



8. Pharmacologic and Pharmacodynamic Animal Studies in Support of Mirinca-mycin Development. Status: bio-availability and tolerability study in primates completed. Development of further work suspended by IPT at WRAIR due to ineffective treatment of relapsing *P. cynomolgi* malaria.

9. Evaluation of Avian Influenza Hemagglutinin Sequences in Wild Birds. Status: Data analyzed and paper submitted for publication.

10. Kwai River Christian Hospital Surveillance of Influenza-like Illness. Status: New protocol submitted to Thai MOPH IRB by Virology.

11. Human Influenza Sentinel Surveillance in Cambodia. Status: Study initiated at 1 site and 1 sub-sentinel site.

12. Evaluation of *In Vitro* Cross-Reactivity with Avian Influenza H5N1 Virus in Healthy Volunteers Vaccinated with a Prime Boost Regimen of Seasonal Influenza Vaccine. Status: *in vivo* part completed. Lab assays in final stages.

13. Retrospective Survey of Severe Malaria in Battambang Referral Hospital, Cambodia, from 2006 to 2008. Status: manuscript in preparation.

14. An Active Malaria Epidemiology Cohort Study in Personnel and Dependents of the Royal Cambodian Armed Forces with Evaluation of a 2 Day Versus 3 Day Treatment 7 Regimen of DHA-Piperaquine for Patients with Uncomplicated Malaria. Status: enrollment completed.

15. Detection and Quantification of *Plasmodium* spp. by 18S rRNA Gene Subunit-Based and Species-specific Real-time PCR Assays. Status: protocol awaiting approval.

16. Leptospira (LPS assay) RAPID PCR Validation Using JBAIDS Molecular Assay Transition Package. Status: study ongoing

17. Malaria Drug Resistance Surveillance in Royal Thai Army Personnel in Thailand. Status: protocol finalized

DEPARTMENT OF RETROVIROLOGY

DEPARTMENT MISSION

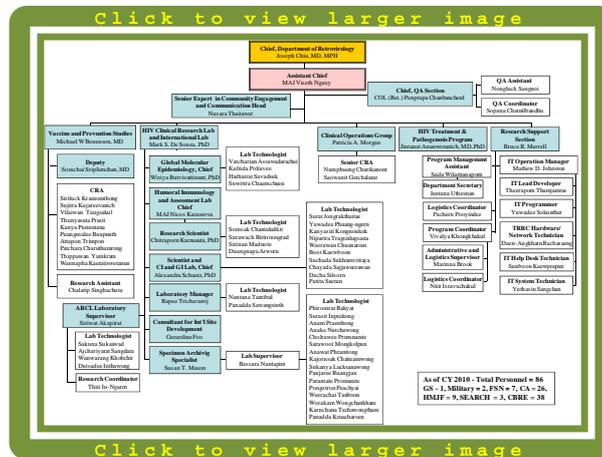
The mission of the Department of Retrovirology is to prepare for and conduct advanced development of preventive HIV vaccines for soldiers. This mission is achieved collaboratively and supported through i) the performance of preclinical and clinical (phase I-III) trials of candidate vaccines and their evaluations for safety, immunogenicity and efficacy, ii) the identification and characterization of potential cohorts for phase III vaccine trials, iii) the establishment of diagnostic assays which differentiate infection from vaccine-induced immune responses, iv) the characterization of HIV viruses circulating in the region, v) the determination of the natural history of HIV infection and disease.

PERSONNEL

The Department of Retrovirology consists of 86 staff that includes 1 GS and 2 Active-Duty Army Officers (1 Medical Corps, 1 Medical Service Corps). Within the department, there are 21 clinical research (4 MDs) and quality assurance staff, 42 laboratory personnel (5 PhD staff scientists), and 13 logistics/IT support staff. An overview of the organization chart is provided below as well as listing of each departmental staff member. The Department of Retrovirology



has added personnel due to initiation of new MIDRP-funded activities and increased laboratory work load resulting from the recently completed Phase III trial that showed modest efficacy in preventing HIV infection.



