

# Serum Concentrations of IL-2 and TNF- $\alpha$ in Patients with Painful Bone Metastases: Correlation with Responses to $^{89}\text{SrCl}_2$ Therapy

Na Fang, MD<sup>1</sup>; Yong Li, MD<sup>1</sup>; Yan-song Xu, MB<sup>1</sup>; Duo Ma, MS<sup>1</sup>; Peng Fu, MS<sup>1</sup>; Hui-qi Gao, MS<sup>1</sup>; Feng-tong Gao, MB<sup>2</sup>; Hai-shan Yang, MB<sup>3</sup>; and Zhi-jie Yang, MB<sup>1</sup>

<sup>1</sup>Department of Nuclear Medicine, First Hospital of HarBin Medical University, HarBin, China; <sup>2</sup>Department of Nuclear Medicine, China-Japan Union Hospital of Jilin University, ChangChun, China; and <sup>3</sup>Department of Radiology, China-Japan Union Hospital of Jilin University, ChangChun, China

We have used  $^{89}\text{SrCl}_2$  for the palliative treatment of painful bone metastases from various malignant diseases. We studied the correlation between serum interleukin-2 (IL-2) and tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) levels and the response to  $^{89}\text{SrCl}_2$  therapy.

**Methods:** Forty-two patients (24 men and 18 women) were treated intravenously with  $^{89}\text{SrCl}_2$  at a dose of 148 MBq (4 mCi).

**Results:** The response rate was 33 of 42 (79%). In the control subjects, serum IL-2 concentrations were higher but TNF- $\alpha$  concentrations lower ( $P < 0.05$ ) than in the patients with bone metastases. After treatment with  $^{89}\text{SrCl}_2$ , IL-2 levels increased and TNF- $\alpha$  levels decreased, with maximal changes at the fourth month after therapy. After comparing the serum levels of IL-2 and TNF- $\alpha$  between responders and nonresponders, we found that these variables did not differ before  $^{89}\text{SrCl}_2$  therapy but differed significantly ( $P < 0.05$ ) after therapy. Responders had higher IL-2 and lower TNF- $\alpha$  concentrations than nonresponders. A good correlation was found between IL-2 and TNF- $\alpha$  levels and the number of metastases and pain score. **Conclusion:**  $^{89}\text{SrCl}_2$  is effective for palliation of bone pain in patients with disseminated bone metastases. In addition to managing pain,  $^{89}\text{SrCl}_2$  can improve immunity and the quality of life for most patients. Further studies are needed to elucidate the roles of IL-2 and TNF- $\alpha$  in the response to  $^{89}\text{SrCl}_2$  therapy and to evaluate their usefulness as indicators of  $^{89}\text{SrCl}_2$  efficacy.

**Key Words:**  $^{89}\text{SrCl}_2$ ; bone metastases; IL-2; TNF- $\alpha$ ; radionuclide therapy

J Nucl Med 2006; 47:242–246

**P**ain is the major presenting symptom in 75% of patients with bone metastases from various malignant diseases (1). If pressure inside the marrow cavity rises to more than 50 mm Hg or the bone periosteum is extended, bone pain may be inevitable (2). Reducing or even eradicating pain and improving the quality of life are the main concerns at the

late stage of bone involvement and may constitute important clinical problems.  $^{89}\text{SrCl}_2$ , a radiopharmaceutical proposed by Pecher in 1942 (3) for bone pain palliation in metastatic disease, has often been used for analgesia in recent years. Several studies have demonstrated the effectiveness of  $^{89}\text{SrCl}_2$  in treating painful bone metastases (4–7).

It is well known that the generation, proliferation, differentiation, and prognosis of malignant tumors correlate closely with the status of the immune system, especially with cell-mediated immunity (8). In addition to T lymphocytes, cytokines such as interleukin-2 (IL-2) and tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) also play an important role in tumor immunity. Previous studies have focused on T lymphocyte subset alterations after  $^{89}\text{SrCl}_2$  therapy (9). To our knowledge, no report has been published on the changes in serum IL-2 and TNF- $\alpha$  in patients receiving this therapy.

Therefore, the aim of this study was to explore a possible relationship between plasma IL-2, TNF- $\alpha$ , and clinical symptoms from bone metastases and to investigate the role of IL-2 and TNF- $\alpha$  in estimating the effectiveness of  $^{89}\text{SrCl}_2$  in reducing pain.

