

Original Article

Comparison of azithromycin vs doxycycline prophylaxis in leptospirosis, A randomized double blind placebo-controlled trial

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Abstract

Introduction: Leptospirosis is an important zoonotic disease in paddy field with 29.5% prevalence rate in Mazandaran province and 4% to 52% mortality rate among hospitalized patients. Prevention is an important strategy for the control of this disease. This study aimed to compare the prophylactic effect of azithromycin versus doxycycline against leptospirosis in an endemic area in north of Iran.

Methodology: In this randomized double-blind placebo-controlled trial, paddy field workers (n = 187) were randomized to receive azithromycin (500mg weekly), doxycycline (200 mg weekly) or placebo starting one week before exposure to paddy field, during and to four weeks after. Paddy field workers aged 18- 65 years who signed the informed consent form were assessed for signs and symptoms of leptospirosis in addition to serologic evidence of the disease 6th and 12th week. Data were analyzed with SPSS version 13 using Chi-square and Fisher exact test and ANOVA.

Results: From June to September 2016, 187 participants were entered the study to receive azithromycin (n = 66), doxycycline (n = 71) or placebo (n = 50). In terms of preventing against clinical leptospirosis, there was not any significant difference between three arms, though there was statistically significant difference of seropositivity after 6 and 12 weeks in comparison to baseline among all three groups (P = 0.029) and between active treatment (eg. azithromycin and doxycycline) groups and placebo group (P = 0.01).

Conclusion: Azithromycin like doxycycline decreased seropositivity without significant effect on clinical leptospirosis.

Key words: leptospirosis; chemoprophylaxis; doxycycline; azithromycin; clinical trial.

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Introduction

Leptospirosis is a zoonotic disease with a protean presentation. It is a tropical region disease, though sometimes occurs in temperate areas [1]. It usually develops in mammals and humans following exposure to the causative organism, *Leptospira interrogans*, through damaged skin. The prevalence of the disease is increasing, especially in the endemic area because of work with naked and unprotected feet in the paddy field [2].

Leptospirosis presents in a broad range manifestation from asymptomatic and only positive serology to severe disease with high mortality and poor prognosis [3-5]. Vasculitis with necrosis of extremities may be seen in complicated patients [6,7] with 4 to 52 percent mortality rate [8-11].

Chemoprophylaxis is the primary approach to prevent leptospirosis among high risk individuals including paddy field workers, but those are not completely effective [11-15] Some studies have shown that doxycycline is effective as a chemoprophylactic agent, though the gastrointestinal side effects is a concern [11,12,14].

Azithromycin has been evaluated as a chemoprophylactic agent, but most studies are in vitro and animal researches. It may be associated with less adverse reactions compared to doxycycline [16,17].

Regarding to endemicity of leptospirosis in our province [18,19] and large number of cases admitted in our ward every year, the aim of this study was to evaluate the efficacy of azithromycin as a

chemoprophylactic agent in leptospirosis in comparison with doxycycline.

Methodology

Study design

This randomized double blind placebo-controlled study was conducted in three different endemic cities for leptospirosis in Mazandaran province in north of Iran including Sari, Ghaemshahr and Jouybar from June to September 2016, collaborated by governmental – primary health care centers. The study was approved by the research council of Mazandaran University of Medical Sciences with ethic committee code of 882.ir.Mazums.REC.94. The study proposal was submitted to Iranian Randomized Controlled Trials (IRCT) with approval code of IRCTIRCT2015052322383N1. The clinical variables measured based on history and physical examination. All participants were evaluated by main research team and next follow up was done by experienced primary health center staffs who were residing and working in the primary health care centers of the study area. All physicians were informed about the study aims, details of methods and re-educated about the clinical signs and symptoms of leptospirosis

Study sample

The inclusion criteria of the study were as follows: age of 18- 65 years old, residing in endemic area, signing the informed consent form (taken from WHO Research Ethic Review Committee) [20], to follow the study protocol including use of drugs according to study design, to accept to come back based on predefined appointments for laboratory screening tests and commitment to report any clinical manifestation experienced during the study period.

Patients with history of hypersensitivity to azithromycin and doxycycline, positive first screening test for leptospirosis, pregnant women, previous history of any severe adverse reaction to doxycycline and azithromycin were excluded.

The eligible subjects were randomized into three groups to receive azithromycin (500 mg weekly, Iran Daru Company, Tehran Iran), doxycycline (200 mg weekly, Exir Company, Tehran, Iran) or placebo starting one week before exposure to paddy field, during (takes around 6 weeks) and to four weeks after that. Health center staff and participants were blind. During the study period, all individuals were asked about any symptoms or signs related to leptospirosis and underwent clinical examination when needed.

Immunoglobulin G (IgG) screening test (ELISA kit, DRG international Incorporation, New York, USA) was done for all individuals at baseline and at weeks 6 and 12 of the study. For participants with signs or symptoms of leptospirosis during study period, a blood sample was drawn two weeks after the onset of symptoms to evaluate the Immunoglobulin M (IgM) anti leptospira titer.

