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Effect of Osimertinib on the expression of serum mmp-7 and mmp-9 in patients with non-small cell lung cancer

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Abstract: The purpose was to investigate the efficacy and safety of Osimertinib in the treatment of advanced non-small cell lung cancer and to analyze its effects on the expression of serum matrix metalloproteinase-7 (MMP-7) and matrix metallo-proteinase-9 (MMP-9). Eighty patients were equally divided into observation and control group. The observation group was given Osimertinib combined with conventional chemotherapy and the other was treated with conventional chemotherapy alone. The short-term efficacy, the levels of serum MMP-7, MMP-9 and adverse reactions were compared. The effectiveness and clinical benefit rate of the observation group were 62.50% and 92.50% respectively, significantly higher than the control group. There was no significant difference in MMP-7 and MMP-9 before treatment however there was a significant difference after treatment, and the serum MMP-7 & MMP-9 levels showed a trend of increasing with decreasing efficacy. After treatment, comparing with control group, serum MMP-7 and MMP-9 levels were significantly lower, the Karnofsky score was significantly higher, and the improvement effect of the quality of life was statistically significant. Besides, the incidence of leukopenia, thrombocytopenia, anemia and gastrointestinal symptoms were significantly lower. In the treatment of patients with advanced non-small cell lung cancer, Osimertinib significantly reduced the expression of serum MMP-7, MMP-9, improved the clinical benefit and quality of life of patients. The clinical efficacy was significant with a high safety.

Keywords: Non-small cell lung cancer, Osimertinib, MMP-7, MMP-9

INTRODUCTION

Lung cancer is one of the most malignant tumors with the highest incidence and mortality worldwide, among which, non-small cell lung cancer (NSCLC) accounts for about 80% (Felip and Rosell, 2008). Lung cancer is difficult to be detected in the early stages, because it is commonly unconventional at diagnosis stage and the disease improves speedily. Seventy percent of NSCLC patients had meta-static lesion and/or critical surgical management at the period of diagnosis (Scagliotti *et al*., 2009; Zhang *et al*., 2010). The survival degree of patients having lung cancer was only around seven percent for ten years. By means of the rise in age, the survival rate of patients will drop consequently (Yoshizawa *et al*., 2010). On behalf of patients with advanced NSCLC, management is largely intended to prolong survival and improve superiority of life. Chemotherapy based on Platinum is presently the first-line treatment in lung cancer patients, that can efficiently postpone the development of the disease and extend the survival spell of patients, on the other hand, still a large ill population with reduced response to chemotherapy, the disease still remains at progress state, and chemotherapy toxicity and side effects play a vital role to put impact over quality of patient's life (Wang *et al*., 2008; Lu *et al*., 2016). Osimertinib (axitinib) is considered as an oral, irreversible and selective inhibitor which aimed to target EGFR activation and resistance mutations. A small molecule of aniline-pyrimidine along with acrylamide group produce a covalent bond with C797 at the ATP binding site edge in catalytic domain of the EGFR gene. It binds irreversibly with a definite mutant form of EGFR, thereby inhibiting the proliferation of cancer cells and producing apoptosis in tumor cells as soon as possible (Bo *et al*., 2016). Few clinical studies are being carried out on Osimertinib to treat advanced non-small cell lung cancer in China. Matrix metalloproteinases (MMPs) have got a excessive homology in their structure. Amongst the enzymes, they are regarded as representative groups and may play title role in case of tumor metastasis, invasion, wound healing, menstrual cycle, and other physiological and pathological procedures (Banerjee *et al*., 2013; Attia *et al*., 2013). On the basis of the above background, this study used Osimertinib combined with chemotherapy in the treatment of advanced non-small cell lung cancer in a controlled study to evaluate its short-term efficacy and safety, and to explore its impact on the expression of serum MMP-7 and MMP-9 in Chinese patients and its anti-cancer mechanism.

MATERIALS AND METHODS

Research subjects

Eighty patients (n=80) with advanced non-small cell lung cancer who were treated in "The First Affiliated Hospital of GuangXi Medical University, China" from April to

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December 2017 were selected as research subjects.