## MATERIALS AND METHODS

### Patients

All patients were selected on the basis of the following criteria: a life expectancy of 6 mo or longer; no critical organ dysfunction; and, during the 3 mo before therapy, no external-beam radiotherapy, chemotherapy, or hormone therapy that was considered to have affected the patient's immunity. In a previous study, we found no significant difference in immunologic status among patients with different types of tumor, but the status of all patients was worse than that of the control subjects. A total of 42 patients (24 men and 18 women; mean age, 54 y) were studied between December 2001 and February 2004. All had painful bone metastases and completed a 6-mo follow-up period.

The documented primary tumors were of the lung ( $n = 14$ ), breast ( $n = 10$ ), prostate ( $n = 8$ ), kidney ( $n = 3$ ), stomach/colon ( $n = 2$ ), and skin (melanoma,  $n = 1$ ). Four were of unknown

Received Jan. 4, 2005; revision accepted Nov. 7, 2005.  
For correspondence or reprints contact: Na Fang, MD, PET/CT Center, TianJin Cancer Hospital, TianJin, 300060, China.  
E-mail: fangna7859@163.com

origin. Those malignancies had been diagnosed using radiography, CT, MRI, or SPECT examinations and confirmed in most patients by postoperative histologic examination ( $n = 31$ ). The presence of bone metastases was identified by whole-body bone scanning with  $^{99m}\text{Tc}$ -labeled methylene diphosphonate. Leukocyte and platelet concentrations were greater than  $3,000/\mu\text{L}$  and  $100,000/\mu\text{L}$ , respectively. No other tumor-oriented treatment was allowed during the entire follow-up period.

The control group consisted of 20 healthy, tumor-free subjects (12 men and 8 women; mean age, 50.6 y). The patient and control groups did not significantly differ in sex or age ( $P > 0.05$ ). Neither group received immunologically altering drugs for at least 3 mo before the study.

All patients gave their written informed consent before inclusion in the study.

### Radiopharmaceuticals and Administration Modalities

For all patients, a standard single dose of 148 MBq (4 mCi) of  $^{89}\text{SrCl}_2$  (Metastron; Amersham) was slowly (about 10 min) injected into the cephalic vein.

### Protocols

**Collection of Blood Samples.** Peripheral venous blood samples (2 mL) were drawn from the patients and the control subjects into nonheparinized tubes before the onset of the therapy and monthly for 6 mo after the injection of  $^{89}\text{SrCl}_2$ . When the nonheparinized blood coagulated, serum was then separated and preserved at  $-20^\circ\text{C}$  before examination.

**Analysis of IL-2 and TNF- $\alpha$ .** Before the examination, the samples were thawed to  $4^\circ\text{C}$  and then mixed and centrifuged at 3,000 rpm for 5 min. The reagents to determine IL-2 (IL-2 radioimmunity kit) and TNF- $\alpha$  (TNF- $\alpha$  radioimmunity kit) were purchased from Biotechnical Laboratory of Beijing Dongya. The  $\gamma$ -counter (GC-2160) was manufactured by USTC Chuangxin Co., Ltd.

**Whole-Body Scanning.** Before therapy and monthly for 6 mo after therapy, all patients received 740–1,110 MBq (20–30 mCi) of  $^{99m}\text{Tc}$ -labeled methylene diphosphonate intravenously and underwent whole-body scanning 2–4 h afterward using a single-head SPECT camera (DPS33000; ADAC) equipped with low-energy general-resolution collimators. The number of bone metastases was calculated using a modification of previously described methods (6,10). The skeleton was divided into 4 regions (skull and spine, throat and shoulder, pelvis, and limbs), and in each region the number of foci suggestive of metastases was scored visually and then summed. The bone scans were evaluated independently by 3 nuclear physicians. If they had different opinions, the maximum score was recorded.

**Pain Score Assessment.** Before therapy and monthly for 6 mo after therapy, the pain score was calculated for all patients by multiplying the severity of pain, on a 4-point scale, by its frequency, also on a 4-point scale (Table 1) (4,7,11). Pain severity also included information about the dosage, type of analgesic drugs administered, and any changes in mobility for daily activities (Table 1). The pain score before therapy was compared with the lowest pain score after therapy, and patients whose pain score had decreased by 2 or more were considered responders.

**Hematologic Toxicity.** A hemogram (white blood cell and platelet counts) was obtained for all the patients before therapy and weekly afterward for a month, then monthly for the next 5 mo. Hematologic toxicity was assessed according to criteria listed in the *Manual of Oncologic Therapeutics* (12).

