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ORIGINAL ARTICLE

# Relationship of Cytokines to Symptom Distress and Symptom Clusters Among Non-small-cell Lung Cancer Patients Receiving Gefitinib Treatment: A Pilot Study

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**Purpose:** Several cytokines involved in the development of sickness behaviors are considered to be related to the development of cancer symptoms. However, the mechanism of cytokines' involvement in symptom relief with gefitinib treatment remains unknown. This study analyzed the relationships between single symptoms/symptom clusters and cytokines in patients with advanced non-small-cell lung cancer (NSCLC) at pretreatment and at 1 month, 3 months, and 6 months after gefitinib treatment.

**Methods:** Fifty-seven patients with NSCLC were recruited via convenience sampling from a group of thoracic oncology patients in Northern Taiwan. Research measures included the use of the M.D. Anderson Symptom Inventory—Taiwan form and enzyme-linked immunosorbent assays. Statistical analyses included descriptive statistics and generalized estimating equation analysis.

**Results:** Positive relationships were observed between interleukin (IL)-2 and nausea ($p < 0.01$), distress ($p < 0.05$), drowsiness ($p < 0.01$), lack of appetite ($p = 0.01$), sum of symptom severity scores ($p < 0.01$), and a gastrointestinal symptom cluster ($p < 0.01$). Positive relationships between IL-6 and sadness ($p < 0.05$), lack of appetite ($p < 0.05$), and pain ($p = 0.014$), and a negative relationship between IL-6 and difficulty remembering ($p < 0.05$) were also observed. In addition, positive relationships were observed between IL-10 and fatigue ($p < 0.01$), lack of appetite ($p < 0.01$), drowsiness ($p < 0.01$), sadness ($p < 0.05$), sum of symptom severity scores ($p < 0.01$), and a general symptom cluster ($p < 0.01$).

**Conclusion:** These results may provide a basis for understanding possible mechanisms of symptom distress in patients with NSCLC; this may possibly lead to the identification of a target for effective symptom management, i.e., focusing on the inflammation pathway for the treatment of detrimental effects of cytokine-induced inflammatory responses.



## 1. Introduction

Lung cancer was the most commonly diagnosed cancer worldwide and the leading cause of cancer death in males in 2008.[1] Moreover, this type of cancer has the second highest yearly mortality rate among all cancers in Taiwan.[2] Non-small-cell lung cancer (NSCLC), which is the cause of the majority of lung cancer cases, is difficult to treat due to the poorly understood pathological mechanisms associated with this disease. Patients with advanced lung cancer are particularly vulnerable to the symptoms of breathlessness, fatigue, and anxiety, which impact patients' functions.[3] Owing to the poor prognosis associated with this type of lung cancer, symptom management is crucial in oncological treatment of NSCLC. Research has shown that the epidermal growth factor receptor is a significant factor in the development and growth of lung cancers.[4] Epidermal growth factor receptor therapy involves the use of specific drugs to attack some special structures of the cancer cell, by taking advantage of the absence of such structures in normal cells; indeed, targeting these specific structures allows such targeted therapies to kill cancerous cells without harming normal cells. The most commonly used drug to treat lung cancer in Taiwan is Iressa (i.e.,

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gefitinib or ZD1839). Iressa is an orally active, selective, epidermal growth factor receptor tyrosine-kinase inhibitor, which blocks signal transduction pathways implicated in the proliferation and survival of cancer cells. However, patients often experience multiple physical symptoms, such as skin toxicity, diarrhea, and acute interstitial pneumonia, during and after treatment.[5,6] Taken together, a cancer-related symptom is the activation of the immune system in response to a tumor, itself, or treatments for the disease.

In animal models, similarities have been identified between cancer symptoms and many symptoms of sickness behavior triggered by the lipopolysaccharides of bacteria, including physiological reactions such as fever, pain, cachexia, and lowered functioning of the hypothalamic–pituitary–adrenal axis and the autonomic nervous system[7]; moreover, behavioral responses, including reduced general activities, drowsiness, cognitive impairment, reduced social and exploratory behaviors, reduced sexual behavior, and reduced food intake, have also been identified in such animal models.[8] Previous studies elaborated on the induction of sickness behaviors similar to cancer symptoms after the injection of cytokines in animals and humans.[7] Together these findings suggest that peripheral levels of proinflammatory cytokines are related to the immune responses that affect physiological, psychological, and behavioral alterations.