Inclusion criteria

The non-small cell lung cancer was confirmed by imaging and pathological examination. The Karnofsky score was ≥60 points. No radiochemotherapy and other anti-tumor treatments were performed 30 days before the selection. The expected survival time was more than 3 months.

Exclusion criteria

Patients with other serious diseases, syndromes or complications

Progressive lung cancer patients

Patients with severe heart, liver and kidney damage

Poor patient compliance, unable to cooperate with therapist

Psychotic patients and critically ill patients

Patients with bone metastases and brain metastases, and the symptoms were not controlled.

Patients whose clinical data could not determine the efficacy and safety

The patients were randomly divided into observation group and control group, each with 40 cases.

The observation group was 36 to 71 years old with an average of (55.62±5.21) years consisting of 22 male and 18 female. The pathological classification was as: 30 cases of squamous cell carcinoma, 8 cases of adenocarcinoma, 2 cases of squamous adenocarcinoma; TNM stage: 14 cases in stage IIIa, 17 cases in stage IIIb, 9 cases in stage IV; Karnofsky score 63 to 87 points, mean (74.67±4.25) points.

The control group patients were 35 to 72 years old with an average of (55.84±6.02) years having 23 male and 17 female. The pathological classification was as: 31 cases of squamous cell carcinoma, 7 cases of adenocarcinoma, 2 cases of squamous adenocarcinoma; TNM stage: 15 cases in stage IIIa, 16 cases in stage IIIb, 9 cases in stage IV; Karnofsky score 63 to 89 points, mean (75.27±3.61) points. There were no significant differences between the two groups in terms of age, gender, pathological type, TNM stage, Karnofsky score (P>0.05), and they were comparable as shown in table 1.

Methods

The control group used conventional platinum-based chemotherapy regimens including GP (gemcitabine + cisplatin), DP (multiple paclitaxel + cisplatin), TP (paclitaxel + cisplatin), and NP (Vinorelbine + Cisplatin), 3 weeks for 1 course of treatment, at least 2 consecutive courses. The observation group was given oral Osimertinib (Axitinib, manufactured by Astra Zeneca) at the same time on the basis of the control group, 80 mg/d, 3 weeks for 1 course of treatment, and 2 consecutive courses of treatment. The adverse reactions in the two groups of chemotherapy were closely observed and treated.

Laboratory detection

Before treatment and after two courses of treatment, the two groups of patients were collected with 5mL of fasting peripheral blood, and centrifuged at 2500r/min for 15 minutes. The upper serum was detected and stored at -20°C. Serum levels of MMP-7 and MMP-9 were determined by enzyme-linked immunosorbent assay. Murine anti-human MMP-7 monoclonal antibody, mouse anti-human MMP-9 monoclonal antibody and kit were purchased from Calbiochem, Germany.

Evaluation standard

Quality of life assessment

Evaluation of patient's quality of life was carried out by Karnofsky score before treatment and after 2 courses of treatment;

Significantly effective

The score was increased by >20 points after treatment;

Effective

The score was improved by 10 to 20 points after treatment

Stable

The score was improved by <10 points after treatment;

Invalid

The score decreased after treatment.

Evaluation of efficacy and adverse reactions

Thoracic CT or MRI was reviewed after 2 courses of treatment. The short-term efficacy was evaluated according to the evaluation criteria of WHO solid tumor efficacy, including complete remission (CR), partial remission (PR), stable lesion (SD), and lesion progression (PD), CR+PR indicated effective, CR+PR+SD indicated clinical benefit.

Ethical approval

The experimental scheme has been approved by the ethics committee of the First Affiliated Hospital of Guangxi Medical University.

STATISTICAL ANALYSIS

The data was analyzed using SPSS 21.0 on PC. Measured data was expressed as mean ± SD ($\overline{x} \pm s$). Samples of the two groups were compared using t-test, and comparisons between groups were performed using analysis of variance. Count data was expressed as a rate, using the χ^2 test. P<0.05 was considered statistically significant.