### Statistical Analysis

All data were expressed as mean  $\pm$  SD. The Student  $t$  test was used to compare groups, and a  $P$  value of less than 0.05 was considered to indicate statistical significance. The extent of bone involvement (estimated by whole-body scanning) and the blood levels of IL-2 and TNF- $\alpha$  were examined using the Pearson correlation coefficient.

## RESULTS

No immediate adverse reactions were noted after the  $^{89}\text{SrCl}_2$  injection. Twenty-one percent of patients experienced a mild, transient worsening of symptoms (pain flare phenomenon) that began 1 d after the treatment, lasted 2–4 d, and did not require additional analgesics. The pain flare phenomenon had no correlation with primary tumor type, age, number of bone metastases, or sex. The palliative effect usually started around the second or third week after the injection. No side effects were reported in tissues other than bone marrow after  $^{89}\text{SrCl}_2$  administration, and no patients required a blood transfusion for hematologic depression. A decrease in the initial white blood cell and platelet counts began during the third or fourth week after the injection. Three months after the injection, white blood cells and platelets had decreased in 21% and 27%, respectively, of patients. And later, both white blood cell counts and platelet counts tended to return to their pretherapeutic values, with a gradual partial-to-complete recovery within 6 mo. Grade I toxicity was about 19% for platelets and 16% for white blood cells. Grade II was

**TABLE 1**  
Scoring System for Evaluating Pain

Severity score	Frequency score
0: none (normal mobility or needing no analgesics)	0: none
1: mild (unable to work or needing occasional analgesics)	1: occasional (less than once per day)
2: moderate (mobile at home or not needing opioids)	2: intermittent (at least once per day)
3: severe (in a chair most of the time or needing low-dose opioids)	3: frequent (1–3 times per day)
4: unendurable (in bed more than 80% of the time or needing high-dose opioids)	4: constant

Pain score = severity score  $\times$  frequency score.

TABLE 2

IL-2 and TNF- $\alpha$  Levels in Patients with Bone Metastases and Control Subjects

Group	TNF- $\alpha$ (ng/mL)	IL-2 (ng/mL)
Control subjects ( $n = 20$ )	$1.35 \pm 0.43$	$4.50 \pm 1.37$
Patients with bone metastases ( $n = 42$ )	$2.77 \pm 0.31$	$2.87 \pm 1.23$
$P$	0.001*	0.001*

\* $P < 0.001$  vs. control subjects.

about 8% for platelets and 5% for white blood cells. No patients experienced severe toxicity (grade III or IV).

The serum IL-2 concentrations of all 42 patients were lower than those of the control subjects, whereas TNF- $\alpha$  concentrations were higher in the patients (Table 2). After  $^{89}\text{SrCl}_2$  therapy, serum concentrations of IL-2 increased, peaking at the end of the fourth month and then gradually decreasing over the following 2 mo. In contrast, serum concentrations of TNF- $\alpha$  decreased after the therapy, reaching a minimum at the end of the fourth month and then gradually recovering to baseline values over the following 2 mo. A good inverse correlation was found between IL-2 levels and the number of metastatic lesions and pain score, and a direct correlation was found between TNF- $\alpha$  levels and the number of metastatic lesions and pain score (Table 3).

We categorized each patient as a responder or a nonresponder according to pain score. No significant differences in age, sex, or type of primary tumor were found between responders and nonresponders (Table 4). In responders, serum IL-2 concentrations increased from  $2.91 \pm 0.56$  to  $4.58 \pm 1.14$  ( $P < 0.001$ ), whereas serum TNF- $\alpha$  concentrations decreased from  $2.78 \pm 0.37$  to  $1.24 \pm 0.55$  ( $P < 0.001$ ). In contrast, no significant changes in IL-2 and TNF- $\alpha$  concentrations were observed in nonresponders ( $P = 0.565$  and  $0.542$ , respectively). The differences in serum IL-2 and TNF- $\alpha$  concentrations between responders and nonresponders were not obvious before therapy ( $P = 0.519$  and  $0.178$ , respectively) but

TABLE 4

Responder and Nonresponder Distribution by Age, Sex, Number of Bone Tumors, and Primary Tumor Type

Parameter	Responders ( $n = 33$ )	Nonresponders ( $n = 9$ )
Age (y)	$53 \pm 20.3$	$58 \pm 21.2^*$
Male	19	5
Female	14	4
Primary tumor type		
Lung	11	3
Breast	8	2
Prostate	7	1
Kidney	2	1
Stomach/colon	1	1
Melanoma	1	
Other	3	1
No. of bone metastases	$11.89 \pm 4.81$	$13.15 \pm 5.74^*$

\* $P > 0.05$  vs. responders.

became statistically significant after therapy ( $P < 0.001$ ) (Table 5).

