

## Original Article

THE VALUE OF PROPER SPUTUM COLLECTION INSTRUCTION  
IN DETECTION OF ACID-FAST BACILLUS<sup>1</sup>Tetsuhisa HIROOKA, <sup>1</sup>Takeshi HIGUCHI, <sup>1</sup>Nakako TANAKA, and <sup>2</sup>Tsuyoshi OGURA

**Abstract** [Purpose] This study was conducted to assess clinically whether intervention with instruction applying respiratory rehabilitation method for expectorating sputum was useful or not to obtain more suitable sputum for smear examination of acid-fast bacilli.

[Subject] All specimens examined were sputa obtained from 163 patients without the instruction group and 161 patients with the instruction group, who visited our out-patients clinic during one year from September 1, 2000 to August 31, 2001 and the following one year, respectively.

[Method] Gross appearance of the sputum according to Miller & Jones' classification (M1, M2, P1–P3) and smear positive rate by fluorescence staining method after N-Acetyl-L-cysteine-NaOH treatment and centrifugation were compared between the two groups.

[Results] M1 and P1 sputa were 21.5% and 21.5% in the no instruction group, while those were 8.1%, 36.6% in the instruction group. Difference in M2, P2 and P3 sputa were not significant between the groups. Smear positive rate was 10.4% in the no instruction group, while it was 21.1% in the instruction group. According to gross appearance of M2, P1 and P2, positive rate was 11.1%, 11.4% and 30.8% in the no instruction group, and 17.7%, 28.8%, and 26.3% in the instruction group. Chest roentgenographic findings judged by type (cavitary and non-cavitary) and extent of the

pulmonary lesions of these smear positive cases (17 in the no instruction group and 34 with the instruction group), revealed that the no instruction group consisted of more predominantly severer disease with cavity and moderately or far advanced lesions as compared with the instruction group.

[Consideration] We could exclude that the difference in gross appearance and smear-positive rate of the sputum specimen between groups without and with the instruction might be due to differences in disease severity between the two groups with and without the instruction.

[Conclusion] The instruction for sputum expectoration seems to be useful to increase positive rate in the smear examination of acid-fast bacilli.

**Key words:** Acid-fast bacterium testing, Respiratory rehabilitation, Sputum collection instruction

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## Original Article

RELATIONS BETWEEN CLINICAL SUBTYPES OF *MYCOBACTERIUM AVIUM* PULMONARY DISEASE AND POLYCLONAL INFECTIONS DETECTED BY IS1245 BASED RESTRICTION FRAGMENT LENGTH POLYMORPHISM ANALYSIS<sup>1</sup>Katsuhiko KUWABARA, <sup>2</sup>Yasushi WATANABE, <sup>1</sup>Koichi WADA, and <sup>1</sup>Toshiaki TSUCHIYA

**Abstract** [Introduction] The epidemiology of *Mycobacterium avium-intracellulare* (MAC) infections has not been completely defined. Recently some reports presented polyclonal MAC infections. The purpose of this study was to reveal the clonal diversity of *Mycobacterium avium* isolates and the relation between clinical subtype of lung disease and polyclonal infection.

[Methods] We categorized pulmonary *Mycobacterium avium* infection to three clinical subtypes, tuberculosis like type, bronchiectasis with preexisting tuberculosis type and nodular bronchiectasis type. *Mycobacterium avium* isolates of 11 patients were studied for their heterogeneity using IS1245 based RFLP analysis. The insertion sequence IS1245 is repetitive element identified only in *Mycobacterium avium*. Standard method of IS1245 based RFLP analysis has been proposed as a suitable technique for typing of *Mycobacterium avium* isolates for epidemiological and taxonomic studies. At least three distinct colonies were subcultured to single clone. The subclones of the isolates were analyzed by IS1245 based RFLP technique and some subclones were also examined by antimicrobial susceptibility test.

[Results] Two of three patients of tuberculosis like type were considered to be monoclonal infection because only a single genotype was identified. And only one of four patients of bronchiectasis with preexisting tuberculosis type was considered to be polyclonal infection despite of long-term observation. Although isolates were collected in two or more occasions in clinical course over one year period, only a single genotype was observed in two patients. In contrast, three of four patients of nodular bronchiectasis type had multiple genotypes.

Isolates recovered from patients with monoclonal infection pattern following long-term treatment with clarithromycin monotherapy became resistant to clarithromycin. In contrast, three strains derived from one nodular bronchiectasis patient were susceptible to clarithromycin despite of long-term chemotherapy including clarithromycin. The susceptibility patterns

of the other drugs were also apparently different. Strain conversion due to repeated polyclonal infection was considered. These results of the antimicrobial susceptibility test supported clonal diversity of the *Mycobacterium avium* infection.