Lung cancer patients often experience multiple symptoms at the same time, i.e., a symptom cluster. Two major symptom clusters, i.e., general and gastrointestinal, have been identified in Taiwanese lung cancer patients.[9] Several cytokines involved in the development of sickness behaviors are thought to be involved in producing symptom clusters.[7] Previous research has shown that behaviors such as fatigue, anemia, and poor cognitive function in cancer patients are related to the effects of interleukin (IL)-6 on the production of red blood cells,[10,11] whereas serum levels of IL-2 have been correlated to fatigue,[12] pain,[13] and wasting syndrome.[14] In addition, IL-10 serum levels are linked to fatigue.[15] Loberg et al[16] indicated that cytokine overproduction was associated with the development of metastases and hastened death. However, only a limited number of studies provided data to support the contribution of cytokines to the development of distress related to single and cluster symptoms.[17] Indeed, Gilbertson-White et al[7] suggested that symptom research should include the measurement of cytokines as biomarkers for cancer symptoms. If symptom clusters and the possible effects of biological metastasis on symptom management can be evaluated in depth, this information can be used to improve the management of NSCLC symptoms. Further research is needed to truly understand the relationship between proinflammatory cytokines and sickness behaviors, which would enhance our ability to develop tools for oncology nurses to help them evaluate multiple concurrent symptoms. Based on this consideration, we hypothesized that cancer-related symptom clusters experienced by NSCLC patients share a common cytokine-based mechanism. Because of the potential involvement of inflammatory cytokines in the development of symptoms, we selected three specific cytokines, i.e., IL-2, IL-6, and IL-10, for further observation; more specifically, we chose to investigate the concentrations of these cytokines in response to physiological and psychological distress. Furthermore, when patients receive treatment with gefitinib, they often experience some degree of relief of cancer symptoms. However, the level of toxicity and disease-related symptoms should be assessed every 28 days, as suggested by Haura et al.[18] Indeed, after treatment, 43.1% of patients experience apparent improvement in symptom distress within 14–410 days (56 days on average).[19] Therefore, in order to continue the evaluation of the prevalence, severity, and level of distress in the course of treatment, the present article provides follow-up data for four time points in the course of treatment, i.e., at pretreatment and at 1 month, 3 months, and 6 months after gefitinib treatment. The purpose of this study was to analyze the relationships between single symptoms/symptom clusters and cytokines in advanced NSCLC patients after receiving gefitinib treatment.

## 2. Methods

### 2.1. Sample and settings

This study employed the convenience sampling method to recruit participants from a larger group of thoracic oncology in- and outpatients at a medical center in northern Taiwan. The following selection criteria were employed: (1) aged over 18 years; (2) with a pathological diagnosis of NSCLC; (3) with clear consciousness and the ability to communicate in Mandarin or Taiwanese; and (4) receiving a daily dose of gefitinib. A medium effect size (0.25) was used to calculate the sample size. A $p$ value $< 0.05$ was taken to be significant. The sample size needed was 25 for each course of treatment to maintain power greater than 70%. In order to improve the latter's quality and efficiency, we attempted a feasibility study designed to test logistics and gather information prior to a larger study; this required a minimum of 10 participants throughout the course of treatment. Overall, 57 NSCLC participants were recruited. In total, 11 patients completed 6 months of sample interval data collection. Note that 25 patients completed the 3-month sample interval; however, ten patients changed treatment, eight patients died and fourteen patients refused treatment, thus reducing the sample size to 11.

### 2.2. Instruments

#### 2.2.1. Demographic and medical characteristics

Relevant demographic and medical information was obtained from each patient's chart and through face-to-face interviews of patients. Demographic data included information on age, occupation, gender, education, marital status, religion, etc. Medical characteristics included disease diagnoses, metastases, treatment methods, and other applicable medical history.

#### 2.2.2. Karnofsky Performance Scale

Scores obtained with the Karnofsky Performance Scale (KPS) are strong predictors of the patients' quality of life.[20] The KPS is rated using a scale of 1–100, where 0 represent dead and 100 represents normal. Scores of $\leq$40 indicate dependency, whereas 50–60 and $\geq$70 indicate the need for assistance and ability to provide self-care, respectively.