Using a fall in TNF- $\alpha$  levels or a rise in IL-2 levels of greater than 25%, compared with the baseline value, we found the sensitivity and specificity of IL-2 increase to be 87.9% and 77.8%, respectively, and the sensitivity and specificity of TNF- $\alpha$  decrease to be 81.8% and 66.7%, respectively (Tables 6 and 7). Patients with an increasing IL-2 level and a decreasing TNF- $\alpha$  level had a better prognosis.

## DISCUSSION

IL-2 is a cytokine released from T helper lymphocytes. It promotes the generation, proliferation, and differentiation of T lymphocytes; enhances the activity of natural killer cells; induces the generation of lymphokine-activated killer cells; and promotes the production of antibodies by B lymphocytes. Through these mechanisms, it plays an important role in antitumor immune responses (13). Pre-treatment serum IL-2 levels have been shown to be of

TABLE 3

IL-2 and TNF- $\alpha$  Levels, Number of Metastases, and Pain Score After  $^{89}\text{SrCl}_2$  Therapy

Parameter	Before therapy	After therapy					
		1 mo	2 mo	3 mo	4 mo	5 mo	6 mo
IL-2 (ng/mL)	$2.87 \pm 1.23$	$2.98 \pm 0.54$	$3.14 \pm 0.81$	$3.53 \pm 1.01$	$4.13 \pm 1.12$	$3.35 \pm 0.96$	$2.94 \pm 1.04$
TNF- $\alpha$ (ng/mL)	$2.77 \pm 0.76$	$2.53 \pm 0.14$	$2.01 \pm 0.42$	$1.86 \pm 0.21$	$1.49 \pm 0.47$	$2.05 \pm 0.26$	$2.76 \pm 0.23$
No. of metastases	$12.16 \pm 4.58$	$11.92 \pm 4.12$	$10.87 \pm 4.05$	$9.63 \pm 3.11$	$9.05 \pm 3.29$	$9.72 \pm 4.00$	$10.39 \pm 3.88$
Pain score	$8.24 \pm 3.71$	$6.35 \pm 3.04$	$5.84 \pm 2.16$	$4.20 \pm 3.87$	$3.99 \pm 2.15$	$4.01 \pm 3.64$	$4.73 \pm 2.81$

$r = -0.91$  for IL-2 lesions,  $-0.87$  for IL-2 pain score,  $0.78$  for TNF- $\alpha$  lesions, and  $0.62$  for TNF- $\alpha$  pain score.

**TABLE 5**  
IL-2 and TNF- $\alpha$  Levels in Responders and Nonresponders Before and After  $^{89}\text{SrCl}_2$  Therapy

Group	Before therapy		4 mo after therapy	
	IL-2 (ng/mL)	TNF- $\alpha$ (ng/mL)	IL-2 (ng/mL)	TNF- $\alpha$ (ng/mL)
Responders ( $n = 33$ )	$2.91 \pm 0.56$	$2.78 \pm 0.37$	$4.58 \pm 1.14$	$1.24 \pm 0.55$
Nonresponders ( $n = 9$ )	$2.72 \pm 0.54$	$2.73 \pm 0.45$	$2.48 \pm 1.16$	$2.41 \pm 0.58$
$P$	0.345*	0.745*	<0.001†	<0.001†

\* $P > 0.05$  vs. responders.

† $P < 0.001$  vs. responders.

independent prognostic value in patients with advanced non-small cell lung cancer (14). A study by Fischer et al. also indicated that IL-2 secretion correlates with long-term survival in patients with small cell lung cancer (15).

