Laboratory Diagnosis of HIV-Associated Infections

Report of an Intercountry Workshop
Bangkok, Thailand, 14-25 July 2003

WHO Project: ICP HIV 001 XW.02

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1. INTRODUCTION

HIV/AIDS has emerged as a major public health problem globally. It has already acquired considerable magnitude in the SEA Region with an estimated 6 million persons carrying the virus, with projections for future not showing any early respite from the pandemic. In the days to come, larger number of people will carry this virus and also present to health care facilities with myriad clinical manifestations, mostly based upon infections by a large number of microorganisms.

Proper management of the patient is one of the important strategies being implemented to mitigate the impact of HIV. The impairment of immune response in the patients makes them susceptible to infection and disease by conventional pathogens as well as by opportunistic microorganisms which are otherwise considered harmless. Microorganisms such as atypical mycobacteria, *Pneumocystis carinii*, *Cryptococcus*, *Cryptosporidium*, *Candida* etc belong to this group. Since these were not associated with human infections till the advent of HIV, the infrastructure and expertise to isolate and identify these are limited in the Region. Management of opportunistic infections is one of the mainstays for improving the quality and longevity of life. Infections due to opportunistic infections can be diagnosed, monitored and managed, provided good laboratory practices and processes are followed.

Realizing the acute need for upgrading the capacity of the Member Countries in establishing accurate diagnosis of these opportunistic infections to support HIV management strategies, a ten-day workshop for microbiologists from the SEA Region was organized at Bangkok, Thailand from 14 - 25 July 2003. Infrastructure and expertise for various HIV-associated infections are available in two different institutes in Bangkok and hence participants were trained for the first week in Siriraj Hospital and at Bumrasnaradura Institute during the second week of the workshop. Apart from the faculty of these two institutes, two experts from India facilitated the workshop. Eighteen participants from Bangladesh, India, Indonesia, Myanmar, Nepal and Sri Lanka attended this workshop. The list of participants and detailed programme can be seen at Annexes 1 and 2.
2. OBJECTIVES

The objectives of the workshop were as follows:

(1) To train the participants in diagnosing opportunistic infections in various clinical specimens obtained from patients with HIV/AIDS with emphasis on good laboratory practices;
(2) To identify country needs for WHO technical and material support in laboratory diagnosis;
(3) To develop strategies and plan for further training at the national level, and
(4) To promote networking of laboratories.

3. INAUGURAL SESSION

The Workshop was inaugurated by Dr Chantapong Wasi, Head of Virology, Siriraj Hospital, Bangkok. She emphasized the importance of diagnosis of HIV-associated infections in the rapidly emerging epidemiology of HIV. Dr Rajesh Bhatia, STP/BCT, WHO/SEARO presented the objectives and expected outcome of the workshop. He also deliberated upon the role of WHO in advocating strengthening of laboratories for management of HIV infection as well as HIV-associated infections in the countries of the SEA Region.

4. PROCEEDINGS

The main aim of the workshop was to provide hands-on training to the participants in updated standard microbiological techniques. These included various internal quality control measures, biosafety activities and acquaintance with external quality assessment schemes (EQAS). After brief presentations, extensive practicals were performed by the participants. The practical training included direct demonstration of the causative agents of HIV-associated infection by various microscopic methods, their culture and final identification tests. Serological techniques to facilitate diagnosis of some of these agents and use of modern molecular tools including PCR techniques and flow cytometry were also demonstrated.
Every practical activity was followed by an interactive session between the faculty and the trainees in which the problems related to the practicals were solved. Participants were also asked to solve some of the problems that might come up in their daily laboratory tasks. In addition, the country-specific availability of infrastructure and skills and measures that they could initiate to strengthen the same, including the training of various categories of staff in laboratory aspects of HIV-associated infections were discussed with the participants. They were provided with adequate technical material in electronic and hard copy form (Model standard operating procedure (SOP) developed by SEARO, Guidelines on biosafety). The presentations made in the workshop were compiled into a compact disc and provided to the participants to use as reference as well as training material in their respective countries.

Since a variety of infections can be grouped as HIV-associated infections, facilities for diagnosis of these may not be available in one institute. Instead of creating fresh infrastructure, the participants discussed the mechanism of networking in their own countries. The organizers of the workshop assured all possible technical support in the endeavours of the participants in establishing diagnostic facilities and providing reference facilities.

5. RECOMMENDATIONS

To Member Countries

(1) The knowledge and experience gained from the workshop in improving infrastructure and skills in diagnosis of HIV-associated infections should be utilized gainfully in capacity building for management of HIV-associated infections.

(2) All laboratories should strive to implement an efficient quality system, so that diagnosis of HIV-associated infections is reliable and matches with the needs of the users.