#### 2.2.3. Taiwanese version of the MD Anderson Symptom Inventory

The Taiwanese version of the MD Anderson Symptom Inventory (MDASI-T) was used in this study and was translated by Lin et al.[21] The original form was developed by Cleeland et al[22] and is a multidimensional assessment form with 19 evaluation items, which are designed to evaluate the severity of multiple cancer symptoms using an 11-point rating scale. Cancer patients assessed the severity of cancer symptoms using the 11-point scale, in which "0" represents "not present" and "10" represents a symptom of severity level that is "as bad as you can imagine"; more specifically, 1–4 points represent a mild level of symptom severity, whereas 5–6 points and $\geq$7 points represent a moderate level and a severe level of symptom severity, respectively. Studies regarding the severity of symptoms in cancer patients have a very high level of internal consistency (i.e., Cronbach $\alpha$ of up to 0.85).[22] The Cronbach $\alpha$ value of the MDASI-T was 0.85. Two major symptom clusters (i.e., general and gastrointestinal symptoms) were identified in Lin el al's[21] study. The general symptoms investigated in this

study include distress, sadness, fatigue, sleepiness, dry month, lack of appetite, difficulty remembering, drowsiness, shortness of breath, numbness, and pain. Gastrointestinal symptoms investigated in this study included nausea and vomiting.

#### 2.2.4. Cytokines

At each visit, a research assistant collected a blood sample (8–10 mL) from each patient between 9 AM in the morning and 12 PM in the afternoon. The samples were stored for 4–6 hours at 4°C until the specimens were transported to the laboratory, where this storage temperature was maintained. To consider the role of cytokine activation and production on the function of the immune system, three cytokines were measured: IL-2, IL-6, and IL-10. The serum was separated from each blood sample and stored at −70°C in accordance with the standard procedures. Serum IL-2, IL-6, and IL-10 levels were tested using enzyme-linked immunosorbent assays (ELISAs); more specifically, the DuoSet ELISA development kit IL2, IL6, and IL10 (R&D Systems, Minneapolis, Minnesota, USA) was used according to the standard operating procedures outlined by the manufacturer. Two parts of general protocol were performed, including plate preparation and assay procedure. Color development was measured at 450 nm using an automated microplate ELISA reader. A standard curve was run on each assay plate using recombinant human IL-2, IL-6, and IL-10 (R&D Systems) in serial dilutions.

### 2.3. Procedures

Prior to conducting this study, approval was obtained from the Human Subject Committee of Taipei Medical University, Taipei, Taiwan. Participants were only asked to undergo venipuncture, which carries minimal risk. However, patients were instructed to use ice packs in some instances (i.e., bruise). As previously mentioned, this study collected data from thoracic oncology in- and outpatients at a medical center in northern Taiwan who were referred by the treating physician. The research assistant explained the research process and purposes of this study to the patients, and those who were willing to participate in the study were asked to sign a written letter of consent. Participation in this study did not affect the patients' right to medical care, and the data provided were kept in strict confidence. During the interview process, the research assistant instructed the patients on how to complete the questionnaire properly; more specifically, patients were requested to fill in demographic and medical characteristic information and MDASI-T over the course of about 10 minutes. If a patient could not complete the questionnaire independently, the research assistant read the questions to the patient and recorded the patient's answers. After the forms were completed, the research assistant extracted 10 mL of blood via venipuncture, and the blood samples were sent to the laboratory for serum separation and analysis. Data collection procedures were repeated at 1 month, 3 months, and 6 months after baseline.

### 2.4. Statistical analysis

Descriptive statistics were used to analyze the demographic and medical characteristic variables, symptom distress, symptom interference, and cytokine levels. Generalized estimating equations (GEEs) were used to account for within-individual correlation arising from repeated measurements on the same individual.[23] Using GEEs for longitudinal data analysis produced more efficient and unbiased regression estimates to analyze the relationships between individual symptoms and cytokine levels by adjusting for other relevant variables (i.e., age and the Karnofsky functional status) and the influence of repetitive measurements (i.e., time effect and baseline as the reference group). Serum levels of cytokines were analyzed in relation to the change in symptom distress over time. In order to evaluate the effect of cytokines on symptoms after adjusting for time effect, age, and KPS score, the working correlation structure was correctly specified as an autoregressive(1) model. Dependent variables included single symptoms, total symptoms, symptom clusters 1 and 2 (i.e., the general cluster and the gastrointestinal cluster, respectively), and independent variables included time period factors, single cytokines, age, and KPS as covariates. The results of the GEE analyses were expressed as coefficients, along with the corresponding 95% confidence interval, associated $p$ values, and estimates of correlations. In each of the models, the level of cytokine was monitored as continuous, and the symptom distress and symptom cluster were evaluated. Only those variables that showed significance at the $p < 0.05$ level were considered in the final model. All statistical analyses were conducted using Statistical Package for Social Science software version 17.0 (SPSS Inc., Chicago, Illinois, USA), and $p < 0.05$ was chosen as the level of statistical significance. Results were depicted as Mean +/− standard deviation.

