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Applied research of detection on the bone marrow cell morphology combined with the serum as CysC and Cr for the diagnose on the early renal injury in multiple myeloma

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Abstract

Our study aims to discuss the expression and clinical significance of the bone marrow cell morphology combined with the serum level of CysC and Cr of the patients with the early renal injury in multiple myeloma. Patients with the early renal injury in multiple myeloma was analyzed retrospectively. 80 cases of the clinical materials were divided into clinical albuminuria group, no-nephropathy group, trace albumin group and normal albuminuria group according to the degree of renal injury. It could be further divided into unfavorable prognosis group and favorable prognosis group according to prognosis. Meanwhile, 20 cases of healthy volunteers receiving physical examination department were included as a control group. The level of CysC and Cr was detected before treatment, 7th day and 30th day after treatment. At the same time, bone marrow morphology was assessed. The level of CysC and Cr in patients with the early renal injury in multiple myeloma was significantly higher than control group before treatment, at 7th day and 30th day after treatment (P<0.05). Their level was significantly reduced at 7th and 30th day after treatment compared with that before treatment with more obvious reduction at the 30th day after treatment. CysC and Cr level in favorable prognosis group was significantly lower than that in unfavorable prognosis group (P<0.05). In conclusion, the combined detection of CysC and Cr level in serum and bone marrow morphology is beneficial for the diagnosis, treatment and prognosis of the early renal injury in myeloma.

Key words: early renal injury in multiple myeloma; CysC; Cr; serum; prognostic.

Introduction

The multiple myeloma is one kind of progressive tumor. Its morphological characteristics are diversified. It has been the main reason to threaten the human life and health seriously. The misdiagnosis and mistreatment are common which could postpone the treatment. The prognosis is poor. There are multiple organs and systems involved in the myeloma. There is intimate connection between the morbidity of the myeloma and the intracellular calcium overload, neutrophil infiltration, increase of inflammatory factor, abnormality of energy metabolism, damage and apoptosis of endothelial cells [1]. The myeloma was malignant proliferated disease. The renal damage was the most obvious. The main clinical feature is the renal insufficiency in multiple myeloma. How to ameliorate the renal damage has been the new challenge on the clinical treatment.

The exact molecular pathogenesis of the early renal injury in multiple myeloma remains unclear [2]. The early renal damage is the main complication for the multiple myeloma. There was important effect of screening on the early detection markers for the treatment and prognosis of this disease. At the present, morphological characteristics of myeloma cells are diversified. The detection on the morphology of myeloma cells was adopted for auxiliary diagnosis. But detection on the morphology of myeloma cells was not applied for the diagnosis on the myeloma along with the development of research. The sensitivity and specificity of detected results was in large difference [3]. The detection on the concentration of the Cr in serum for the assessment on the early renal injury in multiple myeloma was adopted clinically. It was adopted to judge the degree of renal injury. But it was influenced by the renal

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function, renal discharge capacity and ingestion level. Therefore, the sensitivity and specificity are influenced [4]. There was relation between CysC level and the morbidity and development, invasion and metastasis of tumor from existing study. The CysC is degraded and absorbed at nephric tubule completely and could pass through the glomerular filtration membrane. At the present, it was also considered as the important indicator for the glomerular filtration function. It could develop important indicative function on the diagnosis of the early renal injury in multiple myeloma [5]. The best opportunity for recovering the renal function is in three months from the morbidity of myeloma. So early discovery for the renal damage was the key for the prevention and treatment of the complications in multiple myeloma. The renal function of patients could be improved efficiently if the multiple myeloma was diagnosed early and effective therapeutic measures were adopted. In our study, CysC and Cr levels were combined with the bone marrow morphology to assess their role in the diagnosis and treatment of early renal injury in multiple myeloma.

Material and Method

Research Object

The 80 cases of clinical materials of patients with the early renal injury in multiple myeloma from May of 2018 to March of 2020 in our hospital was analyzed retrospectively. Our study was approved by Ethic Committee in our hospital. Then our study was carried out. There were thirty-two cases of women and forty-eight of men. The average age of patients was 59.77±10.48. The age of patients was forty-five to seventy-two. It was divided according to the severity of early renal damage. There were 20 cases in no-nephropathy group, 20 cases in clinical albuminuria group, 20 cases in normal albuminuria group, 20 cased in trace albumin group. The informed consent was signed by patients. It was divided into unfavorable prognosis group and favorable prognosis group according to the condition of prognosis for the early renal injury in multiple myeloma. At the same time, the 20 cases of healthy volunteers underwent physical examination were set as control group. There were eight cases of women and twelve cases of men. The average of age was 59.37±9.46. The range of age was thirty-eight to eighty. There was no statistical significance of general data including age or gender between these groups (P>0.05) (table 1)