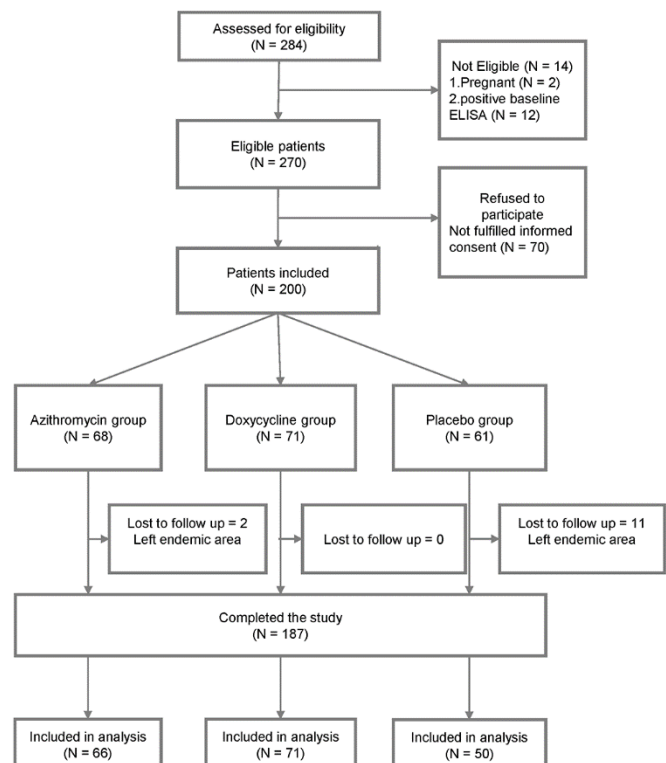
Sample size

Considering 95% two-sided confidence level, 80% power and adjustment for non-respondent of 20%, the samples size was calculated as ninety persons in each group.

Statistical analysis

Data was analyzed using SPSS version 13.0 (IBM Corp. Nie NH, Hull HC and Bent DH, Palo Alto, USA). Chi-square and fisher exact test were used for comparison of qualitative data. To compare the mean differences between three study groups, analysis of variance (ANOVA) was executed. Kolmogorov-Smirnov Test and Levene's test were used to examine the normality of data and homogeneity of variance, respectively. P-value of less than 0.05 was considered a significant difference

Figure 1. Flow-chart of patient inclusion in the study.



Results

We designed flow-chart of patient inclusion for the study as represented in Figure 1.

The mean age of participants was 47.5 ± 10.6 and 66.5% were male. In azithromycin group, 5 cases (7.6%), doxycycline group, 8 cases (11.3%) and placebo group, 12 cases (24%) had positive IgG antibody titer against *Leptospira* at weeks 6 and 12. The positivity of IgG antibody titer in intervening group, azithromycin and doxycycline (9.5%) were statistically different compared to placebo group (12 cases, 24%) (P = 0.01, OR = 3.012, 95% CI: 1.27-7.16).

The IgG seropositivity in azithromycin group was significantly less than placebo group (7.6% vs. 24%; P = 0.013, 95% CI = 1.26-11.8, OR = 3.85). Seropositivity results in doxycycline and placebo groups comparison were not as same as azithromycin (P = 0.063, 95% CI = 0.93-6.63, OR = 2.48). The IgG seropositivity of doxycycline was not as less as azithromycin and in the latter group was different statistically (P = 0.03) (Table1).

During the study, while they were working in the field and receiving chemo-prophylaxis, 13 participants, including 2 cases (2.7%), 9 cases (10.2%) and 2 cases (3.4%) in azithromycin, doxycycline and placebo groups, respectively, developed a clinical syndrome consistent with leptospirosis and had positive IgM antibody titer against *Leptospira*. Fever was the most common symptom experienced by 11 out of 13 of the patients (85%). Other presentations were myalgia, red eye, headache, diarrhea, arthralgia, cough and malaise.

In terms of prevention against clinical disease, no statistically significant results were found in comparison among different groups including azithromycin arm versus doxycycline arm (P = 0.058, 95% CI = 0.05-19.6, OR = 4.1). Azithromycin group versus placebo group (P = 0.8), doxycycline group versus placebo group (P = 0.12) and azithromycin plus doxycycline groups versus placebo group (P = 0.34).

Six out of 71 participants in doxycycline group (heartburn = 2, photosensitivity = 3, vertigo = 1) and 5 out of 66 participants in azithromycin group (epigastric pain = 4, urticaria = 1) developed adverse reaction in whom one in each arm discontinued medication (Table 1).

The rate of side effects due to chemoprophylaxis in two arms did not show significant difference statistically (P = 0.85).

Discussion

In the present study, use of both azithromycin and doxycycline decreased the seropositivity in intervened groups, especially in the former group which was significant statistically. Unlike the seropositivity, the incidence of overt clinical disease was not being influenced by using azithromycin or doxycycline. Of note, comparison among azithromycin and doxycycline showed a trend of preference for azithromycin in terms of prevention against clinical disease (P = 0.058) (Table1).

