Serum changes of PYD, C2C and OC among male brucellosis patients at early period

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Research Article

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Abstract

Objective

Musculoskeletal changes are the most common clinic manifestation of brucellosis,and these musculoskeletal changes are irreversible, so, it is very important to prevention musculoskeletal changes at early stage of human brucellosis .

Method

This was a case-control study. According to diagnostic criteria of human brucellosis in China(WS269-2007),41 male patients were diagnosed as brucellosis patients at early period (within 6 months) and they were not therapied with drug, 44 \ persons were divided into control group with randomly matching. Venous blood samples were collected from all study subjects, and serum PYD C2C and OC were quantitative measured with method of ELISA. Data were analyzed using SPSS 17.0 software. A *p* value < 0.05 was considered to be statistically significance.

Result

The median levels of serum PYD, C2C and OC in the patients group were 278.53 ug/l, 82.23 ug/l, and 8.41ug/l, respectively, while the median levels of serum PYD, C2C and OC in the control group were 210.54 ug/l, 72.74 ug/l and 7.43 ug/l, respectively, there were existing significant differences between patients group and control group (Z = 5.686, 3.997, 3.579, all *P* = 0.000).

Conclusion

Serum levels of PYD, C2C and OC were increased among male brucellosis patients at early period, which might be the indicator biomarkers for osteoarticular changes of human brucellosis at early stage..

Introduction

Brucellosis is a zoonotic disease with worldwide distribution. This disease hinders livestock productivity severely, and remains a major public health problem in mediterranean region, middle East, africa, latin america, and parts of asia 1]. In China, brucellosis have been reported in all 32 provinces, and human brucellosis is endemic in 25 of 32 provinces (or autonomous regions) of mainland China [2]. Brucellosisl poses a serious threat to public health [3].

Brucellosis is transmitted to people by direct or indirect contacting with infected animals or consumption of contaminated foods [4]. Clinical presentation of this disease is nonspecific and highly variable, including in fever, headache, chills, myalgia, and arthralgia [5]. Although several organs and organ

systems may be involved in this disease, musculoskeletal changes are the most common clinical presentations, such as sacroiliitis, spondilitis, peripheral arthritis, osteomyelitis, discitis, bursitis and tenosynovitis [6]. These musculoskeletal changes can lead patients to initially visit general practitioners, and ultimately rheumatology and or orthopedic specialists. Because of variable clinical features and lack of specific symptoms, it may delay diagnosis of musculoskeletal changes and relaive drug therapy [7]. Due to musculoskeletal changes irreversible, so, it is very necessary to prevent musculoskeletal changes of human brucellosis at the early period.

Pyridinoline (PYD) □ type II collagen cleavage neoepitope (C2C) and Osteocalcin (OC) are often used as biomarkers for assessing osteoarthritis (OA). PYD has been validated as useful markers for bone resorption and their levels aresignificantly higher in OA patients [8].The level of C2C may reflect the extent of cartilage degradation in OA patients [9]. OC has routinely been used as reflecting of osteoblastic bone formation and regulating mineralization in the bone matrix [10].

We hypothesized that there were existing abnormal biological changes of musculoskeletal changes among brucellosis patients at early period, while musculoskeletal changes weren't found by radiography.Thus,we can find an effective methds for prevention musculoskeletal changes of human brucellosis at early period.

Only few studies have been related to biomarkers of musculoskeletal changes among human brucellosis. We had found abnormal changes of serum cartilage oligomeric matrix protein (COMP) and C-terminal telopeptide of type II collagen (CTX-II) among brucellosis patients firstly [11,12]. So, we found that it was meaning for expanding biomarkers of musculoskeletal changes to probe the mechanism of musculoskeletal changes among human brucellosis. This was about study of serum PYD, C2C and OC levels in human brucellosis for the first time. So, the aim of this study was to find biomarkers for the musculoskeletal changes of male brucellosis patientsat at early period, and provide a better understanding of this disease.