The included criterion: (1) the content of our study was informed to the patients; (2) The patients participated in this research willingly. And there was no other serious complicating disease. (3) It was conformed to the diagnosis and treatment guideline of multiple myeloma in China. (4) The informed consent was signed by patients. The imageology data was integrated. (5) The increase of bone marrow plasmacyte through tissue biopsy was more than ten percent. There was plasmacytoma. The twenty cases of healthy volunteers included in the check for the physical examination department were set as control group. The included criterion: (1) There was no other serious complicating disease. (2) The volunteers participated in this research willingly. (3) There was no abnormality of renal function.

The exclusion criteria: (1) there was accompanied with rheumatism systemic disease, chronic tuberculosis infection, and increase of abnormal plasmocyte. (2) There was serious osteoporosis and hypophosphorus osteopathy. (3) The collected materials were not integrated. (4) There was accompanied with metastatic carcinoma and serious diseases on the other organs.

Therapeutic method

The Dexamethasone Acetate Tablets were purchased from Lishen Company of Tianjin. (Batch number: SFDA approval number H12020122, specification: 0.75 mg). It was orally administered at the 1st to 4th day, the 9th to 12th day, and the 17th to 20th day. The dosage was twelve milligram in one day. The VAD chemotherapy project was adopted. The intravenous drip was adopted. The Pirarubicin was purchased from WanLe Company of Shenzhen. (Batch number: SFDA approval number H20045983, specification: 10mg). The dosage was ten milligram in one day. It was orally administered at the 1th to 4th day. The Vincristine Sulfate for Injection was purchased from Haizheng of Zhejiang. (Batch number: SFDA approval number H20043326, specification: 1mg). The dosage was 0.4 milligram in one day. The intravenous drip was adopted at the 1th to 4th day.

Main reagent and apparatus

The kit of SYBR Premix Ex TaqTM Ⅱ was purchased from Omega Company. The RNA Extraction Kit was purchased from Takara Company. The RNeasy Mini Kit and miRNA cDNA Synthesis kit was purchased from Thermo Scientific Company. The QPCR ABI7900 apparatus was from Applied Biosystem Company. The Nano Drop2000 micro-spectrophotometer was purchased from Oiagen Company.

Detection of serological sample

Preparation of serum

3 mL peripheral venous blood with an empty belly was extracted at the three time point as before treatment, the 7th day after treatment, the 30th day after treatment. Then it was put into anticoagulant tube. The collected whole blood was transferred into individual deposited tube. It was reserved in liquid nitrogen chronically. The serum was collected after ten minutes of centrifugation at 3000rpm. It was reserved in refrigerator at below seventy degree temporarily. The total RNA was extracted according to the specification of RNeasy Mini Kit. The RNA samples were detected by Nano Drop2000 micro-spectrophotometer quantitatively and qualitatively. The RNA sample conformed to the criteria was put into refrigerator at below eighty degree. The RNA need be extracted again if the RNA D260/D280 was less than 2.0.

Expression of CysC and Cr in serum detected by QPCR

The 1 ug of sample was collected at the time point of before treatment, the 7th day after treatment, the 30th day after treatment. And it was reserved at four degree temporally. The RNA was extracted from the serum of patients with multiple myeloma. The reverse transcription was performed according to the specification of Synthesis miRNA cDNA kit. The reaction condition was five seconds at eighty-five degree, thirty minutes at thirty-seven degree. The primer was synthesized by Guangzhou Fuxiankang Biotechnology Limited Company:

Designed primer sequences:

CysC-F：5’
ACACTCCAGCTGGGTAGCTTATCAGACTGATG
Cr-F：5’ ACACTCCAGCTGGGTAGCACCATCTGAAATC
U6-F：5’ CTCGCTTCGGCAGCACA
U6-R：5’ AACGCTTCACGAATTTGCGTd

U6 was selected as a control. The expression level of CysC and Cr in all samples was detected. The reverse-transcribed product was obtained. The reaction of reverse transcription system was performed by real-time fluorescence quantification PCR (QPCR) apparatus according to the procedure of specification of SYBR Premix Ex Taq Ⅱ TM kit. The total volume was 20 ul. The reaction system: five microliter of cDNA, 0.5 microliter of upstream primer, 0.5 microliter of downstream primer, four microliters of dH_2O, ten microliters of 2x SYBR qPCR Green SuperMix. The five microliters of cDNA were added into RNA. The reaction system: cDNA (diluted at one to twenty), amplified into twenty microliters, decreased up to forty cycles: two minutes at ninety-five degree, fifteen seconds at ninety-five degree, two minutes at fifty degree, reading plate at thirty-two seconds at sixty-degree, sixty degree to ninety-five degree. The analysis on the dissociation curve: the relative expression of miRNA was detected by $2^{-\Delta\Delta Ct}$ method.