IN-HOUSE TRAINING PROGRAMS AND OUTSIDE TRAINING OF PERSONNEL

In-House Training Programs Provided by AFRIMS

1. SOP Training

- CMD-AD-000-02 Preparing and Retiring AFRIMS Standard Operating Procedure
- MD-AD-001-01 AFRIMS SOP Distribution and Annual Review
- CMD-AD-002-01 Forms and Appendices
- CMD-AD-003-01 Contents, Maintenance, Review of Training Files
- CMD-AD-006-00 USAMC-AFRIMS Conductor Orchestrating Training Software

Operational Guidance

- CMD-AD-007-00 USAMC-AFRIMS Personnel Electronic Training Record

Management System Maintenance, Account Creation, and Training

2. Routine Safety and Occupational Health Training

- Fire Prevention, Protection, Report and Investigation
- Accident Illness and Complaints Reporting, Records and Investigations
- Harzard Communication Program
- Routine Prophylaxis and Screening
- Biosafety in Laboratory
- Laboratory Waste Management
- Chemical Safety
- Safety Equipments Usage
- Bloodborne Pathogens Exposure Control Plan
- Respiratory Protection Program
- Post-exposure Prophylaxis and Intervention

3. Non-Routine Safety and Occupational Health Training

- Animal Safety and Bloodborne Pathogens
- Facility Safety and Design and Equipment
- Decontamination and Management
- Regulatory Guidance and Risk Assessment (General and Biological)
- Biosecurity and Transportation



4. Miscellaneous Training
 - Tech Transfer Training
 - UV Meter Use
 - GCLP Seminar for Laboratory Management, Tim Stiles, Qualogy LTD, AFRIMS, Bangkok, 1 March 2010
 - Good Clinical Laboratory Practice (GCLP), Tim Stiles, Qualogy LTD, AFRIMS, Bangkok, 1 March and 3 March 2010
 - Advance Good Clinical Laboratory Practice (GCLP), Tim Stiles, Qualogy LTD, AFRIMS, Bangkok, 4-5 March 2010

In-House Training Programs Provided by Department

- Traditional Concepts of Community Role in Health Care, Introduction to Clinical Research, Research Ethics from Community Perspective, RV217, and Concepts of CAB. Dr. Somchai Sriplienchan and Nusara Thaitawat. All Seasons Hotel, Pattaya, Thailand. 28-29 January 2010.
- Good Participatory Practice (GPP) Guidelines for Biomedical HIV Prevention Trials (produced by UNAIDS and AVAC, 2007). Nusara Thaitawat. ECHO Center, Pattaya, Thailand. 17 February 2010.
- Stakeholders Definition. Nusara Thaitawat. ECHO Center, Pattaya, Thailand. 17 February 2010.
- HIV epidemiology, surveillance. USPACOM Disease Surveillance Workshop, Organizer: PACOM. MAJ Viseth Ngauy. Phnom Penh, Cambodia. 22-25 February 2010.
- Virus sequencing. USPACOM Disease Surveillance Workshop, Organizer: PACOM. Dr. Wiriya Rutvisuttinunt. Phnom Penh, Cambodia. 22-25 February 2010.
- Good Participatory Practice (GPP) Guidelines for Biomedical HIV Prevention Trials (produced by UNAIDS and AVAC 2007). Nusara Thaitawat. ECHO Center, Pattaya, Thailand. 23 February 2010.
- Quality Assurance Training. for Thai Red Cross staff, Organizer: Department of Retrovirology. COL (Ret.) Penprapa Chanbancherd, Sopana Chatnilbandhu, and Nongluck Sangnoi. AFRIMS, Bangkok. 24-25 February 2010.
- Community Engagement and Stakeholders. Nusara Thaitawat. ECHO Center, Pattaya, Thailand. 24 March 2010.
- Training on Good Clinical Laboratory Practices (GCLP): Organizer: Programs for HIV Prevention and Treatment (PHPT). COL (Ret.) Penprapa Chanbancherd, Sopana Chatnilbandhu, and Nongluck Sangnoi. Chiang Mai, Thailand. 1-2 April 2010.
- Vaccination with ALVAC and AIDSVAX to Prevent HIV Infection in Thailand. Special Seminar, Organizer: Emergent Infectious Disease Program, Duke-National University of Singapore. Dr. Joseph Chiu. Singapore. 15 April 2010.
- Immunopathogenesis of Acute HIV Infection and the RV254 Study. Dr. Jintanat. SEARCH TRC, AFRIMS, Bangkok. 20 April 2010.
- Training on Good Clinical Laboratory Practices (GCLP): Organizer: Programs for HIV Prevention and Treatment (PHPT). COL (Ret.) Penprapa Chanbancherd, Sopana Chatnilbandhu, and Nongluck Sangnoi. Bangkok. 29-30 April 2010.
- Laboratory Assessment for CAP Reaccreditation, Organizer: Department of Retrovirology. Siriwat Akapirat. CRC-WRP, Kericho, Kenya. 29 March-2 April 2010 and 3-7 May 2010.
- Laboratory Testing Training (Bhutan staff), Organizer: Department of Retrovirology. Rapee Trichavaroj and Nantana Tantibul. AFRIMS, Bangkok. 1-11 June 2010.