(3) Siriraj Hospital and Bumrasnaradura Institute, Bangkok, Thailand may be approached for any technical support and trouble shooting in establishing quality diagnostic services for HIV-associated infections.

To WHO
WHO should continue to provide technical support to all Member Countries in developing and strengthening infrastructure and skills in laboratory diagnosis of HIV-associated infections.
Annex 1

LIST OF PARTICIPANTS

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Short Term Professional-Blood Safety and Clinical Technology  
WHO, SEARO, New Delhi
Annex 2

PROGRAMME

Monday, 14 July 2003

0830-0900 hrs  Registration
0900-0915 hrs  Course Introduction
0915-1015 hrs  Introduction – Lectures, Instructors and Participants
1045-1215 hrs  Lecture: Overview of Common Opportunistic Infections in HIV Disease / AIDS – Somsith
1315-1430 hrs  Biosafety – Suda
145-1630 hrs  Good Laboratory Practice - Sasiwimon

Tuesday, 15 July 2003

0830-0930 hrs  Biology of mycobacteria – Angkana
0930-1030 hrs  Recent laboratory diagnosis of infection – Dr Paramsivam
1100-1600 hrs  ➢ Laboratory – Angkana and Dr Paramsivam
                ➢ AFB Staining – J. Jearanaisilawong
                ➢ Microscopy and reporting
1400-1500 hrs  Drug resistance in TB – Dr Paramasivam
1500-1700 hrs  ➢ Demonstrations of
                - Direct susceptibility test
                - Indirect susceptibility test
                - Biochemical tests
                - PCR and PCR / REA
                  N. Tingtoy and Dr Paramsivam
                ➢ Questions and Answers – T. Sengna,
                  P.Boonruengsri, D.Techamahapan and Dr Paramsivam
Wednesday, 16 July 2003

0830-0930 hrs  Nocardia and nocardiosis – Nattewan Poonwan
0930-1030 hrs  Candidosis and cryptococcosis – Suporn Foongladda
1100-1200 hrs  
- Laboratory – Angkana Chaiprasert
- Demonstrations of
  - AFB slides
  - Samples slides
  - Colonies
  - Candi/Select plate
  - Sugar Fermentation tests
  - Microdilution susceptibility test
S. Foongladda, P. Ngamskulroongroj, J. Jearanaisilawong, N. Poonwan

1300-1400 hrs  Dimorphic fungal infections – Ariya Jindamporn
1400-1600 hrs  
- Laboratory – Angkana Chaiprasert & team
- Demonstrations of
  - Samples slides
  - Colonies
  - Slide cultures
  - Posters
- Questions and Answers

Thursday, 17 July 2003

0900-1200 hrs  
- Lecture : Investigation in HIV Diagnosis & Monitoring – Suda, Sirirat
- Lab: HIV Serology – Suda, Sirirat & staff

1300-1630 hrs  Viral Infections (Lecture & Lab) – Prasert A, Sontana & staff

Friday, 18 July 2003

0900-1030 hrs  Lecture : Principles of Molecular diagnosis and PCR – Prasert
1030-1100 hrs  Demon : Sample preparation, nucleic acid extraction – Prasert
1100-1200 hrs  Lecture & Demon : HIV–PCR and viral load – Wasun
1300-1430 hrs  Lecture & Demon : Molecular diagnosis for PCP, TB, EBV – Wasun
1430-1700 hrs  Discussion

**Monday, 21 July 2003**

**Bumrasnaradura Institute**

0900-1630 hrs  Bacterial Infections (Lecture & Lab) – Boonchuoy & staff
1030-1100 hrs  Demon : Sample preparation, nucleic acid extraction – Prasert

**Tuesday, 22 July 2003**

0900-1200 hrs  ➢ Overview of parasitic infections in HIV/AIDS – Prof Samantaray
               ➢ Parasitic Infections (Lecture & Lab) – Boonchuoy, Juthatip & staff
1300-1630 hrs  Exercise Unknown Clinical specimens – Boonchuoy, staff and Prof Samantaray

**Wednesday, 23 July 2003**

0900-1200 hrs  Laboratory Exercise – Local staff and Dr Samantaray

**Thursday, 24 July 2003**

0900-1200 hrs  Immunophenotyping, Flow Cytometry – Kovit, Sirirat & staff
1300-1630 hrs  HIV Diagnosis and Monitoring at Bumrasnaradura Institute – Sirirat & staff

**Friday, 25 July 2003**

0900-1000 hrs  Post Test
1000-1400 hrs  General Discussion, Educational Material for Take Home
1400-1500 hrs  Conclusion, Certification Conferment