[Discussion] IS1245 based RFLP analysis possesses a discriminatory power between the isolates on clonal level. This study demonstrates that polyclonal infections are common in nodular bronchiectasis type and monoclonal infections are common in tuberculosis like type and bronchiectasis with preexisting tuberculosis type. And not only simultaneous polyclonal infection but also repeated polyclonal infection were observed in a nodular bronchiectasis type patient.

Drug susceptibility test showed long-term chemotherapy including clarithromycin could change the susceptibility of clarithromycin to resistant in patients with monoclonal infection. In contrast patients with repeated polyclonal infection pattern would avoid drug resistance because of strain conversion. This multiple susceptibility patterns identified in this study would not have been detected by the standard susceptibility test without subculture. And we also need the treatment strategy considering the polyclonal infection.

[Conclusions] Polyclonal infections are considered to be common in pulmonary *Mycobacterium avium* infection, especially nodular bronchiectasis type. Clonal diversity of *Mycobacterium avium* infection is an important factor to perform chemotherapy and drug susceptibility test.

**Key words :** *Mycobacterium avium*, RFLP analysis, IS1245, Polyclonal infection

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## BEIJING FAMILY AND OTHER GENOTYPES OF *MYCOBACTERIUM TUBERCULOSIS* ISOLATES IN OKAYAMA DISTRICT

<sup>1</sup>Ritsuko OHATA and <sup>2</sup>Atsuhiko TADA

**Abstract** [Objective] The purpose of this study is to investigate the distribution of *Mycobacterium tuberculosis* genotypes, such as Beijing family and other genotypes in Okayama district, and to examine the relation between these genotype strains and prevalent strains.

[Methods] The 142 *M. tuberculosis* strains isolated in Okayama City from January 2000 to December 2002 were subjected to IS6110-RFLP and spoligotyping for the population-based study. In addition, 13 strains having 1 to 5 IS6110 copies isolated in Okayama Prefecture excluding Okayama City were also subjected to spoligotyping.

[Results] A total of 103 (72.5%) of 142 strains belonged to Beijing family. Furthermore, 3 of 4 groups of prevalent strains belonged to the family. Twenty one strains out of 39 rest strains could be classified into 10 shared types (STs) of 259 STs in the worldwide spoligotype database. In addition, 15 out of 26 strains from inside and outside of Okayama City having 1 to 5 IS6110 copies showed common unique spoligotype. Moreover, the age of majority patients who were infected with *M.*

*tuberculosis* of the Beijing family or other genotypes were older than 60 years.

[Conclusion] It was suggested that Beijing family had been the main infection source, and the spread of strains of Beijing family and other genotypes occurred considerably in old times, and the unique genotype strains remained for a long time as peculiar strains.

**Key words:** *M.tuberculosis*, Beijing family, Prevalent strains, IS6110-RFLP, Spoligotyping

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————— The 78th Annual Meeting Symposium —————

## TUBERCULOSIS CONTROL PROGRAMME FOR THE ELDERLY WITH SPECIAL FOCUS ON EARLY DETECTION

Chairpersons: <sup>1</sup>Akira KAJIKI and <sup>2</sup>Nobukatsu ISHIKAWA

### Abstract

1. Epidemiological trend of TB and its diagnosis among the elderly : Masako OHMORI (Research Institute of Tuberculosis, JATA)

It is estimated that over 10,000 TB patients will occur among the elderly aged over 75 years of age during 2010 and 2020. Though the new TB patients among the elderly over 65 years of age decreased in number from 1987 to 2001, sputum smear positive patients, which are highly infectious increased by 1.3 times for the same age group, and 2.3 times for that of over 80 years of age. 17.6% of those elderly patients were detected in institutions, either hospitals or nursing homes. These indicate that the elderly will become higher risk of TB for causing early death and infection to others during their admission in the hospitals or nursing homes.

2. Clinical characteristics of TB among the elderly: Nobuhiko NAGATA (Department of Internal Medicine, National Ohmuta Hospital)

Analysis of the 93 elderly TB patients shows that TB diagnosis was delayed among the elderly. For TB diagnosis, 43.2% took over 1 month, and 27.3% over 2 months among the patients over 75 years of age, compared to 22.9% and 9.7% respectively for the patients under 65 years. The prognosis was also poor. Of all who died in the hospital during TB treatment, 40.9% (mostly complicated with other diseases) was among the elderly over 75 years, while it was 6.5% among those under 65 years. Complication of dementia delayed the diagnosis. 79.4% of the smear positive patients over 75 years were diagnosed while admitted in a general ward, and 40% had been admitted 2 weeks or more before diagnosis.