- Laboratory for early detection of HIV infection. PMK & AFRIMS Joint Symposium: Research Highlights on Infectious Disease. Dr. Mark de Souza. Phramongkutklo Hospital, Bangkok. 10 June 2010.
- Laboratory Testing Training (Laos staff), Organizer: Department of Retrovirology. Rapee Trichavaroj, Siriwat Akapirat, Wanwarang Khobchit, and Nantana Tantibul. AFRIMS, Bangkok. 29 June-1 July 2010.
- Review RV217 Protocol, Review of Protocol Modifications for Part B, New HIV Prevention Technologies (NPTs), Review Basics of Clinical Research, Review Basics of Research Ethics, and Good Participatory Practice 2nd edition (NGOs refresher 2010). Dr. Somchai Sriplienchan, Nusara Thaitawat, Patchara Charuthamrong, Wannapha Kaeratiswetanun, Thippawan Yamkram. Montien Hotel, Pattaya, Thailand. 6 August 2010.
- English Class (HON). HON House, Pattaya, Thailand. Patricia Morgan. 24 August 2010.
- RV217 and Communications Skills (outreach team). Nusara Thaitawat, Patchara Charuthamrong, Wannapha Kaeratiswetanun. Camelot Hotel, Pattaya, Thailand. 16 September 2010.
- Touring the Antibody World of HIV. Dr. Nicos Karasavva. AFRIMS, Bangkok. 18 October 2010.
- Good Participatory Practice (GPP) (feedback to finalized 2nd edition). Nusara Thaitawat. ECHO Center, Pattaya, Thailand. 23 November 2010.
- Multi Hybridization Assay Training (Mr. Jun Ryan C. Orbina, Philippines). Organizer: Department of Retrovirology, MERL, TRC, Bangkok. 24-30 October 2010.
- Quality Improvement Visit (Assessment), Organization: Department of Retrovirology. COL (Ret.) Penprapa Chanbancherd and Sopana Chatnilbandhu, TRC, Bangkok. 15 December 2010.