Materials And Methods

Study subjects

This was a case-control study. After clinical examination, serum test and X-ray image examination at hospital, all the study population were excluded normal joint diseases, such as rheumatic fever, osteoarthritis, rheumatic arthritis, paratyphoid fever, and tuberculosis which these joint diseases were similar to human brucellosis at the clinical manifestation. The clinical changes of some of brucellosis patients were fever, weak and headache, some of brucellosis patients have nothing clinical changes, however, due to special occupation and living environment, these study population were checked in Qinghai institute for endemic disease prevention and control from 2017 and 2018 again. Based on epidemiological investiagtion, clinical examination, serological detecting □ 41 male patients were all diagnosed within six month, and were not experienced drug therapy, because some of patients did not definite accuracy infectious time, so we defined the acute patients and sub-acute patients as early period

of brucellosis. Through matching, 44 male control were divided into control group with randomly sampling, whose serum test of RBPT and SAT were negative. The male patients group and the male control group had simliar production or living environment.

Serum samples were collected from study subjects until assayed. Serum levels of PYD, C2C and OC were detected with the method of Enzyme-linked immunosorbent assay (ELISA), ELISA kit were purchased from AIRCo. Ltd (Beijing).

Diagnostic criteria of brucellosis in China

Based on diagnostic criteria of brucellosis in China（WS269–2007）the brucellosis patients are individed into three types, including acute period (within three months after infection, high serum titre and high fever), sub - acute period (among 3–6 months, positive serum titre, and low fever) and chronic period (above six month after infection, positive serum titre, and normal body temperature); the main clinical manifestations are fever, weak, headache, hyperhidrosis and ache of joint and muscle; the serological tests are rose bengal plate test (RBPT, a primary screen test), serum agglutination test (SAT, the titre ≥1:100), and SAT would be excluded vaccinated..

Statistical analysis

Data were analyzed using SPSS 17.0 software（SPSS, Chicago.IL, USA）. The means and standard deviations were calculated for age, which were anlayzed by t test. The data of PYD, C2C and OC were belonged to skewed distribution after analyzing, so, these data werer analyzed with the nonparametric test (Wilcon rank sum test). A p value < 0.05 was considered to be statistically significance.

Results

The average age of brucellosis patients was (39.69±9.98) years, meanwhile that of the controls group was (42.07±13.70) years, there were not a significant difference in age comparison among two groups (t = 0.912, *P* = 0.364). 41 male brucellosis patients were RBPT positive, and SAT above ≥ 1:100, while the results of RBPT and SAT from control group were all negative. Table 1.

Table 1.The general condition of objective person

Conditions	Patients	Healthy control	t	*p*
Numbers	41	44		
Age（years）	39.69±9.98	42.07±13.70	0.912	0.364
RBPT	+	-		
SAT	≥ 1:100	-		

All the 85 persons were tested the serum levels of PYD, C2C and OC. The median levels of serum PYD, C2C and OC in the patients group were 278.53 nmol/l, 82.23 ug/l, and 8.41ug/l, respectively, while the

median levels of serum PYD, C2C and OC in the healthy control group were 210.54 nmol/l, 72.74 ug/l and 7.43 ug/l, respectively. Comparing the results of serum PYD, C2C and OC with rank sum test of the nonparametric test, there were a significant differences among two groups (Z = 5.686, 3.997, 3.579, all *P* = 0.000). Table 2

Table 2 . Serum levels of PYD , C2C and OC of objective person

ers	Patients				Healthy control				Z	*P*
	median	Range	25%Q	75%Q	median	Range	25%Q	75%Q		
ıol/l	278.53	139.58-397.32	233.07	314.72	210.54	86.61-244.89	191.38	224.46	5.686	0.000
l	82.23	46.85-111.12	75.33	92.17	72.74	29.61-94.35	59.28	92.17	3.997	0.000
	8.41	3.33-17.19	7.93	9.09	7.43	2.69-11.42	6.93	8.18	3.579	0.000

Q: inter-quartile range ,

Discussion

Human brucellosis can cause serious complications. A considerable number of brucellosis patients with musculoskeletal changes who had first been referred to rheumatologists and orthopedists may have suffered longer diagnostic delays or even misdiagnoses so, early diagnosis is necessary to avoid these problems [7,13].

