

## ETIOLOGICAL INVESTIGATION OF FARMER'S LUNG — SEROLOGICAL STUDY

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### SUMMARY

The reactions of precipitins in serum against the antigens from two strains of *T. vulgaris* were shown in 46.7 and 66.7% in 30 patients with farmer's lung, significantly higher than those in the control groups, while the reactions against *M. faeni* and *A. fumigatus* were low in the patients' group and not significantly higher than those in two control groups. The results indicated that the main etiological agents of farmer's lung were some strains of *T. vulgaris* in the patients.

### INTRODUCTION

From 1980 through 1981, an epidemiological survey was conducted among 1054 hay grinders in Dafeng County, Jiangsu Province. 120 of them had history of farmer's lung disease. During follow-up study of these 120 grinders, acute episodes of farmer's lung after exposure to mouldy hay dust were seen in 67 of them.<sup>1</sup> Meanwhile, a microbiological study of sputum of these patients and mouldy hay samples from their workplaces was performed. 80 strains of thermophilic actinomycetes were isolated from these samples, 61.2% of them being *T. vulgaris*.<sup>2</sup> In order to confirm whether *T. vulgaris* was the main etiological agent of farmer's lung in that county, we studied the precipitins in serum from the patients using serological method.

### MATERIALS AND METHODS

#### Antigens

Six strains of Thermoactinomycetes, including 4 strains of *T. vulgaris* called 801, 806, 816, 832 and 2 strains of Thermophilic nocardia called 835 and 836, which were isolated from mouldy hay collected from the workplaces of the patients with farmer's lung, were selected and then, antigens were prepared by using a modified Salvaggio's method.<sup>3</sup> Meanwhile, the strains of *M. faeni* 1., *T. vulgaris* 2. and *T. candidus*, one of each, provided by Dr. V. P. Kurup (Medical College of Wisconsin) were also selected to prepare antigens. The antigens were diluted to 30 or 40 mg per ml of normal saline, when they were used. In addition, other antigens including those from *M. faeni* 2., *T. vulgaris* 1. provided by Dr. J. H. Edwards (MRC Pneumoconiosis Unit) and *A. fumigatus* provided by Dr. J. Marx, JR (Marshfield Medical Foundation) were also used for detecting precipitins in serological test. Besides, the extracts from mouldy hay was prepared by using a modified Williams' method and diluted to 12 mg per ml of normal saline, also employed in the serological test.

#### Serum Samples

Serum samples from 30 of these 67 patients were collected just one month after they ground mouldy hay. 30 serum samples from healthy people with no history of exposure to mouldy hay in the same area matched with the patients in sex and age were selected as control group A. Another 29 serum samples were collected from the healthy students in Shanghai Medical University as the control group B.

#### Serological Test

The presence of precipitins against the antigens was tested by using modified Ouchterlony's agar-gel double-diffusion assay.<sup>5</sup>

### RESULTS

It was shown that the reactions against two strains of *T. vulgaris* 1. and 2. were 46.7 and 66.7% in the patients' group, significantly higher than those in the control groups, whereas the reaction against *T. candidus* was 80% higher than that in group B, and it had not much difference with the control group A. Besides, the reactions against *M. faeni* 1. and 2. and *A. fumigatus* were rather low (16.7, 3.3 and 9.1%) in the patients' group and not significantly higher than those in the two control groups.

The reactions against six strains of thermophilic actinomycetes named *T. vulgaris* 801, 806, 816 and 832 and Thermophilic nocardia 835 and 836 ranged from 13.3 to 80.0% in the patients' group. The reactions against *T. vulgaris* 816 was 36.7% in the patients' group, significantly higher than that in the two control groups, and those against *T. vulgaris* 806 and 832 were 80.0 and 33.3% in the patients' group, significantly higher than those in the group B, but not in the group A. Besides, reactions against *T. vulgaris* 801, Thermophilic nocardia 835 and 836 and the extracts of mouldy hay in the patients' group were not significantly higher than those in the control group.

## DISCUSSION

The precipitin test against farmer's lung antigens has been widely used in clinical diagnosis and epidemiological survey of the farmer's lung disease. The positive reactions against these antigens always indicate that the people have the history of exposure to them. Based on these reactions, the etiological agents of farmer's lung could be determined.<sup>6</sup> In our study, the precipitins against a variety of farmer's lung antigens in sera from the patients with farmer's lung in Dafeng County, Jiangsu Province were tested and it was found that the percentages of positive reaction against three strains of *T. vulgaris* 1., 2. and 816 were 46.7, 66.7 and 36.7% in the patients with farmer's lung, respectively, which were significantly higher than those in the two control groups. The results might indicate that the main causative agents were some strains of *T. vulgaris*. The microbiological study of the mouldy hay from that county and sputum from the patients had also indicated that *T. vulgaris* was the dominant thermophilic actinomycetes in the samples, while *M. faeni* was not found in them.<sup>2</sup> So, the findings of our serological study and the microbiological study were consistent with each other.

Pepys had reported that the percentage of positive reaction against *M. faeni* in the patients with farmer's lung was as high as 85% in Britain.<sup>7</sup> So, the main causative agent of the disease was *M. faeni* in Britain. But in Finland, Terho found that the main etiological antigen of farmer's lung was from *T. vulgaris*.<sup>8</sup> Perhaps the difference might be referred to the different way of preparing and storing hay, perhaps also climatic differences and differences in crop types. In Dafeng County as well as other area in east part of China, hay before stocking would be sun-dried as much as it could be and then stocked outdoors. In this instance, the weather is rather humid and warm in these regions, but the time is not long enough for *M. Faeni* to grow in the stacks, which might be the reason why the percentage of positive serological reaction against *M. faeni* was very low in the patients in that county.

It was reported that the reactions against different strains of *T. vulgaris* in the same group of patients with farmer's lung might be significantly different from each other, and similar results could be found from different strains of *T. candidus*.<sup>8,9,10</sup> In our study, it was also found that the reactions against six strains of *T. vulgaris* in the patients with farmer's

lung ranged widely from 13.3 to 80.0%. These findings may indicate that different strains of *T. vulgaris* could have different antigens. Therefore, a variety of strains of *T. vulgaris* should be used to test precipitins in serum from the patients with farmer's lung.

In addition, the reactions against the extracts from Thermophilic nocardia and mouldy hay in the patients was found not significant in this study.

In conclusion, it may be said that the etiological agents of farmer's lung in Dafeng County were mainly from some strains of *T. vulgaris*, but not *M. faeni*, and different strains of *T. vulgaris* should be applied to detect precipitins in serum diagnosis of farmer's lung.

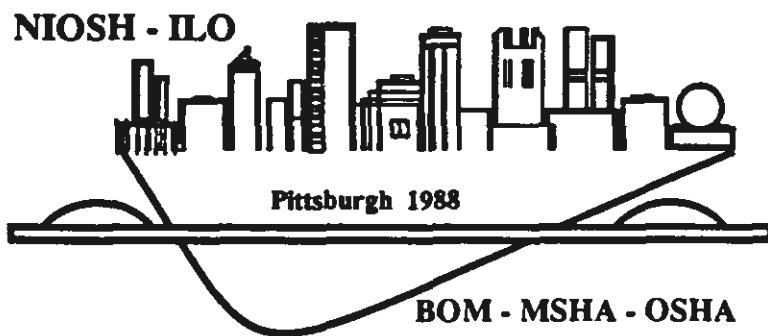
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