3. Use of serological tests for the diagnosis of TB among the elderly : Atsuyuki KURASHIMA (Department of Clinical Research, National Tokyo Hospital)

As TB diagnosis is difficult for the elderly due to non-specific appearances of signs/symptoms, X-ray shadows or negative tuberculin reaction, some serological tests, which have been recently developed, can be useful as a supplementary diagnostic tool. One of them is anti-lipoarabinomannan antibody. 74.3% was positive to this among 148 bacteriologically confirmed TB cases; 77.8% for those aged 65 years and above, and 71.1% for those under 65 years. To anti TBGL antibody, 78% of 170 confirmed TB cases were positive; 75% for those aged 65 years and above, 79.8% for those less than 65 years. Multi-lipo antibody developed by

Japan BCG Laboratory showed higher sensitivity of 91.5%. GPL-core antibody may increase the sensitivity.

4. Mode of TB detection in nursing homes : Shinji SHISHIDO (Research Institute of Tuberculosis, JATA)

15 elderly TB patients who had been diagnosed in 23 nursing homes in the last 5 years were analyzed. The average age was 80.7 years. 10 were male and 5 were female. 11 were sputum smear positive. The symptoms were fever (8), cough (7), wheezing (2), hemoptysis (1), chest pain (1), body weight loss (1), appetite loss (1). Number of patients by time durations before consulting a doctor of within 2 weeks, 2 weeks to 1 month, 1 to 2 months, 2 to 3 months, 3 to 6 months, and more than 6 months were 6, 1, 1, 3, 2, 2 respectively. The prognosis: 4 died within 10 days after diagnosis, 4 needed examinations for the symptoms but were delayed in diagnosis as they were not admitted due to dementia or disability. The orientation and training to the staff of the nursing homes are needed for early TB diagnosis.

5. Programme for the early detection of TB among the elderly : Tadayuki AHIKO (Murayama Public Health Center, Yamagata Prefecture)

Based on the analysis of 138 confirmed TB cases registered in Yamagata Prefecture in 1998, services for early TB detection among the elderly should be 1) periodical chest X-ray examinations by the patients home doctors when underlying high risk diseases such as diabetes or cancer exist, and 2) sputum examination for the symptomatic patients. A survey to 31 local governments conducting special TB services showed the special screening programme for the bedridden elderly in nursing homes was not so efficient due to low quality of the available X-ray facilities and low case detection rate. But these X-ray films can be utilized for comparison with those when any symptom arises.

**Key words** : Tuberculosis control for the elderly, Early detection

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## THE RESULTS OF DOTS STRATEGY

Chairpersons: <sup>1</sup>Hideo OGATA and <sup>2</sup>Takeko YAMASHITA

Topics of symposium and Presenters :

1. The trial of ambulatory intermittent DOT at pharmacies : Masako WADA (Research Institute of Tuberculosis)
2. Recommendation of DOTS, Japan version : Noriko KOBAYASHI (Research Institute of Tuberculosis)
3. Analysis of the factors on defaulter cases, and the cost effectiveness of tuberculosis treatment : Kazuhiro UCHIMURA (Research Institute of Tuberculosis)
4. Visions for the expansion of Japanese DOTS: Toru MORI (Research Institute of Tuberculosis)

**Abstract** Tuberculosis control in Japan is now on the stage of program transition since the declaration of emergency of tuberculosis in 1999. The “21st century DOTS, Japan version” has been proposed under the influence of DOTS experiences in the United States, where drastic reduction of tuberculosis was observed after the introduction of universal DOT. We cannot copy the experiences of US, considering the difference of epidemiological situation of tuberculosis, social background of TB cases and the difference of health infrastructure for TB patients. In the United States, many tuberculosis patients are treated under government health system with DOT. In Japan, TB cases are treated at clinics and hospitals under the integrated health system and public health centers mainly provide prevention services including contact exami-

nation of TB and cohort analysis of TB cases. 21st century DOTS, Japan version, is not the universal system at present but it is on the process of implementation and various kind of new activities have been tried including activities to assure the close collaboration between public health centers and clinics, and DOT activities including hospital based DOT, ambulatory intermittent DOT at pharmacies. Here are presented with various experiences both in the field of collaboration between public health centers and clinics, and in the field of DOT. Also here are presented with the calculation which shows the reduction of total cost of tuberculosis treatment by the introduction of ambulatory DOT, considering the influence of recurrence of defaulter cases which would have occurred without DOT. We consider that this symposium is the interim report of the expansion of Japanese DOTS.

**Key words** : DOTS, Hospital based DOT, Intermittent treatment, Cost effectiveness of tuberculosis treatment

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