

## Histopathology of lung complications in 147 AIDS/HIV infected Thai patients

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- Background** : *Lung involvement is a major source of morbidity and mortality in AIDS/HIV patients. Unfortunately, there has been a lack of information on histopathologic features of lung complications in AIDS/HIV infected Thai patients.*
- Objective** : *To study histopathologic features of lung complications in AIDS/HIV infected Thai patients, and to search for unknown.*
- Setting** : *Chest Disease Institute, the largest national referral center for the treatment of heart and lung diseases in Thailand.*
- Research Design** : *Retrospective review*
- Materials and Methods** : *Microscopic examination of transbronchial lung biopsy (TBLB) slides from AIDS/HIV infected Thai patients who had lung lesions. Tissue slides were stained with Hematoxylain and Eosin (H&E), Ziehl-Neelsen stain for acid-fast bacilli and Gomeri's Methenamine Silver.*
- Results** : *A wide array of histopathologic diagnoses in this study is shown. Interstitial fibrosis (IF) and/or interstitial pneumonitis (IP) were the most common histopathologic diagnosis and were seen in 137 patients (93.20%). Organism-specific diagnoses were made in 77 patients (52.38%). No organism was identified in 68 patients (46.26%). Bronchogenic carcinoma was diagnosed in 2 patients (1.36%).*

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**Conclusions** : *A wide spectrum of lung complications in AIDS/HIV patients is observed in Thailand but with some statistically difference when compared to the Western reports.*

**Keywords** : *HIV, AIDS, Lung pathology, Lung complication, Infection.*

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- เหตุผลของการทำวิจัย** : ข้อมูลที่เกี่ยวข้องกับพยาธิสภาพของภาวะแทรกซ้อนในปอดยังไม่มีโดยเฉพาะในผู้ป่วยคนไทยที่ติดเชื้อ ทั้ง ๆ ที่ภาวะแทรกซ้อนในปอดเป็นสาเหตุสำคัญของการทำให้ผู้ป่วยติดเชื้อ AIDS/HIV ป่วยมากขึ้นรวมทั้งเสียชีวิตได้
- เป้าหมาย/วัตถุประสงค์** : ตรวจสอบเนื้อเยื่อเพื่อศึกษาลักษณะพยาธิสภาพของภาวะแทรกซ้อนในปอดในผู้ป่วยคนไทยที่ติดเชื้อ AIDS/HIV และเพื่อแสวงหาในสิ่งที่ยังไม่รู้
- ประเภทโรงพยาบาล** : สถาบันโรคทรวงอกเป็นศูนย์ตติยภูมิที่ให้การรักษาผู้ป่วยโรคหัวใจและโรคปอดที่ใหญ่ที่สุดในประเทศไทย
- รูปแบบการวิจัย** : การตรวจชิ้นสุตรย้อนหลัง
- สิ่งตรวจและวิธีการทำวิจัย** : ใช้กล้องจุลทรรศน์ตรวจชิ้นเนื้อปอดที่ได้มาจากคนไข้ติดเชื้อ ซึ่งมีรอยโรคในปอด สไลด์ได้ถูกย้อมสี Hematoxylain & Eosin (H&E), Ziehl-Neelsen stain และ Gomeri's Methenamine Silver
- ผลการศึกษา** : พยาธิสภาพที่พบบ่อยที่สุดคือ Interstitial fibrosis (IF) และ/หรือ interstitial pneumonitis (IP) ซึ่งพบได้ 137 ราย (93.20 %) สามารถให้การวินิจฉัยและบ่งบอกเชื้อโรคที่เป็นต้นเหตุได้ 77 ราย (52.38 %) ไม่พบเชื้อโรคที่เป็นต้นเหตุ 68 ราย (46.26 %) ส่วนอีก 2 รายพบว่าเป็น Bronchogenic Carcinoma
- สรุป** : ได้พบความหลากหลายของภาวะแทรกซ้อนในปอดในผู้ป่วยคนไทยที่ติดเชื้อ AIDS/HIV แต่ทว่ามีความแตกต่างกันอยู่บ้างทางสถิติเมื่อเปรียบเทียบกับรายงานของประเทศแถบตะวันตก
- คำสำคัญ / ดัชนีเรื่อง** : เอชไอวี, เอดส์, พยาธิสภาพในปอด, ภาวะแทรกซ้อนในปอด, การติดเชื้อ