Morphological analysis of the bone marrow cells

The three milliliter of bone marrow was extracted through bone marrow puncture. The plate was smeared with cells. Then it was stained with C0135 Reix-jimossa composite dye (Shanghai Beizhuo Biotechnology Limited Company). Then it was classified through microscopic examination under microscope. The cellular morphology was observed. The percentage of the bone marrow cells was calculated.

Statistical method

The SPSS23.0 software was adopted to analyze data. The measure data was represented as mean ± SD and assessed by the One-Way ANOVA among groups or independent t-test between two groups. The Mann-Whitney U test was adopted among the groups of non-normal data. The count data was represented as frequency (percentage). The chi-square test was adopted among groups. $P<0.05$ indicates a significance.

Results

High expression of CysC and Cr in the serum of patients

The results of morphological analysis of the bone marrow cells and detection of qRCR was shown in Table 2 and Figure 1. There was no visible hemorrhage and thanatosis in control group. And there was no obvious change on kidney generally. There was no cytoplasmic vacuole in group without nephrosis. And the structure was normal under light microscope. There was no neutrophil infiltration. There was indistinct boundary of renal cells in normal albuminuria group. And the color was darkened. The hemorrhage and thanatosis was aggravated in trace albuminuria group generally. There were visible progressive cytoplasmic vacuoles in clinical albuminuria group with loss of cell structure. The level of CysC and Cr in the serum of patients with early renal injury of multiple myeloma and the percentage of myeloma cells before treatment was significantly higher than that in control group ($P<0.05$). The percentage of expression quantity of CysC and Cr and myeloma cells before treatment: clinical albuminuria group> trace albuminuria group> group without nephrosis> normal albuminuria group> control group of healthy volunteers. There was statistical significance ($P<0.05$).

Expression of CysC and Cr in patients after treatment

As shown in table 3 and figure 2, there was no obvious change on the nephridial tissue of rats in control group at the 7th day after treatment. There was no visible edema in normal albuminuria group. There was almost no edema under light microscope in group without nephrosis. It was normal basically. And there was no inflammatory cell infiltration. The inflammation of cells was aggravated in trace albuminuria group. There was inflammatory cell infiltration in clinical albuminuria group. And there was progressive enlargement of acinous cells. The expression level of CysC and Cr in the serum of patients with early renal injury of multiple myeloma and the percentage of myeloma cells before treatment was significantly higher than that in control group (P<0.05). The expression quantity at the 7th day after treatment: clinical albuminuria group> trace albuminuria group> group without nephrosis> normal albuminuria group. There was statistical significance between two groups (P<0.05). The expression quantity of CysC and Cr and percentage of myeloma cells in every groups after treatment was significantly higher than that before treatment. There was statistical significance among every groups before and after treatment except the control group at the 7th day after treatment (P>0.05).

Expression of CysC and Cr in patients after 30 days treatment

As seen in table 4 and figure 3, there was no edema and inflammatory cell infiltration in control group at the 30th day after treatment. And the basic structure was normal. There was no destruction and thanatosis in normal albuminuria group. The cells in normal albuminuria group were normal with nearly no inflammatory cell infiltration and edema in group without nephrosis. There was inflammatory cell infiltration in trace albuminuria group. The degree of inflammatory cell infiltration was reduced in clinical albuminuria group after treatment. The expression level of CysC and Cr in the serum of patients with early renal injury of multiple myeloma and the percentage of myeloma cells before treatment was significantly higher than that in control group (P<0.05). There was no obvious difference on the expression quantity at the 30th day after treatment between group without nephrosis and normal albuminuria group (P>0.05). Clinical albuminuria group> trace albuminuria group> group without nephrosis> normal albuminuria group. There was statistical significance among two groups (P<0.05). The expression quantity of CysC and Cr and the percentage of myeloma cells in every groups in the 30th day after treatment compared with the 7th day after treatment was significantly reduced. There was statistical significance among every groups before and after treatment except the control group at the 30th day after treatment (P<0.05).

Role of CycC and Cr level in prognosis

As seen in table 5, the expression level of CysC and Cr in the serum of patients with early renal injury of multiple myeloma and the percentage of myeloma cells in patients with good prognosis after treatment was significantly lower than that in unfavorable prognosis (P<0.05).