TNF- $\alpha$ , another important cytokine in anticancer therapy, is produced primarily by mononuclear macrophages. TNF- $\alpha$  can initiate an intensive immunoinflammation response, induce natural killer cells and macrophagocytes, and produce carcinolysis (16). TNF- $\alpha$  also can damage vascular endotheliocytes and cause thrombosis or hemorrhage, resulting in tumor necrosis or resolution (17), and inhibit tumor cell proliferation by inducing cell apoptosis (18). Most patients with malignant tumors express high levels of TNF- $\alpha$ , and serum concentrations of TNF- $\alpha$  have shown a correlation with tumor burden and progression (19). Increases in TNF- $\alpha$  probably originate from auto-secretion by tumor cells, tumor-infiltrating lymphocytes stimulated by tumor antigen, and circulating monocytes activated by tumor metastases (20). This possibility would be in line with recent findings that, although appropriate concentrations of TNF- $\alpha$  in serum increase immunologic response and inhibit the development of tumors, high levels of TNF- $\alpha$  can paradoxically promote the ability of a tumor to become aggressive and metastasize. TNF- $\alpha$  has been shown to increase adhesion between tumor cells and endotheliocytes, accelerate maturation of the tumor matrix, and increase the gene expression of stromal metalloproteinase (19,21).

The 79% response rate found for  $^{89}\text{SrCl}_2$  by our study is similar to response rates found by other studies (4,22). The measured cytokines fluctuated after  $^{89}\text{SrCl}_2$  therapy, presumably because  $^{89}\text{SrCl}_2$  therapy improves immunologic function by killing tumor cells. After 4 mo, we found

cytokine levels reflecting the pretherapeutic findings; perhaps responding patients should receive a second dose then.

At present, the pain score and the number of metastatic lesions seen on a whole-body bone scan are commonly used to evaluate the effectiveness of radiopharmaceuticals. Although palliation of pain from bone metastases can reflect the status of tumors to some extent, the pain score is influenced by many other factors, including the patient's age, threshold of pain, mentality, and use of analgesics (23). To be meaningful, the pain score must be modified according to analgesic consumption and daily activities. Bone scintigraphy is not an optimal method of following tumor response, because patients who are in end-stage disease and physically weakened can find it difficult to undergo periodic whole-body scanning. Whether bone metastases worsen and spread or the bone begins to repair, the bone scan may show a greater intensity of focal uptake.

In the present study, we found that serum IL-2 and TNF- $\alpha$  levels correlated well with the number of bone metastatic lesions and pain score. Differences between pretherapeutic and posttherapeutic levels of IL-2 and TNF- $\alpha$  were significant in responders but not in nonresponders. In responders, these variables did not differ before  $^{89}\text{SrCl}_2$  therapy but differed significantly after therapy. These results demonstrate that serum concentrations of IL-2 and TNF- $\alpha$  are a useful indicator of the response to  $^{89}\text{SrCl}_2$  therapy in patients with bone metastases.

In summary,  $^{89}\text{SrCl}_2$  therapy improves levels of measured cytokines, presumably by killing bone metastases. Repeating therapy properly might help maintain relatively normal immunity and increase the chance of survival, although no study has yet shown  $^{89}\text{SrCl}_2$  therapy to increase survival. The combination of TNF- $\alpha$  and IL-2

**TABLE 6**  
Sensitivity and Specificity of an Increase in IL-2

Change in IL-2 level	Responders		Nonresponders		Total	
	$n$	%	$n$	%	$n$	%
Decrease	29	87.9	2	22.2	31	73.8
Increase	4	12.1	7	77.8	11	26.2
Total	33	100	9	100	42	100

**TABLE 7**  
Sensitivity and Specificity of a Decrease in TNF- $\alpha$

Change in TNF- $\alpha$ level	Responders		Nonresponders		Total	
	<i>n</i>	%	<i>n</i>	%	<i>n</i>	%
Decrease	27	81.8	3	33.3	30	71.4
Increase	6	18.2	6	66.7	12	28.6
Total	33	100	9	100	42	100

measurement and pain scoring may help in monitoring the therapeutic effects of  $^{89}\text{SrCl}_2$ .

## ACKNOWLEDGMENTS

We thank Li Chen, Changjiu Zhao, Weimin Li, and Jin Zhou for their policy assistance in performing the study. We also thank Dr. Edward Silberstein, a reviewer of the manuscript, for his extensive work in helping to rewrite it. This study was supported by the Institution of Science and Technology in Heilongjiang Province.