Only 25 years ago, we believed that we were winning the battle against infectious disease. Instead, we are losing ground with the appearance of new diseases ranging from AIDS to Ebola and the re-emerging of Tuberculosis (TB) throughout the world.<sup>(1)</sup> According to the World Health Organization (WHO), infectious diseases are the most common cause of death worldwide. AIDS/HIV is an incurable disease and one of the most virulent diseases scourging humanity. It creates catastrophic effects on TB situation in Thailand. Because of its asymptomatic nature and the general public unwillingness to accept the existence of the disease or to take adequate protective measures, HIV has spread efficiently and silently to involve more than a million Thai people. AIDS/HIV pandemic remains virtually unchecked in the many provinces throughout the country.

In the past two decades, evidence has accumulated that lung is the major target of involvement by many infectious diseases, neoplastic and other conditions in AIDS/HIV patients worldwide. Over 80 % of AIDS patients developed lung complications during their disease.<sup>(2, 3)</sup> Data from autopsy studies indicated lung lesions to be up to 90 % of AIDS patients.<sup>(4, 5)</sup> Since lung is a vital organ, its involvement is a major source of morbidity and mortality. Moreover, failure to accurately diagnose lung infection implies that the opportunity for reducing transmission of HIV, TB and other lung infections has been lost.

Of all these years, a great deal of information on clinical manifestations, diagnosis, management, microbiology, immunology of these lung complications has become available, mostly from the United States.

However, data regarding the histopathology of lung lesions are limited. In Thailand, there is much less information on all aspects of lung complications. This research is pursued in response to the demand for more information on pulmonary histopathologic features of AIDS in Thai patients.

### Materials and Methods

A retrospective review was conducted at Pathology Department, Chest Disease Institute, Department of Medical Services, Ministry of Public Health. Chest Disease Institute is a 500-bed tertiary care center located in Bangkok Metropolitan. It is the largest national referral center for the treatment of heart and lung diseases in Thailand. AIDS/HIV Thai patients who underwent fiberoptic bronchoscopy with transbronchial lung biopsy (TBLB) and/or bronchial biopsy (BB) were identified from our surgical pathology records from the period of January 1, 1993 to December 31, 1998. None of these patients received antiretroviral drugs. The above mentioned procedures were performed by chest physicians as a part of diagnostic modalities for lung lesions in AIDS patients. One to six biopsies were obtained from each patient.

All biopsies were processed in tissue processor and multiple serial sections of each patient specimen were routinely prepared for hematoxylin and eosin (H&E) stain and special histochemical stain including Ziehl-Neelsen stain for acid fast bacilli (AFB) and Gomori's Methenamine Silver (GMS) stain. All slides of H&E stain, AFB and GMS stains of TBLB and BB of AIDS patients were pulled out for a microscopic examination. In addition to a bright light microscopic examination, all H&E slides were examined under polarized light.

**Results**

The total number of 296 H&E slides, 296 AFB stained slides, and 296 GMS stained slides from 158 AIDS/HIV patients was available for a review. Eighteen biopsies of 11 patients were considered inadequate due to the following conditions: lack of lung parenchyma, too small of specimens, severe crush artifact, or otherwise non-diagnostic material. These cases were excluded from analysis in this study. The total of 278 biopsies of 147 AIDS/HIV patients was used for analysis. There were 137 males and 10 females with a male to female ratio of 13.7: 10 and age ranged from 19 to 66 years. The clinical data are shown in Table 1.

Since we dealt with infectious diseases, attempts were made to provide organism-specific diagnoses in addition to general pathological diagnoses. A wide array of histopathologic diagnoses in this study are shown in Table 2.

