

# Coming National Program of Epidemiological Survey for Trachoma in China: Prevalence of Trachoma in Northern China

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## Abstract

**Purpose:** To assess the prevalence of and risk factors for trachoma among primary school children in three north districts of China.

**Methods:** The survey was carried out in three north districts of China (Wuqiang county of Hebei province, Yinchuan city of Ningxia Huizu province, and Datong city of Shanxi province). Chester sampling survey was employed in this study. The general state of health and case history of those children were recorded. Sanitation status and habits of both respondent and household, knowledge of health-related issues and the condition of inhabitancy were also questioned. Trachoma was clinically diagnosed according to the simplified classification for trachoma proposed by the World Health Organization (WHO). At the same time, *Chlamydia trachomatis* (*C. trachomatis*) was detected by polymerase chain reaction (PCR) when the child was clinically diagnosed as trachoma.

**Results:** In Wuqiang county, out of 1622 primary school children, 333 children were clinically diagnosed as trachoma and the prevalence of trachoma was 20.5% (95%CI 18.5% to 22.5%). In Yinchuan city, out of 1883 primary school children, 577 children were diagnosed as trachoma and the prevalence was 30.6% (95% CI 28.6% to 32.7%). Out of 1236 primary school children in Datong city, 135 person of trachoma were found, the prevalence of trachoma was 10.9% (95%CI 9.2%-12.6%). The prevalence of trachoma in primary school children from countryside is higher than that in cities ( $P<0.01$ ). The positive rate of PCR for *C. trachomatis* was 64.9% (in 333), 48.9% (in 577), and 63.7% (in 135) in the 3 districts respectively. Narrow living condition, short of clean water and poor personal sanitation habits, and poor knowledge of trachoma were the independent risk factors for active trachoma.

**Conclusions:** Trachoma is still now a remarkable eye health problem in north China. It is necessary to have a nationwide survey for the prevalence of whole country and evaluate mass interventions, both medical and educational, in the community.

**Keywords:** Trachoma; *Chlamydia trachomatis*; Prevalence; Risk factors; Children; Polymerase chain reaction

## Introduction

Trachoma, caused by the obligate intracellular parasite *Chlamydia trachomatis* (*C. trachomatis*), has been prevalent about hundreds years. Today, trachoma is still the leading infectious cause of blindness worldwide [1]. Overall, it is the eighth commonest blinding disease [2]. The burden of trachoma on affected individuals and communities can be huge in terms of both the disability it causes and the economic costs that result. So it is the target of World Health Organization (WHO) initiative for the Global Elimination of Blinding Trachoma by 2020 (GET 2020) [3].

It is believed that trachoma is endemic in 56 countries [4], particularly in the Sahel belt and East Africa. In addition, there are countries in the Middle East, the Indian sub-Continent, and Southeast Asia. The increasing availability of district-level data, based on surveys rather than estimates from countries or districts that have not reported previously have greatly improved knowledge of the trachoma burden.

China was the severely afflicted area of trachoma in the 20 century. Whenever, there had been many scientists and ophthalmologists worldwide made great effort to study the pathogen and to control the blinding disease. It was until 1957 that Tang et al. [5] completed the first successful isolation of *C. trachomatis*, using chicken embryos whose York sacs had been inoculated with material from infected

human eyes in Beijing, China. This significant contribution, which Chinese scientists and ophthalmologists had made, was the great basic work for further studying *C. trachomatis*, treating and preventing the blinding disease.

In China, trachoma is an important public health problem in China, but there are currently few data on prevalence of the disease and no formal trachoma control programs. One study reported a prevalence of less than 2% in Hainan province [6], while another study reported an incidence of 29% in one rural school in Guangxi [7]. In this study, we sought to estimate the status of trachoma in three districts of north China, in order to gather evidence for implementation of control measures.

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## Materials and Methods

### Ethics statement

The parents and the Ethical Review Committee of the China Ministry of Health granted the ethical approval for all protocols and procedures. The equivalent committees of Beijing Tongren Hospital and Capital University of Medical Sciences have approved the study. The survey had been consented by the children and their parents.

### Surveys on the epidemic of trachoma in the primary schools

We followed WHO recommendations for calculating sample size in trachoma prevalence surveys [8]:  $N = e \times d \times b \times (1-b) / c$  ( $e=4$ ,  $d=1.96$ ,  $c=b \times 20\%$ ). Based on previous surveys in other districts [6,7], the expected prevalence of trachoma in children was 25%. Using a precision of 5% and a design effect of 5, the required sample size for estimating TF prevalence in children was 1152 at least in each district.