Outside Training

- Cosmetic Surgery, Dr. Sang Surushtawong, Professor of reconstruction and cosmetic surgery. Police Hospital, SISTERS Office, Thailand. 24 February 2010.
- The SOLID 4 System: A Revolutionary Advancement in the field of Genetic Analysis, Mr. Justin and Yu-Jen Chen. Swissotel Le Concord, Bangkok. 17 March 2010.
- Anal Diseases, Dr. Chitlada Utaipiboon, Care and Treatment and Laboratory Services Section, Global AIDS Program/Thailand, Thailand MOPH-US CDC Collaboration. Pattaya Municipality Health Office, Thailand. 31 March 2010.
- Conference/Seminar: Roche Molecular Scientific Days Update on Clinical Applications and Quality Control, Roche Diagnostic Thailand. Pattaya, Thailand. 1-3 April 2010.
- Full-length Sequencing of HIV from Plasma Training, Dr. Sodsai Tovanabutra, U.S. Military HIV Research Program. Rockville MD, U.S.A. 2 May-29 October 2010.
- HIV Full-length Amplification and Data Analysis, Dr. Sodsai Tovanabutra, US Military HIV Research Program. Rockville MD, U.S.A. 3-7 May 2010.
- The International Society for Laboratory Hematology (ISLH). Brighton, United Kingdom. 10-12 May 2010.
- Henry M. Jackson Foundation for the Advancement of Military Medicine Code of Ethics, On-line. 13 May 2010.
- Biomedical Investigators, Key Study Personnel, Medical Monitors (group 3), CITI Program online training. www.citiprogram.org. 27 May 2010.



- Joint Academic Conference and Research Cooperation Network, PMK & AFRIMS Joint Symposium-Research Highlights on Infectious Diseases. Phramongkutklo Hospital, Bangkok. 10 June 2010.
- Advanced Vaccinology Course, The National Vaccine Committee Office, Department of Disease Control, Ministry of Public Health. Radison Hotel, Bangkok. 15-17 June 2010.
- UNAIDS/AVAC Good Participatory Practice Guidelines for Biomedical HIV Prevention Trials, Deirdre Grant, Program Manager, AVAC, New York. ECHO Center, Pattaya, Thailand. 1 July 2010.
- New Employee Facility Safety Orientation, Online training, Henry M. Jackson Foundation. Rockville MD, U.S.A. 6 July 2010.
- AIDS 2010: XVIII International AIDS Conference, International AIDS Conference. Vienna, Austria. 18-23 July 2010.
- The 9th HIV/AIDS Workshop 2010. Thai AIDS Society, Phramongkutklo Hospital, Bangkok. 15-17 August 2010.
- Risk Communication Workshop, Keith Fulton and Sandy Martinez, AFRIMS-CDC. Century Park Hotel, Bangkok. 30 August-1 September 2010.
- Anti-retrovirals and female hormones using, Dr. Chitlada Utaipiboon and Dr. Akechitra Sukkul, Care and Treatment and Laboratory Services Section, Global AIDS Program/Thailand, Thailand MOPH-US CDC Collaboration, Health and Opportunity Network. Montien Hotel, Pattaya, Thailand. 24 September 2010.
- Life Technologies Sequencing Seminar Series: Genetics Exploring between CE Proven Technology, Justin and Chris. Grand Mercure Fortune, Bangkok. 1 November 2010.
- GPP feedback to finalize 2nd edition. Nusara Thaitawat, Novotel, Chiang Mai, Thailand. 9-10 November 2010.
- Good Participatory Practice Guidelines for Biomedical HIV Prevention Trials Second edition, 2010, Thai NGOs Coalition on AIDS. Ebina House Hotel, Bangkok. 12-13 November 2010.
- NEJM Review CME Program, The Massachusetts Medical Society, Program #362, Correspondence Course. 1 December 2010.
- iPREX (PrEP), WHO, CDC, AVAC, TNCA. Princeton Park Suites Hotel, Bangkok. 8-9 December 2010.
- The Process of Gut Biopsy Sample Collection, Isolation, and Flow Cytometry. Julie Elliott, Center for Prevention Research, UCLA, Los Angeles, U.S.A. 11-17 December 2010.

AWARDS

None

ACCOMPLISHMENTS

In 2010, the Department successfully passed the College of American Pathologist re-accreditation for the clinical laboratories. With the completion of the world's largest phase III HIV vaccine trial (RV144) conducted in Eastern seaboard of Thailand (Chon Buri and Rayong provinces) and the publication of the efficacy result in 2009, the protocol team's effort in 2010 shifted to laboratory analysis. In particular efforts, to characterize the HIV-1 viruses that caused infections and standard immunological analysis to determine what caused the protective effects (correlates of protection). Some of the preliminary findings are:



- HIV subtype E was detected in 59 of 64 individuals.
- High diversity was observed regardless of vaccine/placebo status.
- Breakthrough viruses were not divergent from vaccine sequences.
- CD4 responses to the env protein particularly the V2 region, were predominantly

IL-2+.

