UVEITIS IN THAILAND

Emphasis on clinical patterns and novel developments in diagnostics using intraocular fluid analysis

Uveitis in Thailand
Met nadruk op de klinische manifestaties en nieuwe diagnostische mogelijkheden
gebaseerd op de analyse van intraoculair vocht
(met een samenvatting in het Nederlands)

Proefschrift

ter verkrijging van de graad van doctor
aan de Universiteit Utrecht op gezag van de
Rector magnificus, Prof.dr. J.C. Stoof,
ingevolge het besluit van het college voor promoties
in het openbaar te verdedigen op
donderdag 9 september 2010 des middags te 2.30 uur

door

Kessara Pathanapitoon

geboren op 23 november 1970 te Bangkok, Thailand

Promotor: Prof.dr. A. Rothova

This thesis was financially supported by the Dr F.P. Fischer-Stichting, Amersfoort, Dr. P. Binkhorst Foundation for Ophthalmologic research, Nijmegen; Stichting Blindenhulp, s-Gravenzande; Stichting Nederlands Oogheelkundig Onderzoek, Rotterdam, Rotterdamse Vereniging Blindenbelangen, Rotterdam; Landelijke Stichting voor Blinden en Slechtzienden, Utrecht; Katholieke Stichting voor Blinden en Slechtzienden, Grave, and Stichting Nelly Reef Fund, Ugchelen, the Netherlands.

The publication of this thesis was in part supported by Abbott Nederland, Allergan Nederland and Novartis Nederland.

"True success is not in the learning,
but in its application to the benefit of mankind"

Royal Highness Prince Mahidol of Songkhla, the Prince Father of Thailand

TABLE OF CONTENTS

CHAPTER 1

Introduction

I. GLOBAL BLINDNESS AND VISUAL IMPAIRMENT AND BLINDNESS IN THAILAND

Visual impairment is one of the most common disabilities; it was estimated that 45 million people worldwide are blind, and 200 million people have low vision.[1,2,3] From the population-based studies it emerged that the main causes of blindness worldwide consist of age-related cataract and glaucoma (in addition to refraction abnormalities).[1,3] In the West, the most common causes of blindness comprise retinal diseases, especially age-related macular degeneration (ARMD) and diabetic retinopathy, whereas in developing countries the leading causes of blindness are cataract and glaucoma.[1] One third of the world's blind people live in Asia.[1,3,4] Population-based studies specifying the causes of blindness in Asia point out that the major causes of blindness in Asia include cataract followed by glaucoma and corneal opacities.[1,5-7] The impact of intraocular inflammation on the blindness and low vision in Asia is not known.

The prevalence of blindness in South East Asia is around 0.8%; the specific rates vary from 0.3% in Thailand to 1.5% in Indonesia in 2000.[4,8] In Thailand, the primary ophthalmologic care has been aimed at cataract and glaucoma surgery and the prevention of nutritional diseases, and the prevalence of blindness has decreased dramatically during the past decades.[9] The reported prevalence of blindness in 2000 in Thailand was 0.31% and of low vision 0.8%, which is lower than in most countries in Asia, but these still represent 11% of all registered disabilities in Thailand.[4,8] In 1994, the most common causes of blindness in this region included cataract (75%) followed by corneal opacities, phthisis and glaucoma whereas the most common causes of low vision consisted of cataract and refractive errors.[5] The more recent report on the prevalence of ocular diseases and blindness in elderly Thai subjects from an urban population (Bangkok, 2001) revealed that the major causes consisted of cataract (67%), a combination of cataract and glaucoma (12%), glaucoma solely (4.5%) and ARMD (10%).[10]

In contrast, virtually nothing is known about the causes of blindness and low vision in patients consulting the tertiary centers in Asia, nor have the most frequent disorders and needs of patients consulting these centers been identified. The percentage of blindness and low vision due to infectious diseases, including human immunodeficiency virus (HIV)-related visual impairment, is also not known. Even with the huge HIV burden in South East Asia, cytomegalovirus (CMV) retinitis has not yet been recognized as an important cause of blindness.

