P38 mitogen-activated protein kinase (p38 MAPK) overexpression in clinical staging of nasopharyngeal carcinoma

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Abstract. Molecular biological research on nasopharyngeal carcinoma has been widely practiced, such as VEGF, EGFR, COX-2 expression and so on. MAPK plays a role in cell growth such as proliferation, differentiation, and apoptosis, primarily contributing to gene expression, where p38 MAPK pathway mostly associate with anti-apoptosis and cause cell transformation. The aim of this study is to determine the expression of p38 MAPK in clinical stage of nasopharyngeal carcinoma so that the result can be helpful in prognosis and adjunctive therapy in nasopharyngeal carcinoma. The research design is descriptive. It was done in THT-KL Department of FK USU/RSUP Haji Adam Malik, Medan and Pathology Anatomical Department of FK USU. The study was conducted from December 2011 to May 2012. The Samples are all patients who diagnosed with nasopharyngeal carcinoma in oncology division of Otorhinolaryngology Department. p38 MAPK overexpression was found in 21 samples (70%) from 30 nasopharyngeal carcinoma samples. The elevated of p38 MAPK expression most found on T4 by eight samples (38.1%), N3 lymph node group by nine samples (42.9%), stage IV of clinical staging is as many as 15 samples (71.4%). p38 MAPK most expressed in stage IV clinical staging of patients with nasopharyngeal carcinoma.

1. Introduction
Nasopharyngeal carcinoma (NPC) is a carcinoma derived from the nasopharyngeal epithelium.[¹] Globally, nasopharynx carcinoma is rare cancer with an annual rate of approximately 80,000 cases of which is 0.7% of all cancers. In North America and Europe, the incidence is less than 1 case per 100,000 population, but in endemic areas such as South China (Hong Kong) and Southeast Asia, incidence approximately 20 - 30 cases happen in 100,000 male population and 8 to 15 cases per 100,000 population in female.[²]

Nasopharyngeal carcinoma in Indonesia is the fifth of ten malignant tumours majority throughout the body, whereas, in the ear diseases, nose and throat, nasopharyngeal cancer is on first ranks. Nearly 60% of head and neck tumours are nasopharyngeal cancer.[³] The incidence rate in Indonesia is 6.2 per 100,000 cases or about 12,000 cases annually.[³] In ENT Department of Adam Malik General Hospital Medan, there were 335 new cases of KNF during 2006-2010.[⁵]
Molecular biology studies in nasopharyngeal carcinoma has been widely performed that can help in determining the prognosis in patients with nasopharyngeal carcinoma and assist in providing adjunctive therapy in nasopharyngeal carcinoma. MAPK (Mitogen-Activated Protein Kinase) is an enzyme that plays a role in extracellular stimuli such as growth hormone transfer to the nucleus, and also serves to regulate the expression of almost all genes in the human body. MAPK has three paths in the transduction process of ERK, JNK and p38, where the p38MAPK path is most associated with anti-apoptotic function and the p38MAPK pathway causes cell transformation.\[6\]

Several studies have found the involvement of the p38 MAPK pathway to increase the reaction of cell apoptosis in oral cavity cancer after the administration of tolfenamic acid.\[7\] Nasopharyngeal carcinoma caused by latent infection of Epstein Bar virus, causing changes in nasopharyngeal epithelial cells. In a study conducted by the Polymerase Chain Reaction (PCR) method, p38 MAPK suppression found in nasopharyngeal epithelium that led to cancer cells growth.\[8\]

2. Methods
This is descriptive study, where all the data was taken from the patient examination results in Otorhinolaryngology Department from December 2011 to May 2012. Immunohistochemical examination performed in Anatomic Pathology Department. The population criteria of this study are patients who diagnosed with NPC and never received treatment with radiotherapy, chemotherapy or both chemoradiotherapy. Sample selection of this research is by nonprobability sampling. The variables in this study were the expression of p38MAPK among primary tumour size (T), size of enlarged lymph node (N), and clinical staging.