Discussion

The occurrence of multiple myeloma is complicated. It is a common disease of blood system. The renal injury was the most nerve complication in multiple myeloma which could influence the prognosis of patients. The renal injury was discovered to be treated early which was very helpful to improve the prognosis of patients [6]. Therefore, the diagnosis of the early multiple myeloma was the key for the treatment. At the present, golden standard is still renal biopsy. The indicator of traditional detection method was hysteretic relatively. But the feasibility for carrying out the puncture for renal biopsy was reduced enormously due to the invasive procedure and limitation of patients' engine body diseases. The diagnostic data could be obtained timely if there was renal injury clinically. So, the operability of inspection on the serology and bone marrow morphology is more convenient. The pathogenic condition could be judged conveniently and relatively. The data was obtained timely. And early prevention could be done. So more definite and more accurate indicator was tried to assess the injury of multiple myeloma [7]. The efficient treatment could be given on the early injury of multiple myeloma [8-10].

At present, the indicator of GFR for assessment of the filtering capability of kidney could be evaluated quantitatively. The accuracy for the assessment on the renal function was higher relatively. And the clinical common indicators including creatinine clearance rate could be adopted to assess the renal function of patients partly. But the urine of patients in twenty-four hours need be collected for detection. And the accuracy need be improved. The operative difficulty and workload are larger relatively [11]. The detection on the ^{51}Cr-EDT radioactive nucleus was important indicator for the assessment on the filtering capability of kidney clinically. But the

procession of detection was accompanied with contamination of radioactive ray. And the price of detection was high. The requirement for the testing equipment was high. Therefore, it could not be carried out in all medical institution [12]. The indicator of the detection on the evaluation index for the injury of multiple myeloma including the detection of Cr was convenient, common and accurate indicator relatively along with the improvement on the sensibility and accuracy of detection technology in clinical research continuously. It could develop certain effect on the judgment of the progression of clinical disease [13]. The indicator was found to be influenced by discharge capacity of kidney and body appetite capacity partly along with the improvement on the study continuously. The real-time, specific and sensitive assessment could not be obtained. There was certain hysteresis effect on results probably [14]. The Cys C is cysteine proteinase and could develop inhibitory effect on the development and progression of multiple tumors, transformation and infiltration reported by literatures. The molecular weight of Cys C is small relatively. It could be filtrated through glomerulus and re-absorpted by glomerulus [15]. The Cys C is the important indicator for the injury of glomerulus from the research in recent years. It could be adopted to assess the degree of injury of glomerulus [16-17]. The kidney was excretory organ of Cys C. So, there was important effect on assessment for the function of kidney and glomerulus. Besides, effect of favorable prediction on the occurrence for disease and prognostic was studied. It could be considered as the important indicator for the prediction on the median survival time of multiple myeloma. It might be adopted to judge the condition of occurrence and development of complications such as renal injury [18]. At the present, effect of CysC and Cr on the prevention and prognosis for the early renal injury in multiple myeloma was still unclear. The research significance for judgment on patient's prognosis was important. But it was absent extremely, especially through detection on the expression of CysC and Cr [19-20]. The bone marrow cell morphology could play a beneficial role in the blood diseases. The role of CysC and Cr in the diagnosis and prognostic evaluation for the serum disease in the early renal injury of multiple myeloma was discussed deeply combined with the detection on the bone marrow cell morphology in our study.

The percentage of myeloma cells and expression level of CysC and Cr in the patients' serum in the early renal injury of multiple myeloma before treatment was significantly higher than in control group from the results of our study. The expression quantity of CysC and Cr and percentage of myeloma cells at the 7th day after treatment, the 30th day after treatment: clinical albuminuria group> trace albuminuria group. The expression quantity of CysC and Cr and percentage of myeloma cells was increased at different degree after multiple kinds of treatment. There was increased tendency over time. The expression of CysC and Cr at the 7th day after treatment was significantly higher than that before the treatment. There was certain fallout on the expression quantity at the 30th day after treatment. It was reduced significantly compared with the 7th day after treatment. It was hinted that there was close relation between the expression level of CysC and Cr and percentage of myeloma cells in the patient's serum with early renal injury of multiple myeloma. But the therapeutic effect at the 7th day after treatment was not observed. The expression of CysC and Cr and percentage of myeloma cells was still in the increased state. The condition of disease was not relieved adequately. The therapeutic effect was observed continuously as time went on. The expression level of CysC and Cr and percentage of myeloma cells was fell back at the 30th day after treatment. The expression of CysC and Cr and percentage of myeloma cells in the serum of patients with the early renal injury of multiple myeloma in the group of good-treated patients was significantly lower than in the group of unfavorable prognoses obviously from the results of our study. It was proved further that the detection on the CysC and Cr and percentage of myeloma cells was favorable indicator on the diagnosis and prognosis of the early renal injury of multiple myeloma. There was certain guiding significance for the expression of CysC and Cr and percentage of myeloma cells in the serum of patients with the early renal injury of multiple myeloma on the prediction and prognosis of the disease.