*Pneumocystis carinii* Pneumonia (PCP) was diagnosed in 51 patients (34.69 %). The co-infection of PCP and TB was found in 2 patients (1.36 %). The co-infection of PCP and CMV was found in one patient (0.68 %). Interstitial fibrosis (IF) and/or interstitial pneumonitis (IP) with intra-alveolar exudate were seen in 46 cases. An additional lesion of diffused alveolar damage (DAD) with hyaline membrane was found in 10 out of 46 cases (6.80 %). PCP organisms were confirmed with GMS stain in 40 out of 46 cases (27.21%) with these features. The other 6 cases showed no organism on GMS stain because the tissue in the paraffin block was exhausted after repeated cuttings. These 6 cases were diagnosed as suggestive of PCP. One out of these 6 cases was diagnosed as suggestive of co-infection of PCP and TB. The 5 additional cases of IF and/or IP without a typical intra-alveolar foamy exudate showed PCP organisms on GMS stain. In conclusion, the total of 45 cases of PCP

**Table 1.** Clinical Data.

	Number of AIDS Patient	Percentage
Age range 19 to 66 years		
Mean Age 33 years		
Age Group		
• Under 20 years	1	0.68 %
• 20 – 29 years	52	35.37 %
• 30 – 39 years	62	42.18 %
• 40 – 49 years	25	17.01 %
• 50 – 59 years	4	2.72 %
• 60 – 69 years	3	2.04 %
Total	147	100.00 %
Male	137	93.20 %
Female	10	6.80 %
Male / Female Ratio	137/10	13.70 / 1.0

was confirmed with the identification of organisms on GMS stain.

Tuberculosis (TB) or other mycobacterial infection was diagnosed in 5 patients (3.40 %) based on an identification of acid fast bacilli on Ziehl-Neelsen stain. Histopathologic findings in these 5 cases include one IF and IP, 2 granuloma, and 2 granulation tissue with necrosis, compatible with suppurative inflammation. One out of the 5 cases was a case of co-infection of TB and PCP. Another case of granuloma

with histologic features of PCP showed no organism on special stain. Thus, the diagnosis of suggestive of TB and PCP was made in this case. An additional of 6 cases showed granuloma and suppurative-granulomatous lesions but no organism was identified on special stain. The diagnosis of suggestive of TB or other mycobacterial infection was made in these 6 patients. Other lung complications are self-explanatory as shown in Table 2.

**Table 2.** Final Histopathologic Diagnoses.

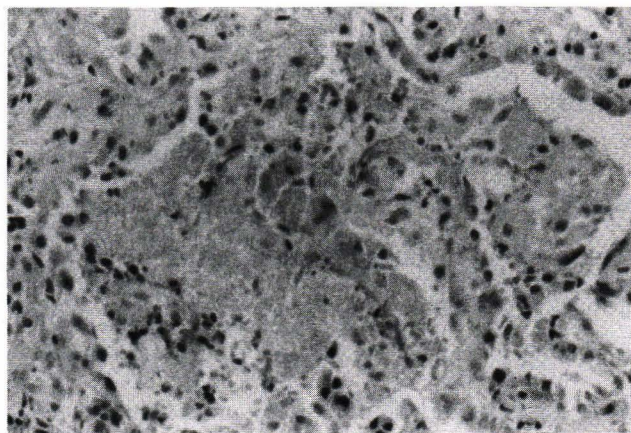
	Number of AIDS Patient (N = 147)	Percentage
I. IF and/or IP with organism-specific diagnoses	77	52.38 %
1. <i>Pneumocystis carinii</i> pneumonia (PCP)	43	29.26 %
2. Suggestive of PCP	5	3.40 %
3. Tuberculosis (TB) or other mycobacterial infection	4	2.72 %
4. TB or other mycobacterial infection and PCP	1	0.68 %
5. Suggestive of TB and PCP	1	0.68 %
6. Granuloma and Suppurative-granulomatous lesion, Suggestive of TB or other mycobacterial infection	6	4.08 %
7. Cytomegalovirus (CMV)	9	6.12 %
8. Cryptococcosis	4	2.72 %
9. Histoplasmosis	2	1.36 %
10. Penicillosis ( <i>Penicillium marneffe</i> )	1	0.68 %
11. CMV and PCP	1	0.68 %
II. General Pathologic Diagnoses	70	47.62 %
1. IF and IP	37	25.17 %
2. IF alone	20	13.61 %
3. IP alone	3	2.04 %
4. Suppurative Inflammation	4	2.72 %
5. Non-Specific Inflammation	4	2.72 %
6. Bronchogenic Carcinoma	2	1.36 %