RESULTS

The judgment of curative effect is based on the Thoracic CT or MRI. The specific judgment method was described in the “Evaluation of efficacy and adverse reactions” part.

Table 1: Comparison of general information between the two groups

Item		Observation Group (N=40)	Control Group (N=40)	T/X^2	P
Age (Years)		55.62±5.2	55.84±6.0	0.694	0.072
Gender (Case)	Male	22	23	1.005	0.063
	Female	18	17		
Karnofsky Score (Point)		74.67±4.2	75.27±3.6	1.017	0.059
Pathological Classification		---	---	---	---
Squamous Cell Carcinoma		30	31	---	---
Adenocarcinoma		8	7	0.861	0.124
Squamous Adenocarcinoma		2	2	---	---
TNM Stage		---	---	---	---
IIIa		14	15	---	---
IIIb		17	16	0.773	0.296
IV		9	9	---	---

TNM system: Tumor, Node and Metastasis system based on the size, number and metastasized of the cell
IIIa: Tumor size category 2 without ciliary body involvement but with extraocular extension ≤5 mm in largest diameter
IIIb: Tumor size category 3 with ciliary body involvement and extraocular extension ≤5 mm in largest diameter
IV: The cancer has spread to other organs or parts of the body

Table 2: Comparison of short-term efficacy between the two groups (case)

Group	Case	CR	PR	SD	PD	Effective [N (%)]	Clinical Benefit [N (%)]
Observation Group	40	2	23	12	3	25 (62.50)	37 (92.50)
Control Group	40	1	19	12	8	20 (50.00)	32 (80.00)
X^2	---					6.742	7.035
P	---					0.026	0.021

Table 3: Relationship between serum levels of MMP-7 and MMP-9 and efficacy in patients (pg/mL, $\bar{x} \pm s$)

Item	Case	MMP-7		MMP-9	
		Before Treatment	After Treatment	Before Treatment	After Treatment
CR+PR	45	184.32±34.57	99.45±26.73	572.25±42.34	231.83±36.35
SD	24	191.35±41.08	161.18±24.25	576.48±37.56	416.05±33.73
PD	11	198.26±45.31	216.94±32.85	579.51±43.67	614.65±39.22
F	---	0.748	5.061	0.962	6.005
P	---	0.263	0.031	0.104	0.029

Table 4: Comparison of serum levels of MMP-7 and MMP-9 in the two groups (pg/mL, $\bar{x} \pm s$)

Group	Case	MMP-7		MMP-9	
		Before Treatment	After Treatment	Before Treatment	After Treatment
Observation Group	40	196.24±41.7	114.62±39.5	571.41±46.33	271.93±44.31
Control Group	40	195.61±46.2	148.55±36.3	573.75±49.16	402.56±42.58
t	---	0.583	5.751	0.712	6.635
P	---	0.357	0.024	0.208	0.017

Comparison of short-term efficacy

The effective rate and clinical benefit rate of the observation group were 62.50% and 92.50%, respectively, which were significantly higher than those in the control group. The difference was statistically significant (P<0.05). as can be seen from table 2.

Relationship between serum levels of MMP-7 and MMP-9 and efficacy in patients

There was no significant difference in serum levels of MMP-7 and MMP-9 before treatment in all patients with CR+PR, SD, and PD (P=0.748, 0.962), but there was a significant difference after treatment (P=0.031, 0.029), and the serum MMP-7, MMP-9 levels showed a trend of

Table 5: Comparison of Karnofsky score and improvement of quality of life before and after treatment in the two groups

Group	Case	Karnofsky Score (Point, $\bar{x} \pm s$)		Improvement Of Quality Of Life [N (%)]			
		Before Treatment	After Treatment	Significantly Effective	Effective	Stable	Invalid
Observation	40	73.92±6.4	93.74±5.2	10 (33.33)	15 (50.00)	4 (13.33)	1 (3.33)
Control	40	74.78±7.54	80.33±6.57	6 (20.00)	9 (30.00)	10 (33.33)	5 (16.67)
T/X^2	---	0.391	7.325	6.834			
P	---	0.622	0.013	0.015			

Table 6: Comparison of adverse reactions in the two groups (n=40)