In conclusion, detection of expression level of CysC and Cr and percentage of myeloma cells maybe be helpful to assess the occurrence, development and prognosis of the early renal injury of multiple myeloma. However, due to limited number of patients included, which is a main study limitation, large cohort clinical samples should be collected to validate the findings in the future.

References

[1] Terpos E, Christoulas D, Kastritis E, Katodritou E, Pouli A, Michalis E, et al. The Chronic Kidney

Disease Epidemiology Collaboration Cystatin C (CKD-EPI-CysC) equation has an independent prognostic value for overall survival in newly diagnosed patients with symptomatic multiple myeloma; is it time to change from MDRD to CKD-EPI-CysC equations? [J]. European journal of haematology. 2013,91(4):347-355.

[2] Kudose S, Suarez-Fuentes C, D'Agati VD, Markowitz GS. The Case | A 53-year-old woman with acute kidney injury and multiple myeloma[J]. Kidney international. 2019,96(4):1045-1046.

[3] Hari P, Reece DE, Randhawa J, Flomenberg N, Howard DS, Badros AZ, et al. Final outcomes of escalated melphalan 280 mg/m (2) with amifostine cytoprotection followed autologous hematopoietic stem cell transplantation for multiple myeloma: high CR and VGPR rates do not translate into improved survival[J]. Bone marrow transplantation. 2019,54(2):293-299.

[4] Shaikh S, Nwankwo C, Lacasse A, Cheng S. Acute kidney injury on chronic kidney disease: from congestive heart failure to light chain deposition disease and cast nephropathy in multiple myeloma[J]. Journal of community hospital internal medicine perspectives. 2019,9(4):319-321.

[5] Nadiminti K, Singh Abbi KK, Mott SL, Dozeman L, Tricot A, Schultz A, et al. VTD-melphalan is well tolerated and results in very high rates of stringent CR and MRD-negative status in multiple myeloma[J]. OncoTargets and therapy. 2017,10:217-226.

[6] Andronesi AG, Tanase AD, Sorohan BM, Craciun OG, Stefan L, Varady Z, et al. Incidence and risk factors for acute kidney injury following autologous stem cell transplantation for multiple myeloma[J]. Cancer medicine. 2019,8(6):3278-3285.

[7] Iacobelli S, de Wreede LC, Schonland S, Bjorkstrand B, Hegenbart U, Gruber A, et al. Impact of CR before and after allogeneic and autologous transplantation in multiple myeloma: results from the EBMT NMAM2000 prospective trial[J]. Bone marrow transplantation. 2015,50(4):505-510.

[8] Lee Y, Goyal K, Prasad B. Acute Kidney Injury in a 31-year-old Male as a Consequence of Multiple Myeloma[J]. Cureus. 2018,10(6): e2881.

[9] Nishihori T, Ochoa-Bayona JL, Kim J, Pidala J, Shain K, Baz R, et al. Allogeneic hematopoietic cell transplantation for consolidation of VGPR or CR for newly diagnosed multiple myeloma[J]. Bone marrow transplantation. 2013,48(9):1179-1184.

[10] Mene P, Giammarioli E, Fofi C, Antolino G, Verde G, Tafuri A, et al. Serum Free Light Chains Removal by HFR Hemodiafiltration in Patients with Multiple Myeloma and Acute Kidney Injury: a Case Series[J]. Kidney & blood pressure research. 2018,43(4):1263-1272.

[11] Wang M, Delasalle K, Feng L, Thomas S, Giralt S, Qazilbash M, et al. CR represents an early index of potential long survival in multiple myeloma[J]. Bone marrow transplantation. 2010,45(3):498-504.

[12] Sprangers B. Aetiology and management of acute kidney injury in multiple myeloma[J]. Nephrology, dialysis, transplantation: official publication of the European Dialysis and Transplant Association - European Renal Association. 2018,33(5):722-724.

[13] Hudier L, Decaux O, Haddj-Elmrabet A, Lino M, Mandart L, Siohan P, et al. Intensive haemodialysis using PMMA dialyser does not increase renal response rate in multiple myeloma patients with acute kidney injury[J]. Clinical kidney journal. 2018,11(2):230-235.