3. Result
An immunoreactive score of 30 nasopharyngeal carcinoma patient’s tissue was found to be over perspective of p38MAPK in 21 samples (70%). The elevated of p38MAPK expression most prevalent foundon T4 by eight samples (38.1%). N3 lymph node group of nine samples (42.9%). Overexpression of p38 MAPK most common found in stage IV is as many as 15 samples (71.4%).

4. Discussion
In this study, there was overexpression of p38 MAPK in 21 tissues of nasopharyngeal carcinoma (70%) of 30 tissues of nasopharyngeal carcinoma. This result seemed to be similar to some studies. Liang\[9\] found overexpression of p38 MAPK in gastric carcinoma, of which 30 samples of gastric carcinoma found to be overexpression of p38 MAPK of 14 samples and higher levels of protein in gastric carcinoma tissue compared to normal mucosa. Research by Fang\[10\] et al. also supported the results of our study, which significant results on MAPK pathway involvement in apoptotic processes, where 19 of MAPK genes pathway found with PCR in patients with nasopharyngeal carcinoma.

**Figure 1.** p38 MAPK expression in nasopharyngeal carcinoma.
NPC is a squamous cell carcinoma developed in nasopharyngeal epithelium, where one of the alleged etiologies is an inflammatory and infection process in the upper respiratory tract caused by Epstein-Barr Virus (EBV). EBV infection as one of the risk factors NPC has a latency period to maintain EBV episodes in the infected nasopharyngeal epithelium, about 20-25 years without symptoms. This leads to EBV infection providing a collection of target cells in the nasopharynx susceptible to exposure to environmental carcinogens as well as subsequent genetic changes in oncogenes and tumour suppressor genes that play a role in malignant transformation of nasopharyngeal carcinoma.\(^{[1]}\)

![Figure 2. Expression of p38 MAPK: (A) primary tumour, (B) lymph node, and (C) clinical staging](image)

This study found the most p38 MAPK overexpression on T4 primary tumour size by eight samples (38.1%). The distribution of p38 MAPK overexpression is increasing in line with the enlargement of tumour size T2 to T4. Sullivan\(^{[12]}\) in his clinical trial study in carcinoma colon found the role of p38 MAPK in the extent of tumour expansion. He provided a p38 MAPK inhibitor, which led to increased apoptosis and reduced proliferation of tumour cells. Liang\(^{[9]}\) et al. in a case-control study of normal and gastric carcinoma tissue, found no significant association between p38 MAPK and age of gastric carcinoma tumour.

The phenotype of carcinoma can be recognized by the presence of apoptotic failure, unlimited replication, invasion and metastasis, angiogenesis, development of drug resistance.\(^{[6]}\) At NPC the emerging regional metastases are metastases to the lymph nodes of the neck. In this study, p38MAPK overexpression was found most frequently in N3 lymph node size (9.9%). Overexpression of p38 MAPK increased in line with lymph node. These results are consistent with a case-control study by Minghua\(^{[13]}\) presumed the role of MAPK pathways in NPC metastases to regional lymph nodes through activation of the enzyme protein. Similar results were also found in a clinical trial study by Sullivan\(^{[12]}\) who suspected MAPK pathway involvement in lymph node metastasis in colon carcinoma. Huang\(^{[14]}\) found an increase in the activity of endogenous MMP p38 associated with invasive and metastatic properties to regional lymph nodes in breast carcinoma.

This study found a predominant of p38 MAPK overexpression most common in stage 4 by 15 samples (71.4%). Increased of p38 MAPK expression in this study was similar with clinical staging. Wan\(^{[15]}\) concluded MAPK involvement in tumorigenesis process in NPC resulting in high expression of the Aur-A protein, which is a protein that causes intracranial invasion and an increased stage of nasopharyngeal carcinoma. It is an important role of p38 MAPK in pathology processes such as inflammation, cell stress, apoptosis, cell cycle and growth, so its activation will increase the phenotype of malignancy in clinical staging.\(^{[16]}\)\(^{[17]}\)

5. Conclusions
Despite p38 MAPK most expressed in stage IV of clinical staging of nasopharyngeal carcinoma, further study of p38 MAPK expression with nasopharyngeal carcinoma before and after therapy with
reference to the results of this study and the results of several studies on expression relationship of p38 MAPK with other carcinoma is needed.

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