- Analysis revealed interferon-gamma production, CD107a-positive T-cells upon restimulation.
- Very low CD8 responses
- Relatively weak NAb response to tier 1 and 2 viruses
- IgG (1 and 3) and IgA responses to vaccine strain antigens were highest.
- IgG and IgA antibodies to HSV gD were detected in the majority of subjects and may minimize HIV infection risk in vaccines.
- Several potential humoral and cellular immunologic assays to assess correlates of protection have been qualified.

In the coming year, efforts will be focused on selecting the most promising and well qualified assay to conduct humoral, cellular and innate immunologic assessment to explore correlates of protection using a case-control design.

The collaborators are also planning clinical studies to extend and build on the modest success of RV 144. Results suggest that the vaccine regimen protected people at lower risk of infection, and the protection appeared to wane over time. One proposed study involves boosting some of the volunteers from RV144 to see if this will extend and increase the immune response (a “boost” is an additional vaccine dose given after the primary doses to increase the immune response to the original vaccine antigens).

Another proposed study will recruit several hundred new volunteers in Thailand who will receive a similar vaccine regimen as in RV144, plus an additional boost at 12 months. This study would provide insight into the benefit of the additional boost and collect more blood and some tissue samples so that extensive research can be conducted on the study participants’ immune responses.

RV152, a study of breakthrough infections in the Phase III trial (RV144) which began in May 2006 closed to enrollment in June 2009 with the completion of RV144. As of January 2011, 61 volunteers have reached study endpoint of CD4 less than 350 cells/ μ L. As is on-going as HIV-infected volunteers from the phase III trial are identified. Study results will be analyzed when approximately 66 primary composite endpoints have accumulated based on an updated statistical analysis plan. In accordance with the new 2010 National Treatment Guideline regarding initiation of anti-retroviral therapy, HIV infected individuals whose CD4 < 350 cells/ μ L are offered treatment. It is estimated that the final analysis will be June 2011.

RV158, the Phase I study of a CRF01_AE MVA vaccine which began on 27th November 2007 in Thailand enrolled twelve volunteers and was completed in November 2008. Laboratory work on samples collected in the clinical trial was conducted in FY 2009 and 2010. Cellular immunogenicity was assessed by a validated IFN γ Elispot assay, an intracellular cytokine staining assay, lymphocyte proliferation and a (51)Cr-release assay. Humoral immunogenicity was assessed by ADCC for gp120 and binding antibody ELISAs for gp120 and p24. MVA-CMDR was safe and well tolerated with no vaccine related serious adverse events. Cell-mediated immune responses were: (i) moderate in magnitude (median IFN γ Elispot of 78 SFC/10(6) PBMC at 10(8) pfu IM), but high in response rate (70% (51)Cr-release positive; 90% Elispot positive; 100% ICS positive, at 10(8) pfu IM); (ii) predominantly HIV Env-specific CD4(+) T cells, with a high proliferative capacity and durable for at least 6 months (100% LPA response rate by the



IM route); (iv) dose- and route-dependent with 10(8) pfu IM being the most immunogenic treatment. Binding antibodies against gp120 and p24 were detectable in all vaccination groups with ADCC capacity detectable at the highest dose (40% positive at 10(8) pfu IM). MVA-CMDR delivered both intramuscularly and intradermally was safe, well-tolerated and elicited durable cell-mediated and humoral immune responses. Volunteers were unblinded as to treatment allocation at both sites in Thailand. Study results were published in PLoS. There is continued work to further characterize the immune response elicited by the candidate vaccine.