[14] Paule B, Quillard J. [Prognostic and therapeutic impact of the cellular morphology and type of bone marrow invasiveness in multiple myeloma] [J]. Bulletin du cancer. 1988,75(6):567-569.

[15] Premuzic V, Batinic J, Roncevic P, Basic-Jukic N, Nemet D, Jelakovic B. Role of Plasmapheresis in the Management of Acute Kidney Injury in Patients with Multiple Myeloma: Should We Abandon It? [J]. Therapeutic apheresis and dialysis: official peer-reviewed journal of the International Society for Apheresis, the Japanese Society for Apheresis, the Japanese Society for Dialysis Therapy. 2018,22(1):79-86.

[16] Chugh S, Kichloo A, Jafri F, Yusvirazi L, Lerner R. Multiple Myeloma as the Underlying Cause of Thrombotic Microangiopathy Leading to Acute Kidney Injury: Revisiting a Very Rare Entity[J]. Journal of investigative medicine high impact case reports. 2017,5(3):2324709617732797.

[17] Fernandez de Larrea C, Tovar N, Rozman M, Rosinol L, Arostegui JI, Cibeira MT, et al. Multiple myeloma in serologic complete remission after autologous stem cell transplantation: impact of bone marrow plasma cell assessment by conventional morphology on disease progression[J]. Biology of blood and marrow transplantation. 2011,17(7):1084-1087.

[18] Kuriakose P, Das S, Mani A. Bone marrow morphology in multiple myeloma[J]. Indian journal of cancer. 1995,32(3):100-103.

[19] Li Cavoli G, Passanante S, Schillaci O, Servillo F, Zagarrigo C, Li Cavoli TV, et al. Haemodiafiltration with ultrafiltrate regeneration in the removal of free light chains in multiple myeloma and acute kidney injury[J]. Nefrologia: publicacion oficial de la Sociedad Espanola Nefrologia. 2018,38(3):337-338.

[20] Wu D, Wang WJ, Zhang M. Bone Marrow Morphology of Multiple Myeloma with Non-bone-related Extramedullary Disease[J]. Zhongguo shi yan xue ye xue za zhi. 2018,26(3):807-811.

Table and Figure legends

Table 1 **comparison of general materials of the subjects in every groups [n (%)]**

data	Control group（n=20）	no-nephropathy group（n=20）	normal albuminuria group（n=20）	clinical albuminuria group（n=20）	trace albumin group（n=20）	F	P
age（t/a）	58.83±8.34	59.02±8.95	59.94±9.16	59.04±8.97	59.34±9.18	0.149	0.867
gender							
men	11（55.0）	11（55.0）	12（60.0）	11（55.0）	12（60.0）	0.210	0.894
women	9（45.0）	9（45.0）	8（40.0）	9（45.0）	8（40.0）		
complications							
Ostealgia and pathological fracture	14（70.0）	10（50.0）	12（60.0）	10（50.0）	12（60.0）	1.145	0.567
Anemia and bleeding tendency	9（45.0）	12（60.0）	11（55.0）	12（60.0）	11（55.0）	1.327	0.544
nervous system injury	10（50.0）	9（45.0）	13（65.0）	9（45.0）	13（65.0）	0.197	0.906

*P<0.05vs control group；△P<0.05vs normal albuminuria group；※P<0.05 vs no-nephropathy group；°P<0.05 vs clinical albuminuria group.

Table 2. **expression of CysC and Cr in the serum of patients with early renal injury of multiple myeloma and morphological analysis on the myeloma cells before treatment**

Groups	Control group（n=20）	group without nephrosis（n=20）	normal albuminuria group（n=20）	clinical albuminuria group（n=20）	trace albuminuria group（n=20）	F	P
CysC（mg/L）	1.372±0.253 △※°	4.264±0.884*△°	3.343±1.696 *※°	7.327±1.644*△ ※°	6.424±1.347*△ ※	3.017	0.037
Cr（mg/L）	2.385±0.673 △※°	4.324±1.037*△°	3.363±1.675 *※°	7.987±1.264*△ ※°	6.584±2.164*△ ※	3.394	0.033
percentage of myeloma cells（%）	5.646±0.646 △※°	12.367±3.056* △°	8.356±1.543 *※°	27.987±7.254* △※°	21.554±5.154* △※	4.341	0.021

*P<0.05vs control group；△P<0.05vs normal albuminuria group；※P<0.05 vs group without nephrosis；°P<0.05 vs clinical albuminuria group. One-way ANOVA analysis.