The study was conducted from Sep. of 2004 to Oct. of 2006. This survey was undertaken in three north districts, including Wuqiang county of Hebei province, Yinchuan city of Ningxia Huizu province, and Datong city of Shanxi province (Figure 1). Across the three districts, there are traversed by Yellow River. A cluster sampling in which class shift was the sampling unit selected primary school children aged 6 to 16 years old. There were fourteen schools investigated randomly, including eight countryside schools and six urban schools.

Senior ophthalmologists saw all children in the study. Clinical examination of the eye involved in careful inspection of the eyelash, the cornea, the lid margin and the tarsal conjunctivas under the slit-lamp microscope. The signs were recorded in fact, and the trachoma was diagnosed according to the WHO simplified grading system

[9]. In the criteria, follicular trachoma (TF) is identified if there are five or more follicular on the surface of the upper tarsal conjunctiva, and intense trachoma (TI) if there is inflammatory thickening of the upper tarsal conjunctiva that obscures more than half of the normal, deep conjunctiva vessels. Trachomatous scarring (TS) is indicated by scarring in the upper tarsal conjunctiva. Before the surveys, the two trachoma graders were standardized against a senior grader, with reliability of  $\kappa = 0.85$  for follicular trachoma and for trachoma intense. All people who handled specimens were trained in proper laboratory techniques using sterile gloves changed between participants.

The general state of health and case history of those children were recorded. Furthermore, sanitation status and habits of both respondent and household, knowledge of health-related issues, and the condition of inhabitanacy were also questioned. The risk factors for trachoma were compared using logistic regression taking into account the community survey structure. If the child was clinically diagnosed as trachoma, a conjunctiva swab would be collected in DNA-free tubes for polymerase-chain-reaction assay of *C. trachomatis*, and stored at  $-20^{\circ}\text{C}$ .

If the child had trachoma, Rifampicin eye drops and instructions on proper use were provided.

### Detection the *C. trachomatis* by plasmid polymerase chain reaction (PCR)

The specific primers designed with reference to Lan [10], for generating a specific fragment of the *C. trachomatis* cryptic plasmid DNA. The primer sequences are: CTP1 (plus strand; 5' TAGTAACTGCCACTTCAT 3'); CTP2 (minus strand; 5'TCCCCTTGTAATTCGTGC 3'). PCR were performed with a final volume of 20  $\mu\text{l}$  containing final concentration of 50 mM KCl, 1.5 mM  $\text{MgCl}_2$ , 10 mM TrisHCl (pH8.3), 0.2 mM dNTP, 25 pM of each primer, 5  $\mu\text{l}$  of sample and 0.5 Utaq polymerase (Promega USA). The PCR amplification consisted of DNA denaturization at  $94^{\circ}\text{C}$  for 5 min, followed by 35 cycles of amplifications with a thermal cycles. Each cycle consisted of a denaturation step at  $94^{\circ}\text{C}$  for 1 min, an annealing step at  $45^{\circ}\text{C}$  for 1 min, and a chain elongation step at  $72^{\circ}\text{C}$  for 1 min. The final elongation step was extended for another 5 min.

Sample containing only the PCR mixture were used as negative control. DNA of *C. trachomatis* TE55 strain (stored by Beijing Institute of Ophthalmology, Beijing Tongren Eye Center, Beijing, China, in 1991) was used as a positive control. Finally, 6  $\mu\text{l}$  of the PCR product was analyzed by 1.5% agarose gel electrophoresis and stained with ethidium bromide.

## Results

### Prevalence of trachoma in Children

There were 4741 children in all, which had been surveyed in the three districts, and the prevalence of trachoma was varied from 10.9% (9.2%-12.6%) to 30.6% (28.6-32.7%) (Table 1). In Wuqiang county, 1622 children were investigation, and 333 cases were diagnosed as trachoma clinically (Table 2). The prevalence of trachoma varied from 7.5% to 40.6% in different schools, averagely 20.5% (95%CI 18.5% to 22.5%). 939 children were from four countryside primary schools, including Beidai school, Changcun school, Sunzhuang school and Jieguan school, being responsible for 11.5% , 11.6% , 38%, and 40.6% of cases respectively. 683 children were from three urban primary schools, including Shiyan school, Nanpai school, and Beipai school, being responsible for 7.5%, 17.7%, and 19.7% of cases respectively. The prevalence of trachoma in country schools is higher than that in urban schools ( $P < 0.01$ ).