To assess the underlying causes of blindness and low vision in patients referred to the tertiary centers in Asia, we investigated the anatomical location and specific causes of blindness and low vision in consecutive new 2951 patients consulting a large university centre in northern Thailand and provide the information on location, etiology and preventability of blindness-inducing disorders (Chapter 2).

II. UVEITIS IN THAILAND: EPIDEMIOLOGY, CAUSES AND ASSOCIATED SYSTEMIC DISORDERS

Uveitis is a major cause of severe visual impairment throughout the world. It comprises a large group of diverse diseases affecting not only the uvea but also the retina, optic nerve and vitreous. Changes in the etiology of uveitis over time have been brought about by effective treatment of specific infectious entities, such as tuberculosis and syphilis, and the emergence of new intraocular infections, such as cytomegalovirus (CMV) retinitis, related to HIV.[11] In addition, combinations of geographical, environmental, nutritional, socioeconomic, ethnic and genetic factors influence the causes of uveitis, and different patterns of uveitis are encountered around the world. The pattern of uveitis observed in the Far East differs considerably from that found in the West.[12,13] For example, sarcoidosis, a relatively common cause of non-infectious uveitis in the West (approximately 9.5%)[14,15], was only noted in 0.2% in a large survey from China[16] and 3.8% from India.[17] However, repeated reports from Japan point out the frequent prevalence of ocular sarcoidosis in uveitis surveys, with the prevalence of 9.5-18% among all with uveitis.[18-20] The other entities in posterior uveitis regularly encountered in the West such as presumed ocular histoplasmosis and birdshot retinochoroidopathy, have -so far- not been observed in the Far East. In contrast, Behcet's disease and Vogt-Koyanagi-Harada disease, which are rarely mentioned in reports from the West (approximately 0.8-2.5%)[14,15], are commonly observed in China (16.5%)[16], Korea and several reports from Japan (28%).[18,20]

Table 1. Causes of uveitis in Southeast Asia (in %)

	Japan Wakabayashi, 2003 [19]	India Singh, 2004 [17]	China Yang, 2005 [16]	Thailand Pathanapitoon, 2008 [54]
Infectious origin				
- Toxoplasmosis	1.7	1.7	0.1	8.7
- Tuberculosis	6.9	10.1	0.7	2.2
- HSV anterior uveitis	3.6	0.8	1.5	0
Association with systemic diseases				
- Sarcoidosis	9.5	3.8	0.2	0
- Spondyloarthropathy	7.9	8.8	3.3	7.2
- Behcet's disease	5.8	1.9	16.5	5.8
- Juvenile idiopathic arthritis	Not applicable	1.6	2	0
Established clinical entities				
- Vogt-Koyanagi-Harada	10.1	3.5	15.9	16
- Sympathetic ophthalmia	2	2.1	1.6	0.7
- Fuchs' heterochromic uveitis	0.5	2.5	5.7	3.6
Undetermined	Not applicable	51	44.8	19

However, large differences in uveitis etiology within Asian countries are also encountered, including differences in the prevalence of diverse infections such as ocular tuberculosis, toxoplasmosis and leptospirosis.[21] The prevalence of non-infectious uveitis entities within Southeast Asia also widely differs; e.g. the previously mentioned prevalence of ocular sarcoidosis is in Japan is much higher than in other countries (Table 1). In Thailand, data on the prevalence of different types of uveitis are lacking. To clarify the anatomical types and causes of uveitis in Thailand, we undertook a prospective study of 200 consecutive new patients with uveitis and determined their specific diagnoses on the basis of clinical manifestations and laboratory results (Chapter 3).