Table 3. **expression of CysC and Cr and morphology of myeloma cells in the serum of patients with early renal injury of multiple myeloma at the 7th day after treatment**

Groups	Control group（n=20）	group without nephrosis（n=20）	normal albuminuria group（n=20）	clinical albuminuria group（n=20）	trace albuminuria group（n=20）	F	P
CysC(mg/L)	1.693±0.376 △※○	5.445±0.877*△○	3.576±0.474 *※○	7.637±1.534*△※○	8.587±1.245*△※	3.764	0.032
Cr(mg/L)	1.383±0.533 △※○	5.525±0.314*△○	3.546±0.375 *※○	7.843±0.352*△※○	8.267±1.364*△※	3.596	0.037
percentage of myeloma cells（%）	2.384±0.545 △※○	9.584±2.678*△○	5.657±1.757 *※○	24.854±5.345*△※○	18.254±3.578*△※	4.201	0.022

*P<0.05vs control group；△P<0.05vs normal albuminuria group；※P<0.05 vs group without nephrosis；○P<0.05 vs clinical albuminuria group. One-way ANOVA analysis.

Table 4. **expression of CysC and Cr and morphology of myeloma cells in the serum of patients with early renal injury of multiple myeloma at the 30th day after treatment**

Groups	Control group（n=20）	group without nephrosis（n=20）	normal albuminuria group（n=20）	clinical albuminuria group（n=20）	trace albuminuria group（n=20）	F	P
CysC(mg/L)	1.493±0.374 △※○	3.272±0.455 *※○	5.052±0.845*△○	8.032±1.265*△※	6.843±1.243*△※○	3.677	0.034
Cr(mg/L)	1.183±0.545 △※○	3.032±0.355 *※○	4.546±0.543*△○	7.663±1.546*△※	6.546±0.356*△※○	3.426	0.039
Percentage of myeloma cells (%)	2.354±0.554 △※○	4.456±1.245 *※○	7.554±1.325*△○	13.254±2.354*△※	17.854±4.452*△※○	3.974	0.028

*P<0.05vs control group；△P<0.05vs normal albuminuria group；※P<0.05 vs group without nephrosis；○P<0.05 vs clinical albuminuria group. One-way ANOVA analysis.

Table 5. **expression condition of prognosis estimation in CycC and Cr in the serum of patients with early renal injury of multiple myeloma after treatment**

Groups	Group with favorable prognosis(n=35)	Group with unfavorable prognosis (n=28)	t	P
CysC (mg/L)	3.493±1.236	6.484±2.155*	3.944	0.022
Cr (mg/L)	3.904±1.275	6.954±1.856*	3.745	0.036
Percentage of myeloma cells (%)	10.454±3.873	26.212±4.787*	4.246	0.024

*P<0.05vs group of unfavorable prognoses.

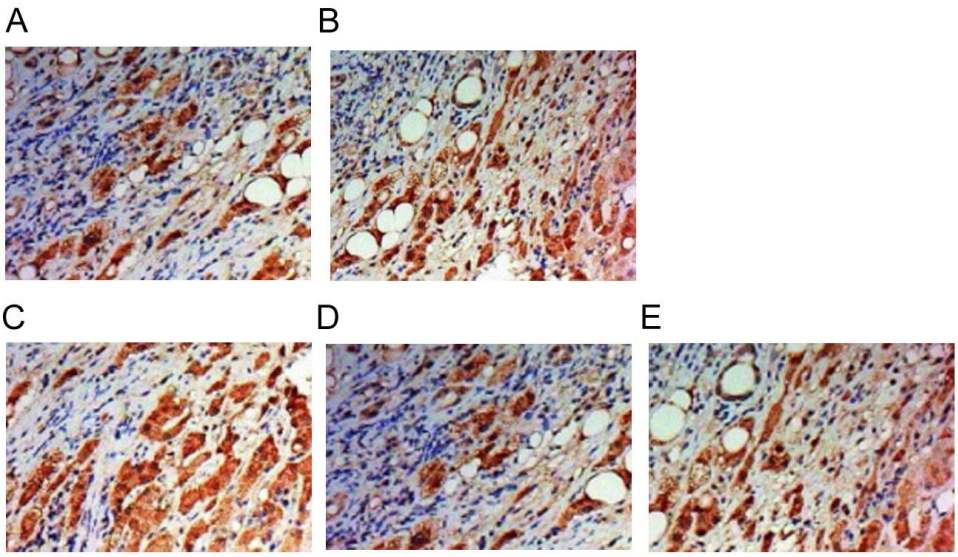

Figure 1**. morphology of myeloma cells in every groups before treatment**
Control (A) normal albuminuria group;(B) group without nephrosis; (C) trace albuminuria group;(D) clinical albuminuria group; (E)control group (×200).