In Yinchuan city, 577 cases were diagnosed as trachoma clinically among 1883 children (Table 3), and the prevalence was 30.6% (95% CI 28.6% to 32.7%). 796 children were from two countryside primary schools, including Fendeng school and Qinshuiyuan school, being responsible for 33.2% overall of cases. In addition, 1087 primary children were from an urban school, Jinfeng incorporate primary school, being responsible for was 28.8% of cases. It was higher in the country schools than in urban school.

A total of 1236 children were examined in Datong city (Table 4). Among which, 135 cases of trachoma were found clinically, and the prevalence of trachoma was 10.9% (95%CI 9.2% to 12.6%). 569 children were from two countryside schools, including Beishan school and Xintianbao school, being responsible for 12.0% and 10.9% of cases respectively. And 667 children were from two urban schools, including No.1 and No.2 of Xinrongqu, being responsible for 9.9% and 11.1% of cases respectively. The prevalence of trachoma in the urban schools was higher than in the country schools, but the difference was insignificant between them.

Overall, the most cases of trachoma were in active trachoma: Trachoma Inflammation Follicular (TF) and/or Trachoma Inflammation Intense (TI). Furthermore, the incidence of *C. trachomatis* in children with TF was higher remarkable than of the patients with TI in all the three districts ( $P < 0.01$ ).

### The results of plasmid PCR analysis

Amplicor PCR analysis revealed that there was an incidence

of 64.9% (in 333), 48.9% (in 577), and 63.7% (in 135) infection in Wuqiang county, Yinchuan city, and Datong city respectively. There were no positive cases in control children. The results of plasmid PCR were different in different grades trachoma patient. The incidence of TI cases is higher than TF cases remarkable ( $P < 0.001$ ).

### Household characteristics and Risks for trachoma

Risk factors for trachoma in these districts included crowding living condition, poor access to water, poor sanitation and shortage of knowledge about trachoma. More population in the family, more likely children had been with trachoma. >5 persons of the family in the compound, children were more likely to have the symptom of trachoma. Besides, children who lodged in school were ease infected by *C. trachomatis* because of crowding and poor sanitation.

An unclean face was the most important characteristic associated with trachoma. There was a significant increase in the prevalence of trachoma among children who had dirty face with nasal discharges and eye discharges ( $P < 0.01$ ). In countryside school, there was lack of water faucet or far to get source of water. It was a difficult thing to access to clean water. The poor personal sanitation was fostered. So, children washed their faces and hands once or twice a week. There was a significant reduction in the prevalence of trachoma among children who had clean hands and faces ( $P < 0.01$ ). In our study, about >50% of children did not know that trachoma being transmitted by contact each other, as well as the symptoms of active trachoma and the way

Districts	No. of children	Range of prevalence in different schools	Follicular trachoma	Intense trachoma	Trachomatous scarring	Mean prevalence (95% CI)
Wuqiang	1622	7.5-40.6%	14.36%	5.98%	0.18%	20.5% (18.5- 22.5%)
Yinchuan	1883	28.8-33.2%	18.69%	7.70%	4.25%	30.6% (28.6-32.7%)
Datong	1236	9.9-12.0%	9.5%	1.4%	0	10.9% (9.2%-12.6%)

Table 1: Prevalence of trachoma in north three districts in China.

	Countryside schools					Urban schools			
	Beidai	Changcun	Sunzhuang	Jieguan	Total	Shiyan	Nanpai	Beipai	Total
No. of children	183	267	208	281	939	361	124	198	683
Follicular trachoma	12 (6.6%)	21 (7.9%)	65 (31.3%)	70 (24.9%)	168 (17.9%)	22 (6.1%)	18 (14.5%)	25 (12.6%)	65 (9.5%)
Intense trachoma	9 (4.9%)	9 (3.4%)	13 (6.3%)	44 (15.7%)	75 (8.0%)	5 (1.4%)	4 (3.2%)	13 (6.6%)	22 (3.2%)
Trachomatous scarring	0	1 (0.4%)	1 (0.5%)	0	2 (0.2%)	0	0	1 (0.5%)	1 (0.1%)
Mean trachoma Prevalence	11.5%	11.6%	38%	40.6%	26.1% (95% CI 23.6 ~28.6%)	7.5%	17.7%	19.7%	12.9% (95% CI 11.2~14.8%)

Table 2: Prevalence of trachoma in different schools in Wuqiang Country.

