin in control group.  | 112 |
| <b>Fig (37):</b> Correlation between CD162 and absolute neutrophil count (ANC) in control group.   | 113 |
| <b>Fig (38):</b> Correlation between CD162 and TNM staging in (Locally invasive tumor) group.  | 115 |
| <b>Fig (39):</b> Correlation between P-selectin and platelet count in (Invasive tumor with regional LN involvement) group.                             | 117 |
| <b>Fig (40):</b> Correlation between sP-selectin and platelet count in (Invasive tumor with regional LN involvement) group.                            | 118 |
| <b>Fig (41):</b> Correlation between P-selectin and grading in (Invasive tumor with distant metastasis) group.   | 120 |
| <b>Fig (42):</b> Correlation between mean values of P-selectin and mean values of soluble P-selectin in all studied groups.                            | 121 |

## **List of Abbreviations**

| <b>Abbreviation</b> | <b>The full term</b>                              |
|---------------------|---|
| ADP                 | Adenosine Diphosphate                             |
| ALT                 | Alanin Aminotransferase                           |
| ANC                 | Absolute neutrophil count                         |
| AST                 | Aspartate Aminotransferase                        |
| BCG                 | Bacillus Calmette-Guerin.                         |
| BPI                 | Bacterial permeability-increasing protein.        |
| C5a                 | Complement 5a                                     |
| cAMP                | Cyclic adenosine monophosphate                    |
| CD                  | Cluster of Differentiation                        |
| CD24                | A mucin-like adhesion molecule on cancer cells.   |
| CD62P               | P-selectin  |
| CD162               | P-selectin glycoprotein ligand-1                  |
| CIS                 | Carcinoma in situ                                 |
| COX-2               | Cyclooxygenase-2.                                 |
| CRC                 | Colorectal carcinoma cells.                       |
| Creat.              | Creatinine  |
| CT                  | Computerized tomography                           |
| DIC                 | Disseminated Intravascular Coagulation            |
| DNA                 | Deoxyribonucleic Acids                            |
| EGF                 | Epidermal growth factor                           |
| ELISA               | Enzyme-Linked Immunosorbent Assay                 |
| FISH                | Fluorescent insito hybridization                  |
| FMLP                | N-formyl-1-methionyl-1-leucyl-1-phenylalanine     |
| FucT-VII            | Fucosyltrasferase enzyme                          |
| GI                  | Low grade malignancy                              |
| GII                 | Moderate grade malignancy                         |
| GIII                | High grade malignancy                             |
| G-CSF               | Granulocyte colony-stimulating factor             |
| GM-CSF              | Granulocyte/ macrophage colony-stimulating factor |
| GP                  | Glycoprotein.                                     |
| GPCR                | G-protein coupled seven transmembrane receptors.  |
| GRO                 | Growth-related oncogene                           |
| H&E                 | Hematoxylin and eosin                             |
| Hb                  | Haemoglobin concentration                         |
| Histopath.          | Histopathology                                    |
| HIT                 | Heparin-induced thrombocytopenia                  |
| HIV                 | Human Immunodeficiency Virus                      |
| HPA                 | Human platelet alloantigen.                       |
| HUS                 | Hemolytic uremic syndrome.                        |
| IL                  | Interleukin                                       |

|                   |  |
|-------------------|--|
| INR               | International Normalized Ratio                       |
| ITP               | Immune thrombocytopenic purpura.                     |
| IVP               | Intravenous pyelography                              |
| kDa               | Killo-Dalton   |
| LOH               | Loss of heterozygosity                               |
| LPS               | Lipopolysaccharide                                   |
| LRR               | Leucine-rich repeated                                |
| MPO               | Myeloperoxidase enzyme.                              |
| MRI               | Magnetic resonance imaging                           |
| mRNA              | Messenger RNA  |
| MS                | Mass spectrometry                                    |
| NCI               | National Cancer Institute                            |
| NADPH             | Nicotinamide adenine dinucleotide phosphate hydrogen |
| NETs              | Neutrophil extracellular traps.                      |
| NF- $\kappa$ B    | Nuclear Factor $\kappa$ B                            |
| NH <sub>2</sub> - | Amino terminal                                       |
| NSAIDs            | Non-steroidal anti-inflammatory drugs                |
| NSF               | Nephrogenic systemic fibrosis                        |
| PAF               | Platelet-activating-factor                           |
| PC                | Prothrombin concentration                            |
| PDGF              | Platelet derived growth factor                       |
| PECAM-1           | Platelet–endothelial cell adhesion molecule-1.       |
| PL                | Phospholipase  |
| PML               | Polymorphonuclear leukocytes                         |
| PMN               | Polymorphonuclear leukocytes                         |
| PSGL-1            | P-selectin glycoprotein ligand-1                     |
| PT                | Prothrombin Time                                     |
| RBCs              | Red Blood Cell Count                                 |
| RNA               | Ribonucleic Acid                                     |
| SABC              | Schistosoma-associated bladder cancer                |
| SCC               | Squamous cell carcinoma .                            |
| sP-selectin       | Soluble P-selectin.                                  |
| T bili            | Total Bilirubin                                      |
| TF                | Tissue factor  |
| TNF- $\alpha$     | Tumor necrosis factor- $\alpha$ .                    |
| TLC               | Total Leukocytic Count                               |
| TNM               | Tumor-node-metastasis staging system.                |
| TP                | Total Protein  |
| TTP               | Thrombotic thrombocytopenic purpura                  |
| TxA <sub>2</sub>  | Thromboxane A <sub>2</sub>                           |
| VEGF              | Vascular endothelial growth factor.                  |
| vWF               | Von Willebrand Factor.                               |
| 2-DE              | 2-dimensional gel electrophoresis                    |
| 3-D               | Three dimensional                                    |

## ABSTRACT

Urinary bladder cancer is a major public health problem being one of the most common malignancies worldwide. In Egypt, this heterogeneous disease has the highest reported world wide incidence "37 per 100,000 populations" in the world due to endemic schistosomiasis. It has been believed that many cancers arise from sites of infection, chronic irritation and inflammation. It has been also suggested that the interaction between circulating platelets and neutrophils influences innate immune functions, contributing to regulate inflammation that means cell-cell interactions are crucial for the host defense mechanism. **P-selectin** is platelet activation marker that has been shown to mediate the rolling of blood cells on the surface of the endothelium and initiate the attachment of leukocytes circulating in the blood to platelets, endothelial cells, and other leukocytes. **PSGL-1** is a disulfide-bonded homodimeric mucin-like glycoprotein on leukocytes that interacts with **P-**, **L-**, and **E-selectin**. In order to investigate the role of **P-selectin** and **PSGL-1** mediated interactions in the microenvironment of the tumor; we measured **P-selectin** on platelets, **PSGL-1** on neutrophils and **sP-selectin** in plasma of 30 patients with **bladder cancer**. Patients were classified into three groups; first group with locally invasive tumor "n=10", second group with regional lymph node involvement "n=10", and third group with distant metastasis "n=10". **Platelet count** was significantly higher in patients with "invasive tumor with distant metastasis" group compared to control and the other two groups; that is due to the frequently observed thrombocytosis in cancer patients. **P-selectin** was significantly elevated in all patient groups as a part of inflammatory and immunological reactions. However among the 3 studied groups, the patients with invasive tumor and distant metastasis showed the highest elevation may be due to