## Discussion

The patients in this study do not represent lung complications of AIDS/HIV in general but rather represent the complex and non-diagnostic cases following routine clinical investigation. In Thailand, management of AIDS/HIV patients with lung complications is oftentimes based on preemptive diagnosis. Although, TBLB is the most common procedure for morphologic diagnosis of lung lesions in AIDS/HIV patients, it is not routinely performed in all of them. TBLB was done only in the difficult and recalcitrant cases. This study revealed a wide array of pulmonary histopathologic changes in the complex AIDS cases of Thai population.

In this study, IF and/or IP were diagnosed in 137 patients (93.20 %) and constituted the most common histopathologic diagnoses. Organism-specific diagnoses were made only in 77 patients (52.38 %). Etiological diagnosis cannot be established in 68 patients (46.26 %). This could be due to the fact that some of these patients had received previous treatment. Moreover, the lung generally responds to various injuries or insulting agents in similar fashion regardless of its etiologic agents. Therefore, ancillary tests beyond histologic evaluation are usually required to discover causative organisms. Unfortunately, supplementary tests such as cultures from tissue biopsies, electron microscopy, immunohistochemical stain, immunofluorescence and molecular diagnostic tests were not available in our institute. We believe these sophisticated tests would increase the detection rate in some cases.

PCP (Figure 1.) was the first extensively reported major opportunistic infection in the history of AIDS epidemic. In fact, the first 3 documented



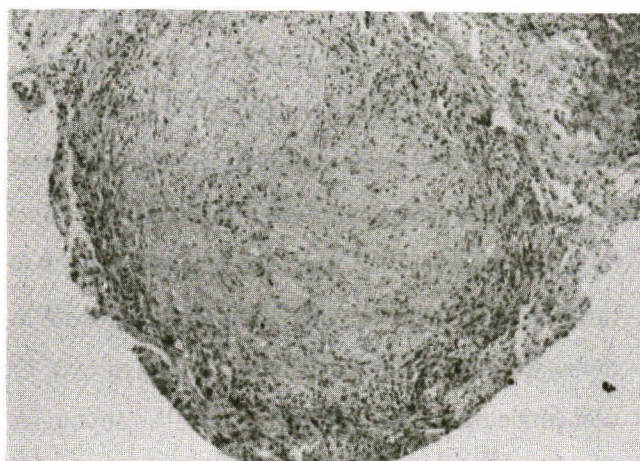
**Figure 1.** PCP with characteristic foamy exudate in alveolar spaces. The interstitium shows inflammatory infiltrate and fibrosis. The diagnosis is confirmed with GMS stain, (H&E stain X 200).

AIDS cases died of PCP. Reports from the United States indicated PCP was the most common lung infection in AIDS patients and was found up to 80 % of them.<sup>(6-8)</sup> In this study, PCP was the most common organism-specific diagnosis and was found in 51 patients (34.69 %). IF and/or IP with eosinophilic foamy exudate in alveoli were the most common histopathologic changes seen in PCP in this study.

PCP was originally considered a protozoon but has recently been classified as fungus based on sequencing analysis of ribosomal RNA and mitochondrial DNA.<sup>(9,10)</sup> Just like other fungi, PCP is best demonstrated on GMS stain. They present in alveolar exudate as thin-walled, round to oval or crescentic cysts within which blue dot-like structures (sporozoites) are visible. Other features such as granulomatous inflammation, necrotizing pneumonia with cavitation and lymphocytic interstitial pneumonitis (LIP) like lesion have been described in AIDS with

PCP as uncommon histopathological changes.<sup>(11-14)</sup> However, these changes were not seen in our study. Vasculitis with or without parenchymal necrosis, a rare feature of PCP was not found in this study either.