III. INFECTIOUS UVEITIS

1) Infectious uveitis in Thailand: Serological investigations

The early distinction of infectious from non-infectious causes of uveitis is crucial for their further management as regards their prognoses; the treatment options for infectious and non-infectious ocular inflammations are entirely different. Treatment of infectious uveitis with antibiotics may lead to improvement or even a cure of ocular disease and might also prevent further systemic involvement. The immunosuppressive treatment of infectious uveitis can be harmful, especially in areas where acquired immune deficiency syndrome (AIDS), tuberculosis, and other infectious diseases are common.[22] It is essential that the diagnosis of intraocular infection should be made before the immunosuppressive treatment is initiated.

Infectious etiology was documented in at least 20–30% of all uveitis cases in the West.[23] In that part of the world, herpes viruses and *Toxoplasma gondii* are the most common infectious agents involved in intraocular inflammations. In contrast, a higher percentage of infections in uveitis were observed in Asia and Africa, accounting for from 11.9%- 50 % of cases. Tuberculosis was the most common infectious cause of uveitis in India (10.1%: 125/1233) and Japan (6.9 %: 13/189). [17,19] Herpetic uveitis was the most common infectious cause in China (1.5%: 26/1752) and North Africa (11.9 %: 56/472). [16,24] Toxoplasmosis was the most common ocular infection in West Africa (43%: 40/93 of all with infections).[25]

In Thailand, cytomegalovirus (CMV) retinitis is a well-known cause of retinitis in human immunodeficiency virus (HIV)-infected patients. [26,27] However, the prevalence and specific causes of infectious uveitis in Thailand have not yet been systematically studied.

In chapter 4, in order to determine which infectious agents might play a role in the etiology of infectious uveitis, we determine the seroprevalence of various infections (known to cause uveitis in other parts of the world) in 101 consecutive Thai patients with non-HIV uveitis and in 100 HIV-infected patients with retinitis and compare the results with 100 non-uveitis controls.

2) Infectious uveitis in Thailand: Intraocular fluid analysis

Uveitis can be caused by numerous infectious agents. Clinical experience and appropriate diagnostic tests are useful for the accurate diagnosis and treatment of specific uveitis entities. In the West, the most common causes of infectious uveitis are *herpes simplex virus* (HSV) type1 and 2, *varicella zoster virus* (VZV) and *Toxoplasma gondii*. In immunocompromised patients, especially in those with HIV infection, cytomegalovirus (CMV) is the most common pathogen involved in ocular disease. The blood examinations in patients with infectious uveitis do not discriminate whether the antibodies or DNA found are a consequence of (past) systemic infection (and might be therefore coincidental), or indicate a cause of active intraocular disease. Therefore, for the definitive diagnosis of infectious uveitis, the analysis of intraocular fluid is required. Unfortunately, in Thailand, analysis of intraocular fluids is not readily available; the lack of these novel diagnostic tests in local laboratories and the costs involved prevented systematic research on these ocular infections in this country.

Immunosuppression due to AIDS is a serious public health problem that is occurring in epidemic proportions in Thailand. The Departments of Epidemiology and Statistics of the Division of Epidemiology, Ministry of Public Health reported that since 1984-2003, there were 312,429 AIDS patients registered in Thailand. The five most common opportunistic infections in Thailand are caused by *Mycobacterium tuberculosis* (66,575 pts; 26 %); *Pneumocystis carinii* (48,574 pts; 19%); Cryptococcus (38,464 pts; 15%) and Candida (11,974 pts; 5%); in addition, bacterial pneumonia was diagnosed in (8,425 pts; 3%).[28] Cytomegalovirus retinitis (CMVR) is the most common ocular opportunistic infection in patients with AIDS, occurring in up to one third of the patients; the prevalence and incidence of other opportunistic infections in ocular disease are not known. Previous study from the pre-HAART (highly active antiretroviral therapy) era in Thailand identified a prevalence of CMV retinitis in 33% of the population with AIDS (130/395 patients) using clinical assessment together with PCR on intraocular fluids.[29] Diagnosis of CMV retinitis in Thailand is usually based on clinical findings. However, overlapping funduscopic findings may occur and make it difficult to distinguish from necrotizing retinitis caused by VZV, HSV or by *T.gondii*. Furthermore, simultaneous infection of the retina by more than one pathogen makes it more complicated to have an accurate diagnosis of the disease. In the HAART era, an entirely different spectrum of ocular disease emerged in HIV-positive patients and includes immune recovery uveitis, toxic reaction to various drugs used for HIV infection (e.g. cidofovir, rifabutin). While the prevalence of co-infection with syphilis is rising, the prevalence of opportunistic infections has decreased.[30] It is important to discriminate between the infectious and non-infectious ocular infections in this population.