Group	Leukopenia		Thrombo cytopenia	Anemia		Liver Damage		Renal Damage	Gastrointestinal Symptoms			
	I-II	III-IV	I-II	III-IV	I-II	III-IV	I-II	III-II	I-II	III-IV	I-II	III-IV
Observation	7	1	8	1	4	1	7	0	1	0	6	1
Control	15	5	14	5	12	6	8	1	1	0	13	5
X^2	5.093		4.561	6.794		0.785		0.692	5.056			
P	0.034		0.036	0.028		0.142		0.158	0.037			

increasing with decreasing efficacy as shown in table 3.

Comparison of serum levels of MMP-7 and MMP-9

There was no significant difference in serum levels of MMP-7 and MMP-9 before treatment between the two groups (P=0.357, 0.208). After treatment, serum MMP-7 and MMP-9 levels in the observation group were (114.62±39.59) pg/mL and (271.93±44.31) pg/mL, which were significantly lower than those in the control group (P=0.024, 0.017) as shown in table 4.

Comparison of karnofsky score and improvement of quality of life before and after treatment

After treatment, the Karnofsky score of the observation group was (93.74±5.21) points, which was significantly higher than that of the control group (80.33±6.57) points (P=0.013). The improvement effect of the quality of life of the observation group after treatment was statistically significant compared with the control group. (P=0.015) as shown in table 5.

Comparison of adverse reactions

The incidence of leukopenia, thrombocytopenia, anemia, and gastrointestinal symptoms in the observation group were significantly lower than those in the control group (P=0.034, 0.036, 0.028, 0.037) as shown in table 6.

DISCUSSION

Lung cancer is the most common malignancy with the highest incidence and mortality in the world. Smoking and air pollution are high risk factors for lung cancer (Ioannidis *et al*., 2013; Gridelli *et al*., 2011). Lung cancer is divided mainly into small cell lung cancer and non-small cell lung cancer dealing with pathological. Presently, the common chemotherapy and radiotherapy approaches in patients with lung cancer often leads into severe toxicity and adverse possessions. Research and development of anti-lung cancer medications remained a warm issue (Ando *et al*., 2006). Osimertinib is the 3rd epi-dermal growth factor receptor tyrosine kinase inhibitor [EGFR-TKI] permitted to treat the locally advanced and/or metastatic, non-small cell lung cancer with EGFR gene T790M mutation. Efficacy is substantial, and is well tolerated. Side effects are more simply recognized than the 1st and 2nd generation EGFR-TKI (Wang *et al*., 2014). Patients who are treated with other EGFR-TKI therapy and experience drug resistance, Osimertinib is a novel choice for such patients (Greig, 2016). Related literature has shown, that Osimertinib, can be linked in blocking downstream beckoning alleyways in EGFR, the chief probable tools are “Hindering mutant EGFR homo dimerization” and “Tempting EGFR mutant cell deprivation” (Rosell *et al*., 2011).MMP-7 gene, has a strong matrix-degrading activity, and a extensive assortment of substrate specificity, that includes fibronectin; type IV collagen; laminin; proteoglycan type I gelatin and soluble elastin substrates. It is largely articulated in the cytoplasm of definite cancer cells or tumor infiltration marginal edges, and is fewer controlled by undesirable tissue inhibitor in matrix metallo-proteinase (TIMP). At what time normal cells and/or cancer tissue secrete MMP-7, they are completely in the custom of sedentary zymogen (ProMMP-7). ProMMP-7 that might be stimulated by means of various factors and MMPs may also control their personal goings-on by interactions. It can be understood that in physical or pathological circumstances, MMP-7 controls the deprivation course of ECM through interaction with other associates of the MMPs family like MMP-1, 3, and 9, (Newby *et al*., 2006). MMP-7 can also cause the migration of endothelial cells. At the same time, the proteolytic activity of MMP-7 expressed in endothelial cells relaxes the structure of blood vessels, making it

easier for tumor cells to infiltrate blood vessels. MMP-9 is a warm plug in tumor investigation in current ages. It is one of the fellows of the metallo-proteinase family that are closely connected to tumorigenesis, development, and its immigration. It contributes in the route remodeling by stimulating collagenase-degrading structures in the respiratory tract and lungs. It also destroys extracellular matrix and basement membrane on the shallow of lung cancers to encourage tumor attack and metastasis. In the interim, it is a significant watchdog of angiogenesis in MMP-9 tumors (Eccles, 2004; Kumar *et al*., 2012). Research has established that MMP-9 is allied with cancer lymph node or distant metastasis and has a noteworthy consequence over cancer prognosis (Yurdakul *et al*., 2014).