Radiography is a useful method for diagnosing musculoskeletal changes of human brucellosis; however, radiography showed a low level of sensitivity in the early stages[14] so, the diagnosis of musculoskeletal changes in this disease is often delayed. Musculoskeletal changes of human brucellosis is frequently destructive, with associated osteopenia and cartilage damage [15]. Metalloproteinases might be a promising procedure for determining osteoarticular changes of human brucellosis [16]; gamma interferon (IFN- γ) and tumor necrosis factoralpha (TNF-α) are involved in the pathophysiology of brucellosis and are closely related to the inflammatory activation of the disease [17]. But these biomarkers are not specific and do not provide a definite clinical diagnosis. So, it is very necessary to find biomarkers for reflecting musculoskeletal changes among human brucellosis.

PYD derive from the breakdown of collagen. PYD has been validated as usefulmarkers for bone resorption and their levels are significantly higher in OA patients [18]. Urinary PYD had a significant correlation with radiographic grades of OA though the synthesis of osteophytes, sclerosis of subchondral bone and synovial degeneration as well as cartilage degeneration in the joints of OA [19]. C2C is a degradation of 3/4 fragment of type II collagen. C2C may reflect the extent of cartilage degradation and significantly increased both in serum of OA patients [20]. C2C has also been shown to correlate with knee degeneration in patients with symptomatic knee osteoarthritis [21]. OC is one of the most abundant noncollagen bone proteins in bone matrix, which has been used as a biochemical marker of bone formation and bone turnover [22]. The synthetic of OC significantly increased, suggesting the potentiality of bone tissues in the remodeling processes [23]. In this study, the serum levels of PYD, C2C and OC

among male patients were higher than that of healthy control with a statistic significance,indicating that there were abnormal changes of collagen metabolism in bone and cartilage and abnormal changes of bone formation and bone turnover.

Brucellosis is more common in males [24]. Worldwide, males are affected more often than females, with a ratio of 5:2–5:3 in endemic areas [13]. Male patients predominance likely reflects the exposure pattern of human brucellosis. Male patients predominated in the patients with spinal brucellosis, which was the foremost cause of the debilitating and disabling complications of brucellosis [25]. Thus, serum changes of PYD C2C and OC among male brucellosis patients at early period are meaningful, and we also exlcuded effect on osteoporosis caused by female with menopause and age.

Meanwhile, there were a few limitations in the study. First, there were lack of data of radiography among these patients at early stage. Second, brucellosis patients could happen abnorml changes at early stage, but we also should know these changes at different stages. Third, we should compare others osteoarthritis with brucellosis. Further research was needed.

Conclusion

Through this study, we found that serum levels of PYD, C2C and OC were increased among male brucellosis patients at early stage, which might be related with abnormal changes of collagen metabolism in cartilage and bone formation and bone turnover.The serum levels of PYD, C2C and OC might be the indicator biomarkers for osteoarticular changes of human brucellosis at early stage. Further research was needed to probe the relationship between these biomarkers and brucellosis patients with/without the osteoarticular clinical symptoms at early stage.

Declarations

Ethical Approval and Consent to participate

This study was in compliance with the ethical principles in the Declaration of Helsinki of the world medical association and was approved by the ethics committee of the Qinghai institute for endemic disease prevention and control. All subjects signed an informed consent form.

Consent for publication

All the authors have read the manuscript and have agreed to submit it in its current form for consideration for publication in the Journal.

Availability of data and materials

Data can be got in whole paper

Competing interests

None of the authors has any conflicting interests to declare.

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Authors' contributions

Zhijun Zhao and Qiang Li written this paper and carried out all the experiment;Jiquan Li and Lansheng Hu performed the statistical analysis; Li Ma and Jiming Wang carried out the samples selection; Liqing Xu and Zhijun Zhao conceived of the whole study and participated in its design and coordination and helped to draft the manuscript.

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