Currier JR, Ngaoy V, de Souza MS, Ratto-Kim S, Cox JH, Polonis VR, Earl P, Moss B, Peel S, Slike B, Sriplienchan S, Thongcharoen P, Paris RM, Robb ML, Kim J, Michael NL, Marovich MA. Phase I safety and immunogenicity evaluation of MVA-CMDR, a multigenic, recombinant modified vaccinia Ankara-HIV-1 vaccine candidate. PLoS One. 2010 Nov 15;5(11): e13983.

RV212, a cross-sectional study to screen for and generate broadly neutralizing monoclonal antibodies from HIV infected individuals seeks to generate broadly neutralizing monoclonal antibodies (mAbs) from volunteers who are HIV infected and have broadly cross-reactive serum neutralizing activity. The protocol began enrollment in August 2007 and completed in July 2008. Only a few Thai volunteers were identified with broadly, cross-reactive antibodies. Isolation and production of monoclonal antibodies is ongoing. AFRIMS will conduct genotyping, antibody isolation and purification, and construct a CRF01_AE antibody pool for reagents.

In collaboration with the University of Hawaii and the Thai Red Cross AIDS Research Centre (SEARCH – Southeast Asia Research Collaboration with Hawaii), the Department is currently working on several protocols. RV233 is a HIV incidence cohort study in clients receiving anonymous HIV counseling and testing service. In addition the study investigated HIV subtype and genotypic resistance among HIV seroconverters. A total of 992 subjects, from 1,075 subjects screened, were enrolled between 1 August 2008-5 August 2009. Among 992 subjects enrolled, 32.8% were heterosexual men, 27.2% were men who have sex with men (MSM) and 33.5% were women (the remainder consisting of men with indeterminate sexual preferences and transgendered individuals). Baseline prevalent HIV infection was identified in 105 subjects (10.5%) at baseline visit, 4.9% of heterosexual men, 18.1% of MSM and 7.5% of women. There were 2 HIV seroconversion, both MSM, during the study period. This gave a calculated HIV incidence of 2.3 infections/100 person-years (95% confidence interval, CI, 0.6, 9.1) among MSM and the overall calculated HIV incidence of 0.56 infections/100 person-years (95% CI 0.14, 2.55). Due to the very low HIV seroconversion rate among an overall study participants along with the lower than expected follow-up rate, a decision was made to close the study early in August 2009. The last study visit occurred in December 2009. Data analysis and manuscript preparation is currently in progress.

RV254, another collaboration with SEARCH and the Thai Red Cross investigated the incidence, demographics, HIV subtype and genotypic resistance in acute HIV infection within a high-risk Thai cohort at the Thai Red Cross Anonymous Clinic (TRCAC), which has an HIV prevalence of about 17%. TRCAC uses 4th generation enzyme-linked immunoassay (AxSYM) for HIV diagnosis. AxSYM-negative samples are pooled to detect acute HIV infection by nucleic acid testing (NAT) using Roche Amplicor v1.5 ultrasensitive assays. Acute HIV infection samples were AxSYM-negative, NAT positive. Additional acute HIV infections were identified if the sample is AxSYM and NAT positive but first generation sensitive EIA (HIV-1 Microelisa System, Organon Teknika, Durham, NC). Demographic and risk behavior data from the TRCAC questionnaires were collected.



Between 20 April 2009 to 31 December 2010, 24,430 samples were prospectively screened to identify 40 subjects with AHI. Twenty-six were identified by pooled NAT (negative HIV IgM antibody – Fiebig I/II) and 14 were identified by sequential EIA (positive HIV IgM antibody – Fiebig III/IV). Thirty-three enrolled in the study. The AHI incidence was around 2/1000 screened. The staging of acute HIV infection was Fiebig I/II in 13 and the rest are Fiebig III/IV. Three were women and 30 were men. Risk factors were heterosexual transmission in 6 and homosexual transmission (men) in the rest. 84% had acute retroviral syndrome. The mean HIV RNA and CD4 at enrollment were 119, 325 copies/ml (min 1,202 - max 337,105) and 426 cells/mm³ (min 218 - max 740). HIV subtypes were CRF_01AE in 16, B in 4, CRF01_AE/B in 1 and pending results in 2. Three subjects who were partners had primary resistance to NNRTI. The majority of subjects agreed to optional procedures. Some subjects co-enrolled in a local treatment protocol in which they received either 5 drugs (megaHAART) or 3 drugs (HAART).