This present study used Osimertinib in combination with conventional chemotherapy to treat patients with advanced non-small cell lung cancer. Its short-term efficacy and clinical benefit were greater as compare to those with conventional chemotherapy alone, suggesting that Osimertinib adjuvant therapy, effectively improved non-small cell lung cancer patient's short-term efficacy, clinical benefit rate and enhanced prognosis. The initial study of the association among serum MMP-7, MMP-9 levels and short-term efficacy in this study presented that there were significant differences in serum MMP-7 and MMP-9 levels after treatment in patients with different efficacy, which augmented with lessening effectiveness. The outcome of treatment on MMP-7 and MMP-9 expression may be related to the therapeutic influence. Osimertinib enhanced the inhibition of MMP-7 and MMP-9 expression in the patients with better short-term efficacy and clinical benefit of the observation group. Osimertinib played an adjuvant anti-tumor effect by inhibiting the expression of MMP-7 and MMP-9, and the specific mechanism still needs further research to prove.

The quality of life of tumor patients is generally low, which seriously affects the patient's tolerance to chemotherapy and has a serious impact on the patient's physical and psychological rehabilitation prognosis. While using chemotherapy to treat tumors, it produces serious toxic and side effects, leading to hair loss, nausea, vomiting, fatigue, etc., while leukopenia, thrombocytopenia, and hemoglobin decrease make the patient's resistance and physical strength significantly lower, their mental state is poor, and the quality of life after treatment is difficult to improve significantly.

CONCLUSION

The results of this study indicate that Osimertinib combined with chemotherapy effectively improved the quality of life of patients, reduced adverse reactions and had a high safety. Hence, the treatment of Osimertinib combined with chemotherapy for patients with advanced non-small cell lung cancer effectively enhanced the inhibition of the expression of MMP-7 and MMP-9 related to tumor neovascularization, reduced the side effects of chemotherapy and improved the quality of life. It has the effect of reducing toxicity and increasing efficiency. It is safe and worthy of application.

REFERENCES

Ando M, Okamoto I, Yamamoto N, Takeda K, Tamura K, Seto T, Ariyoshi Y and Fukuoka M (2006). Predictive factors for interstitial lung disease, antitumor response, and survival in non–small-cell lung cancer patients treated with gefitinib. *J. Clin. Oncol.*, **24**(2): 2549-56.

Attia M, Huet E, Gossard C, Menashi S, Tassoni MC and Martelly I (2013). Early events of overused supraspinatus tendons involve matrix metalloproteinases and EMMPRIN/CD147 in the absence of inflammation. *Am. J. Sport. Med.*, **41**(4): 908-17.

Banerjee P, Jana S, Chakraborty S and Swarnakar S (2013). Inflammation and MMPs in alcohol-induced liver diseases and protective action of antioxidants. *Indian. J. Biochem. Biophys.*, **50**(5): 377-86.

Bo MK, Bae YH and Koh J (2016). AZD9291 overcomes T790 M-mediated resistance through degradation of EGFR L858R/T790M, in non-small cell lung cancer cells. *Invest. New. Drug.*, **34**(4): 407-15.

Eccles SA (2004). Parallels in invasion and angiogenesis provide pivotal points for therapeutic intervention. *Int. J. Dev. Biol.*, **48**(3): 583-98.

Felip E and Rosell R (2008). Pemetrexed, non-small-cell lung cancer, NSCLC. *Ther. Clin. Res. Man.*, **4**(1): 579-85.