TB (Figure 2.) is currently the leading cause of death in AIDS/HIV infected patients worldwide.<sup>(15)</sup> It was found to be the primary cause of death in 32 % of AIDS/HIV patients in an autopsy study in a West African City.<sup>(16)</sup> About a quarter to a third of AIDS patients presented with pulmonary infection with TB and non-tuberculous mycobacteria.<sup>(17-19)</sup> In this study, pulmonary TB and/or other mycobacterial infections were diagnosed in 12 patients (8.16 %). This indicates that clinical diagnosis of TB in AIDS/HIV patients is sometimes difficult to make despite a widespread recognition that TB is often a feature of AIDS. Clinical diagnosis of TB was made based on positive AFB on sputum smear or culture and an abnormal Chest X-rays. Apparently, these routine laboratory tests failed to diagnose TB in almost 10 % of recalcitrant cases. Histopathologic features in our studies included



**Figure 2.** Tuberculosis. TBLB shows a non-caseating granuloma. The diagnosis is confirmed with Ziehl-Neelsen stain for AFB, (H&E stain X100).

granulomas, suppurative-granulomatous inflammation, and acute inflammation with necrosis, mostly compatible with necrotizing pneumonitis. Inflammatory pseudotumor, an extremely rare feature of mycobacterial infection was not found in this study.

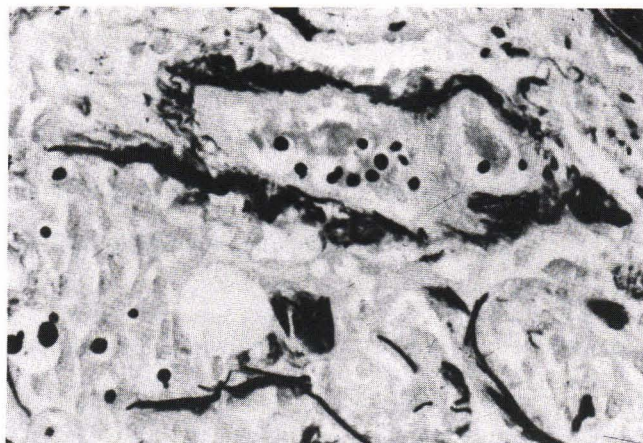
Cytomegalovirus (CMV) is the most common viral infection in AIDS/HIV patients.<sup>(20)</sup> While there is a controversy regarding the pathogenic role of this organism, it is reasonable to consider it a true infection whenever the cytopathic changes are seen in tissue biopsy. One thing to be mentioned, a positive culture of CMV from the respiratory tract is not diagnostic for CMV infection since this organism is commonly isolated from many tissues, organs and body fluids. Furthermore, CMV seropositivity does not indicate current infection. It indicates a previous exposure and can be detected in 40 to 100% of the world's adult population. In this study, the diagnosis of CMV pneumonia was made based on the findings of enlarged cells with intranuclear and/or intracytoplasmic inclusions.

Cryptococcosis (Figure 3.) is the second most common cause of fungal pneumonia in AIDS/HIV patients. IF and IP with Cryptococcal yeasts were seen in all four cases of Cryptococcal pneumonia in our study. This concurred with the western study that IF was the most common histopathologic finding of Cryptococcal pneumonia in AIDS/HIV patients.<sup>(21)</sup>