## 3. Results

### 3.1. Demographic information and characteristics of the sample

Table 1 shows the demographic characteristics of the sample. The average age of the participants was 62.33 ± 12.61 years. The

**Table 1** Demographic and medical characteristics of sample ($N = 57$)

| Variable | |
|---|---|
| Age (y) | 62.33 ± 12.61 |
| Educational level (y) | 9.39 ± 5.60 |
| Living with family | 4 ± 2.04 |
| Karnofsky score | 84.87 ± 10.97 |
| Karnofsky score (1 mo, $n = 40$) | 84.91 ± 9.66 |
| Karnofsky score (3 mo, $n = 25$) | 89.13 ± 2.88 |
| Karnofsky score (6 mo, $n = 11$) | 88.18 ± 6.03 |
| Variable | $n$ (%) |
| Sex | |
| Male | 24 (42.1) |
| Female | 33 (57.9) |
| Employment status | |
| Employed | 14 (24.6) |
| Unemployed or retired | 43 (75.4) |
| Marital status | |
| Married | 50 (87.7) |
| Divorced | 2 (3.5) |
| Widowed | 4 (7.0) |
| Single | 1 (1.8) |
| Educational level | |
| None | 9 (15.8) |
| Elementary | 14 (24.6) |
| Junior high | 9 (15.8) |
| Senior high | 5 (8.8) |
| College or above | 20 (35) |
| Religious affiliation | |
| Buddhism | 33 (57.9) |
| Taoism | 9 (15.8) |
| Protestant | 4 (7.0) |
| None | 11 (19.3) |
| Treatment | |
| Operation | 21 (36.8) |
| Chemotherapy | 29 (50.9) |
| Radiotherapy | 18 (32.1) |
| Metastasis | |
| Yes | 47 (82.5) |
| No | 10 (17.5) |
| Stage of cancer | |
| II | 1 (1.7) |
| III | 7 (12.3) |
| IV | 49 (86) |

**Table 2** Levels of cytokines (pg/mL) in Taiwanese patients with non-small-cell lung cancer

| Cytokine | Baseline ($n = 57$) | 1 mo ($n = 40$) | 3 mo ($n = 25$) | 6 mo ($n = 11$) |
|---|---|---|---|---|
| IL-2 | 72.16 ± 58.47 | 65.69 ± 61.24 | 62.34 ± 66.59 | 85.98 ± 90.18 |
| IL-6 | 64.35 ± 39.17 | 66.64 ± 40.59 | 58.35 ± 40.56 | 55.29 ± 33.27 |
| IL-10 | 87.99 ± 68.59 | 85.70 ± 68.29 | 68.39 ± 73.62 | 61.73 ± 88.56 |

Data are presented as mean ± standard deviation.
IL = interleukin.

average KPS score for the baseline sample was 84.87 ± 10.97. The score for the 6-month sampling declined to 88.18 ± 6.03 from the 3-month score of 89.13 ± 2.88.

### 3.2. Cytokine levels

ELISA was performed for all blood specimens that were drawn at predetermined times during the treatment period. A significant change was noted in the IL-2 level (85.98 ± 90.18 pg/mL) in the 3rd month (Table 2). Levels of each cytokine and each symptom distribution throughout the course of treatment in Taiwanese patients with NSCLC are shown in Figure 1.

### 3.3. Summary of GEE analysis for symptom severity as predicted by cytokines after adjusting for time effects

This study employed a GEE model and found that as IL-2 increased, symptoms of nausea, distress, lack of appetite, and drowsiness; total symptoms; and a gastrointestinal symptom cluster also increased (Table 3). Moreover, as IL-6 increased, pain, lack of appetite, and sadness also increased (Table 3), but difficulty remembering decreased. As IL-10 levels increased, symptoms of fatigue, lack of appetite, drowsiness, and sadness; total symptoms; and a general symptom cluster also increased (Table 3).

The $Z$ value of the general symptom cluster and the IL-10 concentration was 5.01, and the estimated coefficient was 0.04, which suggests that the general symptom cluster and the IL-10 concentration were significantly and positively correlated. This indicates that a unit increase in the concentration of IL-10 during gefitinib treatment of NSCLC would lead to an increase in the severity of the general symptom cluster by 0.04 points.