Greig SL (2016). Osimertinib: First Global Approval. *Drugs*, **76**(1): 263-73.

Gridelli C, De MF, Di MM, Cortinovis D, Cappuzzo F and Mok T (2011). Gefitinib as first-line treatment for patients with advanced non-small-cell lung cancer with activating epidermal growth factor receptor mutation: Review of the evidence. *Lung Can.,* **71**(1): 249-257.

Ioannidis G, Souglakos J and Georgoulias V (2013). Predicting toxicity in advanced lung cancer patients treated with platinum-based chemotherapy. *Lun. Can. Man.*, **2**(1): 281-93.

Kumar VK and Rao JS (2012). MMP-9 and uPAR regulated glioma cell migration. *Cell Adh. Mig.*, **6**(2): 509-512.

Lu S, Cheng Y, Zhou CC, Wang J, Yang JCH, Zhang PH, Zhang XQ, Wang X, Orlando M and Wu YL (2016). Meta-analysis of first-line pemetrexed plus platinum treatment in compared to other platinum-based doublet regimens in elderly east asian patients with advanced nonsquamous non-small-cell lung cancer. *Clin. Lung Can.*, **17**(3): e103-12.

Lynch CC, Hikosaka A, Acuff HB, Martin MD, Kawai N, Singh RK, Vargo-Gogola TC, Begtrup JL, Peterson TE,

Fingleton B and Shirai T (2005). MMP-7 promotes prostate cancer-induced osteolysis via the solubilization of RANKL. *Can. Cel.*, **7**(2): 485-96.

Newby AC (2006). Matrix metalloproteinases regulate migration, proliferation, and death of vascular smooth muscle cells by degrading matrix and non-matrix substrates. *Card. Vas. Res.*, **69**(2): 614-24.

Rosell R, Molina M A, Costa C, Simonetti S, Gimenez-Capitan A, Bertran-Alamillo J, Mayo C, Moran T, Mendez P, Cardenal F and Isla D (2011). Pretreatment EGFR T790M mutation and BRCA1 mRNA expression in erlotinib-treated advanced non-small-cell lung cancer patients with EGFR mutations. *Clin. Can. Res.*, **17**(5): 1160-68.

Scagliotti G, Hanna N, Fossella F, Sugarman K, Blatter J, Peterson P, Simms L and Shepherd FA (2009). The differential efficacy of pemetrexed according to NSCLC histology: A review of two Phase III studies. *Oncol.*, **14**(3): 253-63.

Wang Z, Duan J and Guo Q (2008). A phase II clinical trial of celecoxib combined with platinum-based chemotherapy in the treatment of patients with advanced NSCLC as first-line treatment. *Chin. J. Lung. Can.*, **11**(1): 425-30.

Wang Z, Chen R, Wang S, Zhong J, Wu M, Zhao J, Duan J, Zhuo M, An T, Wang Y and Bai H (2014). Quantification and Dynamic Monitoring of EGFR T790M in Plasma Cell-Free DNA by Digital PCR for Prognosis of EGFR-TKI Treatment in Advanced NSCLC. *Plos. One.*, **9**(1): e110780.

Yoshizawa A, Fukuoka J, Shimizu S, Shilo K, Franks TJ, Hewitt SM, Fujii T, Cordon-Cardo C, Jen J and Travis WD (2010). Overexpression of phospho-eIF4E is associated with survival through AKT pathway in non-small cell lung cancer. *Clin. Can. Res.*, **16**(1): 240-248.

Yurdakul A, Akyurek N, Yilmaz S, Karakaya J, MEMİŞ L and Ozturk C (2012). Prognostic impact of matrix metalloproteinases (MMP-9 and MMP-2) and vascular endothelial growth factor expression in non-small cell lung cancer. *Turk. J. Med. Sci.,* **42**(1): 281-288.

Zhang JG, Wang JJ, Zhao F, Liu Q, Jiang K and Yang GH (2010). MicroRNA-21 (miR-21) represses tumor suppressor PTEN and promotes growth and invasion in non-small cell lung cancer (NSCLC). *Clin. Chim. Acta*., 4**11**(11): 846-852.