Polymerase chain reaction (PCR) is a technique, which uses DNA fragments from specific agents (called primers) and DNA polymerase which amplifies the complementary DNA -if present- in the sample. The value of PCR for analysis of intraocular fluids has repeatedly been reported and was especially successful in the detection of viruses.[31-33.]

Recently, intraocular fluid analysis by PCR has become available at the Division of Clinical Microbiology, Department of Medical Technology, in the Faculty of Associated Medical Sciences, Chiang Mai University, Thailand and has been developed in collaboration with the Department of Ophthalmology and Virology of the University Medical Center Utrecht, the Netherlands.

Detection of active intraocular antibody production might also help to diagnose infectious agent involved in uveitis. The mere presence of an intraocular antibody is not indicative of active antibody production within the eye, because the antibodies found might have been passively leaking from the peripheral circulation across the blood-eye barrier. The measurement of total IgG in the serum and within the eye together with the measurements of specific antibody in the serum and within the eye is necessary and comparing these results gives a so-called Goldmann-Witmer coefficient (GWC). If this coefficient is positive, it means that specific antibodies are produced in the eye and levels are higher than could be explained by a passive leakage from the blood. Since the combined analysis of intraocular samples by PCR and GWC has been reported to be superior to PCR analysis solely, at present, we introduce and evaluate the technique of GWC determination in our uveitis laboratory.[31,32]

In the chapter 5, we evaluate the results of PCR analysis for HSV, VZV, CMV and T. gondii of intraocular fluid in 100 non-HIV infected patients with uveitis of unknown origin and 47 HIV-infected patients with uveitis evaluated in our centre between May 2006 and October 2009 and evaluate the usefulness of PCR for the diagnosis of uveitis of unknown cause in Thailand.

3) Infectious uveitis: Intraocular and plasma HIV1- RNA loads and exploration of HIV-induced uveitis

Ocular involvement in human immunodeficiency virus (HIV)-infected patients was in the pre-HAART era caused by progressive immune dysfunction and the most ocular abnormalities resulted from opportunistic infections and malignancies common to AIDS. In the HAART era, the prevalence of opportunistic (ocular) infections has decreased while the prevalence of immune recovery uveitis has increased.[30] Although HAART has led to a profound decrease in morbidity and mortality of infected people by suppressing HIV replication, the virus continues to evolve slowly during therapy even when patients achieve non-detectable levels of HIV in plasma. HIV-1 persists latently in infected memory CD4+ T cells. Various reservoirs and sanctuary sites harboring HIV such as the central nervous system were established in vivo during antiretroviral therapy.[34-38] Such compartments can harbor residual viruses, allow a persistence of HIV even in patients with otherwise effective antiretroviral treatment and suppression of HIV load in plasma and might theoretically also induce drug resistances.[34-41] In addition, the HIV itself might in such cases induce specific disorders, such as previously reported HIV- encephalitis and HIV-associated dementia in CNS.[40, 41, 42]

Ocular tissues might potentially represent a virus reservoir. Several cases of suspected HIV-associated uveitis with no evidence of other intraocular infectious agents causing uveitis than HIV were reported.[43] In the eye, the possibility of compartmentalization and the local replication of HIV and its possible clinical consequences have not yet been systematically assessed.