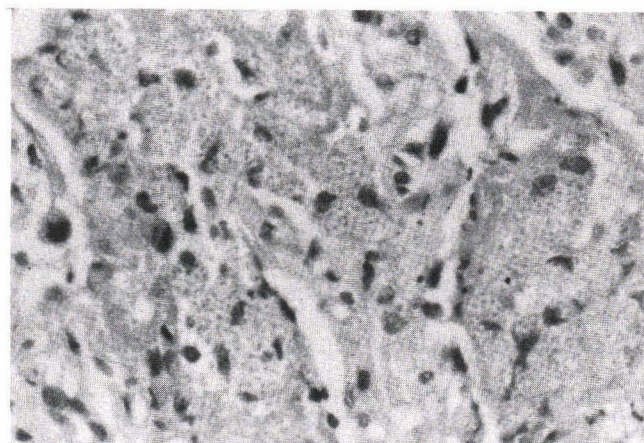
Histoplasma pneumonia (Figure 4.), an uncommon lung infection in Thailand was diagnosed in two cases. Histopathologic features of Histoplasmosis included one case of IF and IP and another case of IF and IP with a few tiny granulomas.

*Penicillium marneffe* (Figure 5.) has almost never been seen in the United States but was reported



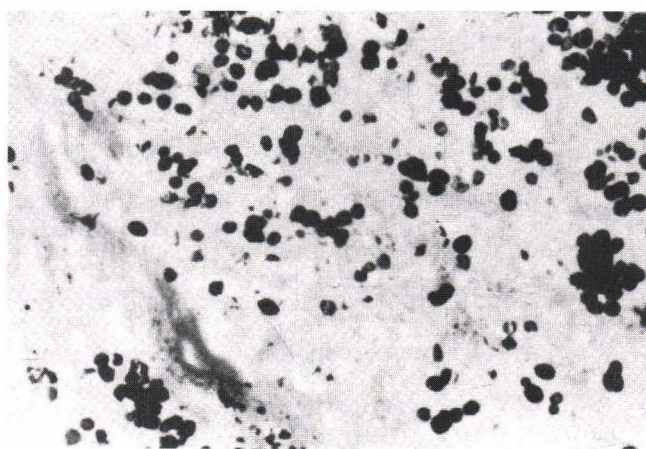


**Figure 3.** Cryptococcal pneumonia. Yeasts are present within the alveoli and interstitium of lung. They are round to oval and vary considerably in size. Some form a single bud. The capsule is seen as a clear halo with GMS stain, (GMS stain X 400).



**Figure 4.** Histoplasmosis. TBLB of an AIDS patient shows IP, IF and numerous yeasts of *Histoplasma* in alveoli and interstitium, (H&E stain X 400).

as the third most common infection of AIDS/HIV patients at Maharaj Nakorn Chiangmai Hospital, Thailand. Therefore, Penicillosis was included in Thai clinical criteria of AIDS and in Communicable Diseases



**Figure 5.** Penicillosis. Numerous yeasts of *Penicillium marneffeii* are seen in TBLB. They can be differentiated from *Histoplasma* or PCP by the presence of a typical clear zone at the center on GMS stain, (GMS stain X 600).

Control (CDC)'s case definition of AIDS.<sup>(22)</sup> We detected only one case of Penicillosis in this study.

The four cases of suppurative inflammation were most compatible with pyogenic bacterial pneumonia. The bacteria responsible for it could be *Pseudomonas*, *E. coli*, *Klebsiella*, *Enterobacter*, *Streptococcus* or *Staphylococcus*. Unfortunately, facility for culture of tissue specimen was not available. Therefore, the causative agents could not be identified in these cases.

Kaposi's sarcoma and malignant lymphoma are the two most common neoplastic complications of AIDS in the industrial countries.<sup>(23, 24)</sup> However, they were not seen in our study. Instead, two cases of bronchogenic carcinoma were diagnosed in our study. One of them was a case of moderately differentiated squamous cell carcinoma and another case of non-small cell carcinoma. More research is required and hopefully will shed more light on the

epidemiology, pathogenesis, and treatment of HIV associated neoplasms.

### Conclusions

As has been reported in the West, a wide spectrum of lung complications in AIDS/HIV patients is also observed in Thailand. Differences in patient population and geographic distribution and the application of sophisticated diagnostic techniques result in statistical variations regarding the incident of these illnesses.

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