Three abstracts have been accepted for presentation at the Conference on Retroviruses and Opportunistic Infections in Boston in February 2011. Key findings to be reported are:

- Mega-HAART in early Fiebig stage AHI may prevent CD4 depletion of the sigmoid colon and render gut and peripheral HIV RNA undetectable.
- Mega-HAART may reduce viral burden and promote mucosal immune restoration, indicated by the increased frequency of CD4+CCR5+ T cells in the sigmoid colon and could be a crucial component of a functional cure.
- Acute HIV infection is associated with detectable virus in CSF as early as 5 days post exposure.
- CNS inflammatory markers are noted early as detected by CSF cytokines and MRS metabolites.
- The CSF cytokine pattern may differ from that in plasma during acute infection.
- In this population of clinic attendees, almost 2 out of every 1,000 were in AHI.
- Men with AHI had variable HIV RNA from undetectable to very high in SP and AL regardless of route of HIV acquisition.
- HIV RNA in SP, but not in AL, was higher in later stages of AHI. Similarity of primary resistance mutations suggests lack of compartmentalization during AHI.

This study will likely complete enrollment of 40 subjects by mid 2011. We are amending the protocol to increase to 75 subjects and to allow foreigners to enroll. In 2011, we will perform more immunologic and virologic analysis using the stored samples in order to understand what occurs in the different compartments very early in HIV infection. These tests will include immunophenotyping (blood, gut), immunohistochemistry (gut), viral load (blood, gut, CSF, genital secretion), HIV sequence (blood, gut, genital secretion) and cytokines and chemokines (blood, CSF).

The RV 243 protocol entitled “Assessment of neutralizing antibody (NAb) in participants from phase I/II Trials of ALVAC-HIV (vCP1521) priming with Chiron gp120 B/E, Sanofi-Pasteur oligomeric gp160, or AIDSVAX™ B/E gp120 B/E boosting against a newly developed, standardized panel of HIV-1 isolates”. This study aims to use the TZM-bl cell line Luciferase Reporter Pseudovirus NAb assay method. It will also aim to use archived plasma samples from previous phase I/II prime-boost HIV vaccine studies (RVs 132 and 135). The objectives of this protocol are (1) to compare cross-clade NAb among samples from HIV uninfected volunteers who have received prime-boost regimens of ALVAC vCP1521 and three different Env subunit protein boosts (2) to compare the frequency and titers of NAb induced among the three protein boost regimens and (3) to evaluate the evolution of NAb, i.e., the change in immunogenic responses whether NAb is detectable and/or shows variation in its expression, among volunteers during



the prime-boost regimen. Luciferase Reporter Pseudovirus Nab assay method has successfully transferred to AFRIMS Retrovirology laboratory to assay RVs 132 and 135 clinical samples from Dr. Montefiori's Laboratory for AIDS Vaccine Research and Development, Duke University Medical Center, Durham, NC, U.S.A.

Summary of Findings from Work Completed in 2010 Include:

1. RV135 regimens provided much lower neutralizing antibody titers than VAX003 but equal positive response rates to the sensitive virus; i.e., MN.3 - the vaccine strain
2. RV135 serum samples provided significantly stronger neutralizing antibody activity than the plasma samples on the very sensitive virus; i.e., MN.3
3. RV135 serum samples had less background neutralizing antibody activity comparing with the plasma samples

These results were very valuable in planning RV144 studies. In the coming year, more strains of pseudoviruses will be available for TZM-bl luciferase neutralizing antibody assay with RV135 high-dose. Samples from the low-dose group of RV135 will also be assayed in order to study dose effectiveness to neutralizing antibody activities.