	Countryside schools			Urban schools
	Fendeng	Qinshuiyuan	Total	Jinfeng
No. of children	404	392	796	1087
Follicular trachoma	60(14.9%)	81(20.7%)	141(17.8%)	211(11.2%)
Intense trachoma	42(10.4%)	29(7.4%)	71(8.9%)	74(6.8%)
Trachomatous scarring	32(7.9%)	20(5.1%)	52(6.5%)	28(2.3%)
Mean trachoma Prevalence	33.2%	33.2%	33.2% (95% CI 29.9~36.5%)	28.8% (95% CI 26.1~31.5%)

Table 3: Prevalence of trachoma in different schools in Yinchuan City.

	Countryside schools			Urban schools		
	Beishan	Xintianbao	Total	No.1 of Xinrongqu	No.2 of Xinrongqu	Total
No. of children	275	294	569	352	315	667
Follicular trachoma	30(10.9%)	23(7.8%)	53(9.3%)	33(9.4%)	31(9.8%)	64(9.6%)
Intense trachoma	3 (1.1%)	9 (3.1%)	12 (2.1%)	2 (0.5%)	4 (1.3%)	6 (0.9%)
Trachomatous scarring	0	0	0	0	0	0
Mean prevalence	12.0%	10.9%	11.5%	9.9%	11.1%	10.5%

Table 4: Prevalence of trachoma in different schools in Datong City.

to protect from it. Age and gender did not affect the incidence of trachoma in the survey.

## Discussion

Trachoma, as a public health problem, has been paid much attention to by ophthalmologist and sociologist. The WHO has made many estimates of the trachoma endemic countries worldwide and other organizes [11-16]. The most recent estimate, suggests that there are currently about 40 million people with active trachoma and 8.2 million with trichiasis [17]. These estimates have generally been produced with models that have relied on the results of a limited number of surveys conducted in a few endemic countries. The results have been extrapolated across these countries and to other countries in the same region.

In China, just as Burton and Mabey reported [18], where pockets of the disease are thought to exist: even with a low prevalence, the contribution from the most populous country could make a profound difference in the global burden of trachoma. China is indeed on the WHO list of areas endemic with trachoma, and this survey confirmed that designation. The survey relied on the clinical examination of children for evidence of trachoma using WHO guidelines. The prevalence of trachoma overall in this survey was lower than reports from Mali and Thiopia, which were 39% [19] and 51% [20] respectively. However, in one countryside schools of Wuqiang county, it was responsible for 40.6% of cases. The high rate was very close to which reported from Ethiopia and Sudan, where active trachoma is often found out more than 50% in children [21,22]. The prevalence of trachoma was closed to the report of Tengzhou City of Shandong Province, which being responsible for 22.2% of cases in 2676 rural children [23]. On the face of it, the active trachoma was still a remarkable eye health problem in the primary schools in North-China.

Trachoma is, first and foremost, a disease of poverty. It thrives in remote, marginalized, and displaced populations [24]. Restricted by historical, social and natural conditions, the economic and social development of the three districts where we surveyed are backward relatively. And the natural environment is also poor, without sufficient precipitation. Especially in Yinchaun city, it is very windy and dusty all the year round. The prevalence of trachoma was high both in countryside schools and in urban schools. Economic has been developing in Datong city recently, so the prevalence of trachoma was lower than the other districts comparatively. Worldwide, the incidence of trachoma has fallen as the social progress. One hundred years ago, trachoma was widespread in Europe and North America, but faded away during the first half of the 20th century as living conditions improved [25].

Risk factors for active trachoma comprise a mix of individual characteristics and household factors [19,26]. Risk factors for trachoma are generally things that favor the transmission of *C. trachomatis* from one person to another. The transmission of *C. trachomatis* from infected to noninfected individuals is necessary to sustain trachoma in endemic communities. Several routes of transmission are probably involved including direct spread (close contact), at communities [27], fomites, and flies on the face [18]. Crowded living conditions, for example with several young children sleeping in the same bed, probably promote transmission. The clustering probably reflected the importance of transmission of infection between members of the same family and transmission between families with close socialites.