In chapter 6, we determine intraocular and plasma HIV-1 RNA loads in 40 HIV infected patients with and without intraocular opportunistic infections and study the HIV dynamics across the blood-retinal barrier. In addition, we describe clinical manifestations of patients with detectable intraocular HIV load, with the emphasis on patients with intraocular HIV loads exceeding the HIV loads of plasma.

IV. NON-INFECTIOUS UVEITIS ENTITIES

1) HLA-B27associated acute anterior uveitis (AAU) in Thailand

Acute anterior uveitis (AAU) is the most frequent type of uveitis encountered in West Europe and the US, accounting for about 50% of all cases of uveitis.[44,45] In most uveitis series, the prevalence of HLA-B27 in patients with AAU was about 50%, ranging from 19% to 82% across the different racial groups.[44,46-49] In Asia, similar findings to those in the Western population were reported for the Indian subcontinent, but by contrast, in Japan the frequency of AAU was reported to be low.[46,47] In Thailand, the eventual presence and clinical features of HLA-B27-associated AAU are not known.

To study whether HLA-B27-associated AAU plays a role (if any) in the Thai population, we studied first the frequency of HLA-B27 in the healthy Thai population without AAU and second, in patients with acute anterior uveitis from the same area. In addition, we describe the clinical features of HLA-B27-positive AAU in this particular population (chapter 7).

2) Ocular sarcoidosis in Thailand

Sarcoidosis affects people of all racial and ethnic groups and can occur at all ages, though; there is a great variety of incidence and prevalence throughout the world.[50,51] In the United States, the majority of patients are black with a prevalence of 40 per 100,000, compared to 5 per 100,000 among White Europeans.[50,51] In contrast to Japan, sarcoidosis is less common in India, Asia, New Zealand and mainland China.[51] So far, it is not known whether the rarity of sarcoidosis in Southeast Asia is genuine or whether sarcoidosis in this geographic area remains underdiagnosed. Prevalence of sarcoidosis among patients with uveitis in the West is approximately 9.5%[14,15], in Japan 9.5-18%[18-20], and in India [21] and Taiwan 4%[52], but virtually no patients with ocular sarcoidosis have been identified in mainland China.[16,52] In Thailand, systemic sarcoidosis has rarely been diagnosed and so far, no cases with ocular sarcoidosis have been reported.

In chapter 8, we search for the signs of pulmonary sarcoidosis in 209 new consecutive patients with uveitis and describe ocular features of Thai patients with signs of associated pulmonary sarcoidosis.

3) Chronic central serous chorioretinopathy associated with serous retinal detachment in a series of Asian patients

Vogt-Koyanagi-Harada (VKH) syndrome is a systemic disease, probably of autoimmune origin and is characterized by various ocular, cutaneous and neurological abnormalities. It represents a most frequent form of non-infectious uveitis in Thailand.[53] Diagnosis of VKH is made on clinical grounds including ocular features which are chronic bilateral granulomatous panuveitis in association with serous retinal detachment, development of sunset glow phenomenon and Dalen-Fuchs nodules and systemic manifestations which are vitiligo, poliosis, alopecia, signs of meningeal irritation, and auditory disturbances. Revised diagnostic criteria for Vogt-Koyanagi-Harada disease were published in 2001 (Table 2).[54] Since there exists no definitive proof of VKH, the diagnosis is made by the above indicated diagnostic criteria and by exclusion of other causes of uveitis. In practical situations however, the strict adherence to these criteria is not feasible and the clinical suspicion of VKH usually rises in patients with uveitis who develop serous retinal detachments.

In chapter 9, we report on a masquerade syndrome mimicking the VKH disease in 7 patients chronic central serous chorioretinopathy (CSR) associated with bullous retinal detachment and report on their clinical presentations, response to treatment and signs which differentiate patients with severe CSR from those with VKH.

The main objective of this thesis is to elucidate the causes of uveitis in Thailand, to report on their clinical manifestations and to increase our knowledge in the field of (at times blinding but frequently treatable) intraocular inflammations.