RV217d is a multi-site cohort study designed to define risk behaviour, incidence of HIV in high risk population (MSMs, SWs, and TGs) and to identify individuals with acute HIV infection to support the full characterization of host response and viral dynamics in HIV pathogenesis. Volunteers are enrolled and followed for two years with blood collection every 6 months after baseline studies. Alternating 6 months the volunteers receives counseling and HIV prevention education. Twice a week the volunteers provide a capillary blood specimen for sensitive testing of very early HIV infection. Those identified as recently infected are studied intensively for ten visits and then followed for an additional 5 years.

The protocol began 23 July 2009. More than 823 potential volunteers have been interviewed and 422 volunteers have taken the ACASI questionnaire. 416 volunteers passed the screening visit and 333 were successfully enrolled. Fifty potential volunteers were already HIV infected at baseline for an overall prevalence of approximately 12%. Fourteen volunteers were captured during the very early stages of HIV infection. Retention to the large blood draw visits and to finger stick small blood volume (twice weekly collections) has been approximately 75%. In September 2010 the project successfully met the criteria for progressing to Part B (an incidence of at least 3.7/100 person years with 30% of the infections being in Fiebig stages I or II).

COLLABORATIONS

- Ministry of Public Health, Department of Disease Control (DDC), Nonthaburi
- Vaccine Trial Centre, Faculty of Tropical Medicine, Mahidol University, Bangkok
- Division of AIDS, NIAID, NIH
- Siriraj Hospital, Faculty of Medicine, Mahidol University, Bangkok
- AFRIMS- Division of Research (Thai component)
- Phramongkutklao Army Medical Center, Bangkok
- Data Management Unit, Faculty of Tropical Medicine, Mahidol University, Bangkok
- The Thai Red Cross AIDS Research Centre
- Hawaii AIDS Clinical Research Program, John A. Burns School of Medicine, University of Hawaii
- Laboratory for AIDS Vaccine Research and Development, Duke University Medical Center, Durham, NC, U.S.A.



- Collaboration of AIDS Vaccine Discovery
- Comprehensive Antibody - Vaccine Immune Monitoring Consortium (CA-VIMC)
- Sanofi-Pasteur
- VaxGen, Inc. (now Global Solutions for Infectious Diseases)

SUMMARY OF FUTURE PLANS AND STRATEGIES

The immediate focus of the coming year is additional laboratory studies of archive RV144 specimens to identify correlates of vaccine efficacy and better understand the immune response. Two advisory groups (Scientific and Product Development) made up of international experts and trial collaborators have been established to advise the sponsor on ways to improve the ALVAC prime gp120 boost vaccine regimen and future clinical development of the vaccines. Plans are underway to begin three follow-up studies to investigate immunologic response following late boosting and intense evaluation of immunologic profile following each vaccination. A phase IIB trial is planned for FY2012 to evaluate early efficacy of this vaccine regimen in community risk and high risk (MSM) population. In preparation of this phase IIB study, activities for development of cohort studies to characterized suitable population will begin in Q1 2011. These studies will involve new collaborators at Chiang Mai University; Phramongkutklao Hospital and Medical College of Medicine of the Royal Thai Army.

The Department of Retrovirology will continue to serve as one of the USMHRP's major testing platforms for phase I-II studies of newer vaccine candidates which will involve further testing of the subtype E (CRF01_AE) MVA vaccine candidate currently in phase I testing and newer DNA vaccine candidate. Further development of populations suitable for more advanced testing in phase IIB and III vaccine trials will be pursued through cohort studies of high-risk populations in Bangkok and Pattaya.

DEPARTMENT OF VETERINARY MEDICINE

DEPARTMENT MISSION

To protect military personnel and their families against tropical disease threats through pre-clinical product development of new prophylactic and therapeutic drugs and new or improved vaccines.

To fulfill this mission the Department of Veterinary Medicine (DVM) conducts biomedical research in animal models and zoonotic disease surveillance, provides veterinary expertise and research animals that are free of confounding diseases to intra- and extramural collaborators, and ensures that all animals receive humane, proper, and safe care and that the USAMC-AFRIMS' Animal Care and Use Program complies with appropriate laws, regulations and guidelines.