The $Z$ value of the gastrointestinal symptom cluster and the IL-2 concentration was 6.83, and the estimated coefficient was 0.004, suggesting that the gastrointestinal symptom cluster and IL-2 concentration were significantly and positively correlated. This indicates that a unit increase in the concentration of IL-2 during gefitinib treatment of NSCLC patients would lead to an increase in the severity of the gastrointestinal symptom cluster by 0.004 points.

## 4. Discussion

These data represent the first longitudinal analysis of cytokine production and symptom distress in patients with NSCLC in Taiwan. The strength of the present study is its longitudinal nature, which contrasts with previous cross-sectional studies in which the patient groups were not followed throughout the time of cancer treatment, to reliably associate the symptom status with the cancer and its treatment. Our study found that symptom improvement increased slightly and was correlated with a few cytokines after receiving gefitinib treatment. Significantly, the lack of appetite experienced by NSCLC patients was related to IL-6 levels. In addition, levels of cytokines prior to treatment were relatively high in most cases and tended to decline in the 1st month after treatment. By the 3rd month after treatment, cytokine levels had significantly dropped with some relief of symptoms. The fact that the serum cytokine level changed significantly with obviously improved symptoms implies that relationships exist among cytokine levels, the tumor, and symptoms. Although this may suggest that gefitinib treatment is effective in treating patients with NSCLC and improves symptom severity, cell-cycle targeted therapy has been used, and tumor response results should be observed closely for indications of levels of cytokines and severe symptom responses. In fact, this is worthy

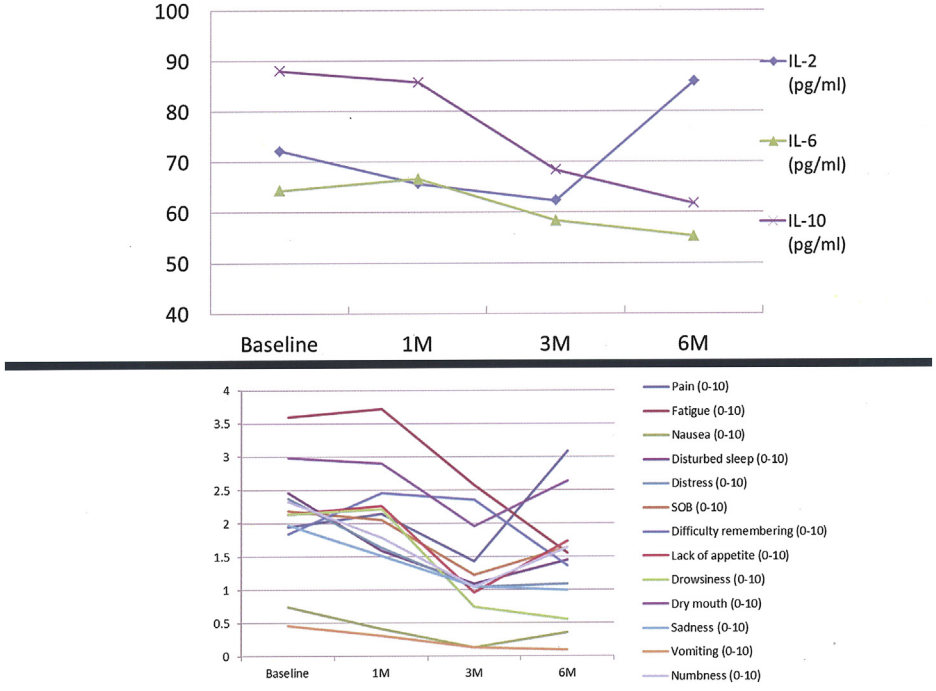


**Figure 1** Levels of cytokine and symptom distress in Taiwanese patients with non-small-cell lung cancer. IL = interleukin; SOB = shortness of breath.

**Table 3** Changes in IL-2, IL-6, and IL-10 related to changes in symptom distress and symptom cluster over 6 months of gefitinib treatment