## REFERENCES

- Nielsen OS, Munro AJ, Tannock IF. Bone metastases: pathophysiology and management policy. *J Clin Oncol*. 1991;9:509–524.
- Campa JA III, Payne R. The management of intractable bone pain: a clinician's perspective. *Semin Nucl Med*. 1992;22:3–10.
- Pecher C. Biological investigations with radioactive calcium and strontium: preliminary report on the use of radioactive strontium in treatment of metastatic bone cancer. *U Cal Public Pharmacol*. 1942;2:117–149.
- Pons F, Herranz R, Garcia A, et al. Strontium-89 for palliation of pain from bone metastases in patients with prostate and breast cancer. *Eur J Nucl Med*. 1997;24:1210–1214.
- Kraeber-Bodéré F, Campion L, Rousseau C, et al. Treatment of bone metastases of prostate cancer with strontium-89 chloride: efficacy in relation to the degree of bone involvement. *Eur J Nucl Med*. 2000;27:1487–1493.
- Kasalický J, Krajská V. The effect of repeated strontium-89 chloride therapy on bone pain palliation in patients with skeletal cancer metastases. *Eur J Nucl Med*. 1998;25:1362–1367.
- Dafermou A, Colamussi P, Giganti M, et al. A multicentre observational study of radionuclide therapy in patients with painful bone metastases of prostate cancer. *Eur J Nucl Med*. 2001;28:788–798.
- Pardoll D. T cells take aim at cancer. *Proc Natl Acad Sci U S A*. 2002;99:15840–15842.
- Li Y, Fang N. The changes of the immunological indexes in the bone metastases patients treated with  $^{89}\text{SrCl}_2$  [in Chinese]. *Chin J Nucl Med*. 2004;24:220–221.
- Black GM, Zivanovic MA, McEwan AJ, et al. Sr-89 therapy: strontium kinetics in disseminated carcinoma of the prostate. *Eur J Nucl Med*. 1986;12:447–454.
- Lin XT. Assessment of radionuclide therapy on the patients with painful bone metastases [in Chinese]. *Chin J Nucl Med*. 1995;15:123.
- Wittes R. *Manual of Oncologic Therapeutics*. Philadelphia, PA: Lippincott; 1989:1–318.
- Gong FL. *Medical Immunology*. 1st ed. Beijing, China: Science Publication; 2003:238–252.
- Orditura M, Romano C, De Vita F, et al. Behaviour of interleukin-2 serum levels in advanced non-small cell lung cancer patients: relationship with response to therapy and survival. *Cancer Immunol Immunother*. 2000;49:530–536.
- Fischer JR, Schindel M, Bulzebruck H, et al. Long-term survival in small cell lung cancer patients is correlated with high interleukin-2 secretion at diagnosis. *J Cancer Res Clin Oncol*. 2000;126:730–733.
- Logan TF, Gooding WE, Whiteside TL, et al. Biological response modulation by tumor necrosis factor alpha (TNF-alpha) in a phase Ib trial in cancer patients. *J Immunother*. 1997;20:387–398.
- Nooijen PT, Manusama ER, Eggermont AM, et al. Synergistic effects of TNF-alpha and melphalan in an isolated limb perfusion model of rat sarcoma: a histopathological, immunohistochemical and electron microscopical study. *Br J Cancer*. 1996;74:1908–1915.
- Wong GH, Kaspar RL, Zweiger G, et al. Strategies for manipulating apoptosis for cancer therapy with tumor necrosis factor and lymphotoxin. *J Cell Biochem*. 1996;60:56–60.
- De Vita F, Orditura M, Infusino S, et al. Increased serum levels of tumor necrosis factor-alpha are correlated to soluble intercellular adhesion molecule-1 concentrations in non-small cell lung cancer patients. *Int J Mol Med*. 1998;1:605–608.
- Wu S, Boyer CM, Whitaker RS, et al. Tumor necrosis factor alpha as an autocrine and paracrine growth factor for ovarian cancer: monokine induction of tumor cell proliferation and tumor necrosis factor alpha expression. *Cancer Res*. 1993;53:1939–1944.
- Hafner M, Orosz P, Kruger A, et al. TNF promotes metastases by impairing natural killer cell activity. *Int J Cancer*. 1996;66:388–392.
- Silberstein EB. Advances in our understanding of the treatment of painful bone metastases. *J Nucl Med*. 2000;41:655–657.
- Li Y, Xu WG, Ma D, Xu YS. The evaluation of the therapeutic effects of  $^{89}\text{SrCl}_2$  and  $^{153}\text{Sm-EDTMP}$  with radionuclide imaging [in Chinese]. *Chin J Biologicals*. 2002;15:374–375.