A limited number of reports showed efficacy of doxycycline as a prophylactic drug in human. The prophylactic usefulness of doxycycline against leptospirosis was firstly demonstrated in a study on US Army deployed in Panama. Takafuji *et al.* carried out a clinical trial for efficacy of doxycycline when American soldiers training in Panama. They found that weekly administration of doxycycline was effective as a preventive agent in 95% of cases [11]. In a similar study, Sehgal SC *et al.* examined the prophylactic role of doxycycline, 200 mg once weekly in residents of a rural area of the Andaman Islands, a high endemic area with annual outbreaks of leptospirosis associated with flooding. They reported that doxycycline prevents occurrence of overt clinical disease in 50% of cases [12] In contrary to above mentioned studies (Takafuji and Sehgal *et al.*), our study did not show efficacy of doxycycline in terms of preventing the clinical disease.

Table 1. Characteristics of participants and sero-positivity and clinical symptoms after chemo-prophylaxis.

10	All (n = 187)	Azithromycin (n = 66)	Doxycycline (n = 71)	Placebo (n = 50)	P-value
Age (mean ± SD)	47.5 ± 10.6	47.3 ± 8.9	48.2 ± 11.0	46.6 ± 12.4	0.63
Sex (%), male	66.5	59.5	70.5	69.5	0.695
Clinical symptoms during study, N (%)	13 (6.9)	2 (2.7)	9 (10.2)	2 (3.4)	0.08
IgM seropositive (N, %)	13 (6.95)	2 (2.7)	9 (10.2)	2 (3.4)	0.08
IgG seropositive, (N, %)	25 (13.4)	5 (7.6)	8 (11.3)	12 (24)	0.03
IgG seropositive, (N, %)	----	5 (7.6)	----	12 (24)	0.01
IgG seropositive, (N, %)	----	----	8 (11.3)	12 (24)	0.06
IgG seropositive, (N, %)	---		13(9.5)	12 (24)	0.01
Adverse drug reactions (N, %)	----	5 (7.6)	6 (8.4%)	----	0.85

A pilot, double-blinded, randomized controlled study assessed the effectiveness of single dose of doxycycline versus placebo in people with a heavy exposure to leptospira in São Paulo, SP, Brazil. The seropositivity and development of clinical leptospirosis was not statistically different between doxycycline and placebo. These researchers believed that their results may be due to small sample size [13]. The results of this study were similar to our study as doxycycline could not prevent the clinical leptospirosis.

Brett-Major DM *et al.* did a Cochrane study on antibiotic prophylaxis against leptospirosis. They concluded that weekly administration of 200 mg doxycycline has no clear efficacy on symptomatic disease or seropositivity and on the other hand produces some gastro intestinal adverse reactions like nausea and vomiting which prevents working in the paddy field [14]. recent study showed efficacy of doxycycline on seropositivity but no preventive efficacy on clinical disease and showed some adverse reactions of doxycycline particularly photosensitivity that decreases adherence.

Doxycycline as a tetracycline drug has some gastrointestinal adverse reaction which may cause low adherence [16] and using a better adverse drug reaction profile such as azithromycin would be a reasonable alternative. As noted, Azithromycin was successfully used as a prophylactic agent in leptospirosis in animal and in vitro studies [16,17] and to the best of our knowledge, there is not any randomized placebo-controlled study comparing the prophylactic role of azithromycin with doxycycline in human high risk population. In our study, azithromycin was associated with some adverse reactions (7.6%) but wasn't statistically significant in comparison to doxycycline.

Prophylactic administration of antibiotics is a cost-effective approach to prevent complications of leptospirosis [21]. Galloway *et al.* noted the benefits and cost effectiveness of both empirical (within 4-7 days of onset of symptoms) and prophylactic therapy to traditional therapy which initiated 7-day after onset of disease. Among three different approaches, the prophylactic administration of antibacterial agent has been associated with a higher survival rates.

Despite the previous studies, the efficacy of doxycycline as a prophylactic agent in endemic or epidemic settings remains unclear. Animal studies demonstrated that azithromycin is both effective and cost-benefit in terms of prophylaxis against leptospirosis, but the clinical evidences of prophylactic usage of azithromycin are poor and current evidences are not strong. The efficacy of chemoprophylaxis with

doxycycline (200 mg once a week) or azithromycin (in pregnant women and children) is being disputed, but focused pre-and post-exposure administration is indicated in instances of well-defined short-term exposure. Another benefit of azithromycin is its safety in pregnant women and children [22].

In fact, regarding to mentioned studies with anecdotal results, in contrast to effect on serologic response, chemoprophylaxis does not have significant effect on clinical disease. Regarding to current data so far which are just in animal and in vitro, it needs more study in human as a chemo- prophylactic agent.

Limitations of the study

The study was limited by using ELISA as a very sensitive test to detect the seropositivity against leptospirosis, instead of MAT test, which is the gold standard test recognized with high specificity for detecting the seropositivity of leptospirosis. MAT is a very expensive test and is not routinely used in clinical practice and no reference laboratory.

Conclusions

Azithromycin was associated with less seropositivity in paddy field workers compared to doxycycline and similar adverse reactions. To prevent overt clinical leptospirosis there was not different effect between two chemo-preventive regimens.

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