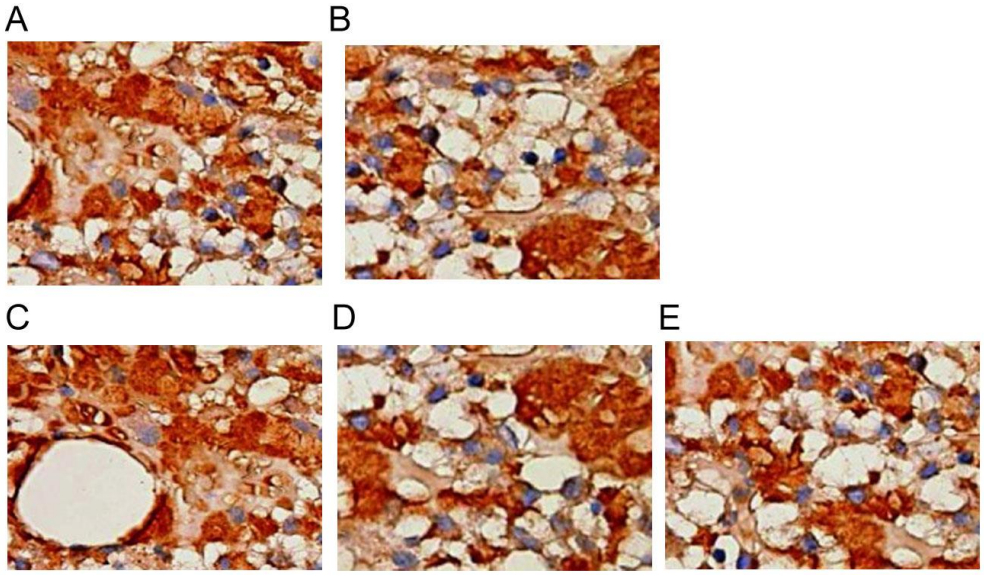

Figure 2. **morphology of myeloma cells in every groups at the 7th day after treatment**
Control (A) normal albuminuria group;(B) group without nephrosis; (C) trace albuminuria group;(D) clinical albuminuria group; (E)control group (×200).

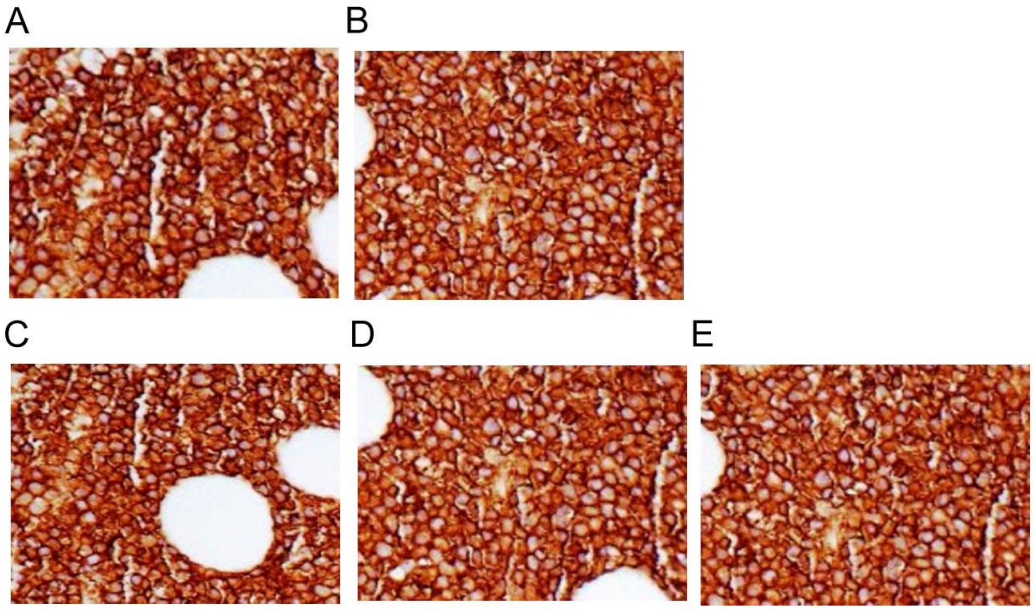

Figure 3. **morphology of myeloma cells in every groups at the 30th day after treatment**
Control (A) normal albuminuria group; (B) group without nephrosis; (C) trace albuminuria group; (D) clinical albuminuria group; (E)control group (×200).