| Symptom item | $\beta$ | SE | 95% confidence interval | | $Z$ | $p$ |
|---|---|---|---|---|---|---|
| | | | Lower | Upper | | |
| *Changes in IL-2* | | | | | | |
| Pain | 0.008 | 0.0061 | −0.004 | 0.020 | 1.614 | 0.204 |
| Fatigue | 0.008 | 0.0041 | 0.000 | 0.016 | 3.555 | 0.059 |
| Nausea | 0.004 | 0.0014 | 0.001 | 0.006 | 6.938 | 0.008** |
| Sleep disturbance | −0.001 | 0.0046 | −0.010 | 0.008 | 0.058 | 0.810 |
| Distress | 0.012 | 0.0060 | 0.000 | 0.024 | 4.157 | 0.041* |
| SOB | 0.001 | 0.0032 | −0.005 | 0.007 | 0.137 | 0.711 |
| Difficulty remembering | −0.001 | 0.0024 | −0.006 | 0.004 | 0.206 | 0.650 |
| Lack of appetite | 0.010 | 0.0042 | 0.002 | 0.018 | 5.542 | 0.019* |
| Drowsiness | 0.009 | 0.0032 | 0.003 | 0.015 | 7.733 | 0.005** |
| Dry mouth | −0.002 | 0.0048 | −0.011 | 0.008 | 0.129 | 0.720 |
| Sadness | 0.005 | 0.0066 | −0.008 | 0.018 | 0.592 | 0.442 |
| Vomiting | 0.000 | 0.0004 | 0.000 | 0.001 | 0.869 | 0.351 |
| Numbness | 0.005 | 0.0071 | −0.009 | 0.019 | 0.482 | 0.487 |
| Total | 0.004 | 0.0022 | 0.000 | 0.009 | 4.014 | 0.045* |
| General symptom cluster | 0.053 | 0.0290 | −0.004 | 0.109 | 3.298 | 0.069 |
| Gastrointestinal symptom cluster | 0.004 | 0.0015 | 0.001 | 0.007 | 6.825 | 0.009** |
| *Changes in IL-6* | | | | | | |
| Pain | 0.014 | 0.0064 | 0.002 | 0.027 | 5.058 | 0.025* |
| Fatigue | 0.000 | 0.0067 | −0.013 | 0.013 | 0.002 | 0.967 |
| Nausea | 0.004 | 0.0034 | −0.002 | 0.011 | 1.758 | 0.185 |
| Sleep disturbance | −0.008 | 0.0090 | −0.025 | 0.010 | 0.748 | 0.387 |
| Distress | 0.013 | 0.0121 | −0.011 | 0.036 | 1.064 | 0.302 |
| SOB | 0.002 | 0.0086 | −0.015 | 0.019 | 0.047 | 0.828 |
| Difficulty remembering | −0.013 | 0.0045 | −0.022 | −0.004 | 8.301 | 0.004** |
| Lack of appetite | 0.012 | 0.0048 | 0.002 | 0.021 | 6.029 | 0.014* |
| Drowsiness | 0.006 | 0.0089 | −0.011 | 0.024 | 0.499 | 0.480 |
| Dry mouth | −0.009 | 0.0110 | −0.030 | 0.012 | 0.680 | 0.410 |
| Sadness | 0.020 | 0.0063 | 0.008 | 0.033 | 10.531 | 0.001** |
| Vomiting | −0.001 | 0.0006 | −0.002 | 0.000 | 1.512 | 0.219 |
| Numbness | 0.021 | 0.0177 | −0.013 | 0.056 | 1.435 | 0.231 |
| Total | 0.004 | 0.0033 | −0.003 | 0.010 | 1.264 | 0.261 |
| General symptom cluster | 0.045 | 0.0424 | −0.038 | 0.128 | 1.135 | 0.287 |
| Gastrointestinal symptom cluster | 0.004 | 0.0035 | −0.003 | 0.011 | 1.337 | 0.248 |
| *Changes in IL-10* | | | | | | |
| Pain | 0.004 | 0.0038 | −0.004 | 0.011 | 1.030 | 0.310 |
| Fatigue | 0.005 | 0.0018 | 0.001 | 0.008 | 7.389 | 0.007** |
| Nausea | 0.002 | 0.0015 | −0.001 | 0.005 | 1.342 | 0.247 |
| Sleep disturbance | 0.001 | 0.0043 | −0.008 | 0.009 | 0.026 | 0.871 |
| Distress | 0.009 | 0.0046 | 0.000 | 0.018 | 3.510 | 0.061 |
| SOB | 0.003 | 0.0020 | −0.001 | 0.007 | 1.834 | 0.176 |
| Difficulty remembering | −0.001 | 0.0027 | −0.007 | 0.004 | 0.200 | 0.655 |
| Lack of appetite | 0.007 | 0.0033 | 0.000 | 0.013 | 3.969 | 0.046* |
| Drowsiness | 0.006 | 0.0029 | 0.000 | 0.011 | 3.989 | 0.046* |
| Dry mouth | −0.002 | 0.0037 | −0.009 | 0.006 | 0.177 | 0.674 |
| Sadness | 0.014 | 0.0049 | 0.004 | 0.023 | 7.884 | 0.005** |
| Vomiting | 0.000 | 0.0002 | −0.001 | 0.000 | 1.344 | 0.246 |
| Numbness | 0.005 | 0.0075 | −0.010 | 0.020 | 0.455 | 0.500 |
| Total | 0.003 | 0.0016 | 0.000 | 0.007 | 4.958 | 0.026* |
| General symptom cluster | 0.044 | 0.0197 | 0.005 | 0.083 | 5.009 | 0.025* |
| Gastrointestinal symptom cluster | 0.002 | 0.0016 | −0.001 | 0.005 | 1.086 | 0.297 |