Similarly, water scarcity probably promotes transmission, because less water is available to use for face and hand washing. An unclean

face was the most important characteristic associated with trachoma in our study. The prevalence of trachoma also reduced remarkable in the study of Cumbenland et al. [28], if children had clean hair, no nasal discharge, no eye discharge, and did not have flies on their face. The prevalence of human and animal faces was associated with flies and an increased risk of active trachoma in children in his study. In addition, children had a 2.5 times higher risk of active trachoma if they lived in households that reported defecation close to the house, or kept cows close to the house. That was indeed a question especially in countryside, but there were few animals in the households in our study. In addition, most children were lodged in school rather than in their family.

Unawareness of trachoma is also an important risk factor. Because people do not understand the transmission and prevention methods of trachoma, it is hard to eliminate trachoma fundamentally. It is urgent to conduct various popularized activities, strengthen mass media propaganda and face-to-face education to prevent trachoma epidemic among children. Both children and teachers were told to improve their personal hygiene in reducing the communication of trachoma. Teachers should make children wash their hands often, especially before they eat and after using the toilet.

In general, the diagnosis of trachoma is made on clinical grounds including symptoms and signs of conjunctiva. Laboratory testing to detect *C. trachomatis* is typically unavailable and unaffordable for clinical care in the epidemiology survey. However, the most patients were in TF stage and the symptoms and signs were mild. Especially, it is difficult to differential diagnosis trachoma from follicular conjunctivitis only by clinical manifestation if patients are children. Recently, laboratory tests have been used broadly to detect *C. trachomatis* infection on the eye in many trachoma surveys [1,6,29]. The common assays include isolation in cell culture, direct fluorescent antibody, microscopy of conjunctiva scrapings, enzyme immunoassay, serology, and nucleic acid amplification. PCR directed at plasmid genes is thought to be the most sensitive and specific for the diagnosis of *C. trachomatis* infection relatively [24].

In this study, the detection of *C. trachomatis* was PCR performed directly on conjunctiva specimens from children with trachoma clinical. The incidence varied from 48.9% to 64.9%. It was most important that plasmid PCR analysis confirmed the presence of *C. trachomatis*. In other trachoma surveys, the incidence of *C. trachomatis* plasmid PCR was about 30%-90% [29,30].

The incidence of *C. trachomatis* in children with TI was higher remarkable than of the patients with TF in the survey. It was probably because the pathological changes of their conjunctiva were mild, and few copies of *C. trachomatis* were present.

The result of PCR was reflected by sampling of conjunctiva scrapings, transport and preservation specimen, and operating in the PCR perform. Besides, it was possible that these and other organisms are able to reproduce the clinical signs of active trachoma, particularly in those who previously had a follicular reaction to ocular *C. trachomatis* infection [30]. It was necessary to place of laboratory tests in community assessment. While, more recent estimates have been more stringent, using only surveys with a national sampling frame and the updated WHO list of endemic countries [14]. Laboratory test should be an additional role for certifying the presence of trachoma as a public health problem.

Measures to control trachoma have been stepped up in an effort to meet the WHO-supported target. In 1997, WHO launched the Global Alliance for the Elimination of Trachoma (GET) for the control of trachoma based on the 'SAFE' strategy [31]. The strategy has four

components: Surgery, Antibiotics, Face washing and Environmental sanitation. Areas where trachoma is endemic are often extensive, but the extent to which neighboring communities are affected by the disease can vary considerably. It is clear that control cannot be achieved though management of symptomatic individuals presenting to health care facilities. It is necessary to assess the needs of each community in turn. In China, the economical development is not balanced between north and south, and the north is quite backward. Moreover, the medical service condition is also poor, especially in the countryside, where lacks the basic medical treatment facilities and doctors. As a result of the relatively ineffective immune response, repeated infection of the individual by *C. trachomatis* is common with age in an endemic environment. This leads to a recurrent chronic inflammation, which is associated with the development of scar tissue within the conjunctiva over many years. The WHO currently recommends the use of either oral Azithromycin (single dose) or topical tetracycline (twice daily for 6 weeks) [32,33]. In addition, the current recommendation is for mass drug administration of entire endemic communities to be conducted annually for several years. It is really a significant problem with a high burden of disability to us.

Anyway, trachoma is still prevalent in north China. It is associated with Hygiene and awareness of knowledge about trachoma. This study demonstrates the need for a broad target interventions and a powerful approach to the prevention from trachoma in endemic districts. Further surveys should be evaluated about mass interventions, both medical and educational, in the community. We need the support at all levels by government and international community. More efforts should be taken for elimination of the blinding trachoma in the most populous country before 2020.

#### Conflicts of Interest Statement

The authors have no conflicts of interest concerning the work reported in this paper.

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