*$p < 0.05$.
**$p < 0.01$.
IL = interleukin; SE = standard error; SOB = shortness of breath.

of further study and could lead to mechanism-driven symptom management strategies.

In research on relationships between individual symptoms and cytokines, correlations have been noted in a few studies. A study by Meyers et al[24] pointed out that more than 70% of untreated lung cancer patients experience difficulty remembering, and more than one-third of these patients have frontal lobe damage. Studies in recent years have suggested that IL-6 is related to cognitive function. Scalzo et al[25] pointed out that the performance of IL-6 is evidenced by the fact that brain functions, such as memory and learning, are damaged in Parkinson disease patients. Significant positive collections were observed between IL-6 and cognitive function. In addition, when the IL-6 level is relatively high, integration and attention decline.[11,26] By contrast, the present study found that difficulty remembering was negatively correlated with the IL-6 level. In fact, our results indicate that a unit increase in the level of IL-6 will reduce the severity of difficulty remembering by 0.01 points on the MDASI-T's 0−10 scale. The hypothesis that IL-6 is mainly proinflammatory and neurodegenerative is also challenged by studies that have produced results suggesting that cytokines have several anti-inflammatory and immunosuppressive actions and may play a downregulating role in inflammatory conditions.[27]

In spite of this, functions of IL-6 are similar to those of major factors in the inflammatory response, which can individually alter neuroendocrine activity, learning and memory, and pleasure functions.[28] Although our findings remain speculative and contradict the work of previous findings, they do lend some credence to the core concept that inflammatory markers are related to cognition. However, IL-6 is also commonly believed to affect the growth of red blood cells adversely, which may result in fatigue and anemia.[15] Therefore, enhanced circulating levels of IL-6 could contribute to symptom distress, leading to a further decline in the functioning of NSCLC patients. The IL-10 level is highest prior to treatment and tends to decline with treatment progression, possibly because IL-10 can prevent the production of other cytokines[29]; moreover, actions of IL-10 are multifaceted.[30] For these reasons, cytokine levels can provide the evidence needed for the design of treatment approaches. This study did not discuss relevant influencing factors, such as adjunct treatment or biochemical tests, that may have affected the results. However, a preliminary understanding of cytokines helps elucidate the function of the immune system.

In this study, IL-2 was found to be related to loss of appetite, drowsiness, distress, nausea, total symptom severity score, and a gastrointestinal symptom cluster (i.e., nausea and vomiting). However, some of the literature has indicated that IL-2 is mainly correlated with fatigue and drowsiness because the release of IL-2 can affect the 24-hour sleep cycle of cancer patients, thereby leading to fatigue and drowsiness all day long.[15,31] Note that Cheville et al[3] mentioned that impaired lung function and a reduction in oxygen supply can lead to shortness of breath and fatigue in patients. Although the previously identified correlation between fatigue and IL-2 was not observed in our study, fatigue in patients often triggers other relevant symptoms. However, the underlying mechanism of fatigue has not been elucidated.

Only one recent article discussed the effects of diet on cell function by comparing 21 anorexic patients to 19 healthy individuals and found that the serum levels of IL-2 in anorexic patients was relatively higher,[32] confirming that IL-2 is positively correlated with a loss of appetite, nausea, and the gastrointestinal symptom cluster. Relationships between IL-2 and symptoms in the human body are still uncertain. However, our data are in accordance with previous findings that a correlation exists between IL-2 and factors related to the stomach and intestines. The presence of individual symptoms and a symptom cluster indicated their correlation with IL-2, thus confirming the correlation between symptoms. Moreover, a decline in IL-2 followed by a spike from the 3$^{rd}$ month to the 6th month after receiving gefitinib treatment was noted. The cycle-targeted therapy will help investigators understand symptomatology and the cytokine-release syndrome better; hence, future studies should attempt to address these topics.

This study also found that IL-6 and IL-10 were related to loss of appetite. Ninety percent of cancer patients have symptoms of anorexia—cachexia,[33] which are often related to the cancer itself or the cancer treatment. The literature emphasized that IL-6 is related to the gastrointestinal system mainly because it induces the production of corticotropin-releasing hormone, which inhibits appetite. Moreover, experiments in mice showed that IL-6 can result in nausea, loss of appetite, and weight loss.[34,35] Serum albumin levels reduced due to secretion of IL-6 mainly because IL-6 reduces the synthesis of hepatocytes.[36] As a result, loss of appetite and cachexia, ultimately resulting in reduced adipose tissues and muscle mass, occur subsequently. Szczesny et al[37] noted that IL-6 was the best indicator of postoperative complications (such as anorexia and cachexia) for 57 patients after lung surgery. Cachexia (i.e., weight loss and loss of appetite) in patients with prostate cancer was significantly correlated with IL-6.[38] In summary, the aforementioned results support the correlation of IL-6 with nausea and loss of appetite.

In this study, IL-6 and IL-10 were found to be related to sadness. According to our results, a unit increase in the level of IL-6 cytokine would lead to an increase in the severity of this symptom by 0.02 points on the MDASI-T's 0—10 scale, and a unit of increase in the level of IL-10 would reduce the psychological sadness symptom severity by 0.01 points. We also found that a unit increase in the level of IL-2 could increase the psychological distress symptom severity by 0.01 points. Some literature has asserted that depression is correlated to cytokines, particularly IL-6.[30,39] Previous studies involving the injection of IL-2 in animals and humans found that sickness behaviors were similar to symptoms of depression (e.g., loss of sleep, loss of appetite, loss of interest, and fatigue).[40] A recent preliminary study on the signal molecular profiling database has suggested that IL-2 is associated with postpartum depression in women within 4 weeks of delivery.[41] Yet, some of the literature indicated that depression and cytokines are not directly correlated. Marques-Deak et al[42] compared 46 patients with untreated serious depression with 41 healthy individuals and found that IL-6 values in the blood did not change; in a further study of female patients with different types of depression, they found no significant change in serum IL-6 levels.[42] Regarding the relationship between IL-6 and depression (i.e., sadness), Alesci et al[43] found an apparent correlation in a study of nine patients diagnosed with serious depression, and Krogh et al[44] found that patients with depression had higher levels of IL-6. Hence, the findings of the present study are consistent with those of previous studies. Although the relationship between IL-10 and psychological sadness has not been described in the literature, further exploration would be worthwhile because this is the only possible explanation for the observed negative physiological factors, especially fatigue, thereby leading to a negative energy balance and stimulating the secretion of IL-10. This results in long-term stress in patients and stimulates the hypothalamic—pituitary—adrenal axis further, which generates mental distress in patients.[33] Cytokines appear to play a role in the "yin—yang compensation effects"[45]; hence, negative feedback by cytokines may reduce its serum level. According to research findings, the level of IL-10 does not decline with treatment time, whereas the severity of psychological sadness obviously improves. Nevertheless, a large amount of articles discussed physiological changes and changes in patients working through the disease that resulted in negative emotions. Clearly, the physiological and psychological balance of cancer patients should not be neglected when dealing with symptoms.

### 4.1. Implications for practice

In summary, cytokines have been implicated in symptom development in NSCLC. Furthermore, various cytokines have been related to multiple symptoms, all of which are common in cancer. Although correlations between symptoms and cytokine levels found in this study were both similar and different as compared to that of previously published findings, the previously noted significant correlation between IL-6 and the symptom of loss of appetite was confirmed. Therefore, the data suggest that inflammatory cytokines are possibly involved in the development and relief of symptoms in NSCLC patients receiving gefitinib treatment. Note that the sample population in this study was small. However, the paucity of empirical research with a longitudinal design exploring the influences of cytokines on symptom development and severity makes additional studies necessary and important. In fact, this study may serve as a pilot for further research with larger samples. Additional work is needed to investigate carefully the longitudinal patterns of dynamic change in physical and psychological

symptoms that may be affected by disease progression, medical comorbidities, and drug toxicity, in order to strengthen the validity of these results.

The relationship between cytokines and cancer is controversial and is much discussed; therefore, this study provides a basis for understanding the underlining inflammatory mechanisms involved in the pathophysiology of symptom distress in patients with NSCLC receiving gefitinib treatment. Ultimately, these results can help identify a target for effective symptom management, i.e., a target focusing on the inflammation pathway for preventing or minimizing the detrimental effects of cytokine-induced inflammatory responses.

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