Table 2. Diagnostic Criteria for Vogt-Koyanagi-Harada Disease

Complete Vogt-Koyanagi-Harada disease (criteria 1 to 5 must be present)

1. No history of penetrating ocular trauma or surgery preceding the initial onset of uveitis.
2. No clinical or laboratory evidence suggestive of other ocular disease entities.
3. Bilateral ocular involvement (a or b must be met, depending on the stage of disease when the patient is examined).
   a. Early manifestations of disease.
      (1) There must be evidence of a diffuse choroiditis (with or without anterior uveitis, vitreous inflammatory reaction, or optic disk hyperemia), which may manifest as one of the following:
         (a) Focal areas of subretinal fluid, or
         (b) Bullous serous retinal detachments.
      (2) With equivocal fundus findings; both of the following must be present as well:
         (a) Focal areas of delay in choroidal perfusion, multifocal areas of pinpoint leakage, large placoid areas of hyperfluorescence, pooling within subretinal fluid, and optic nerve staining (listed in order of sequential appearance) by fluorescein angiography,
         and
         (b) Diffuse choroidal thickening, without evidence of posterior scleritis by ultrasonography.
   b. Late manifestations of disease.
      (1) History suggestive of prior presence of findings from 3a, and either both (2) and (3) below, or multiple signs from (3):
      (2) Ocular depigmentation (either of the following manifestations is sufficient):
         (a) Sunset glow fundus, or
         (b) Sugiura sign.
      (3) Other ocular signs:
         (a) Nummular chorioretinal depigmented scars, or
         (b) Retinal pigment epithelium clumping and/or migration, or
         (c) Recurrent or chronic anterior uveitis.
4. Neurological/auditory findings (may have resolved by time of examination).
   a. Meningismus (malaise, fever, headache, nausea, abdominal pain, stiffness of the neck and back, or a combination of these factors; headache alone is not sufficient to meet definition of meningismus, however), or
   b. Tinnitus, or
   c. Cerebrospinal fluid pleocytosis.
5. Integumentary findings (not preceding onset of central nervous system or ocular disease).
   a. Alopecia, or
   b. Poliosis, or
   c. Vitiligo.

Incomplete Vogt-Koyanagi-Harada disease (criteria 1 to 3 and either 4 or 5 must be present)

1. No history of penetrating ocular trauma or surgery preceding the initial onset of uveitis, and
2. No clinical or laboratory evidence suggestive of other ocular disease entities, and
3. Bilateral ocular involvement.
4. Neurologic/auditory findings; as defined for complete Vogt-Koyanagi-Harada disease above, or
5. Integumentary findings; as defined for complete Vogt-Koyanagi-Harada disease above.

Probable Vogt-Koyanagi-Harada disease (isolated ocular disease; criteria 1 to 3 must be present)

1. No history of penetrating ocular trauma or surgery preceding the initial onset of uveitis.
2. No clinical or laboratory evidence suggestive of other ocular disease entities.
3. Bilateral ocular involvement as defined for complete Vogt-Koyanagi-Harada disease above.

REFERENCES

1. Resnikoff S, Pascolini D, Etya'ale D, Kocur I, et al. Global data on visual impairment in the year 2002. Bull World Health Organ 2004; 82: 844-51.
2. Resnikoff S, Pararajasegaram R. Blindness prevention programmes: past, present, and future. Bull World Health Organ 2001; 79: 222-6.
3. Pascolini D, Mariotti SP, Pokharel GP, Pararajasegaram R, et al. 2002 global update of available data on visual impairment: a compilation of population-based prevalence studies. Ophthalmic Epidemiol 2004; 11:67-115.
4. World Health Organization: Table 61 Status of eye health and eye care in the SEA Region, by country, 2000. Health Situation in the South-East Asia Region, 1998-2000. Available at http://www.searo.who.int/LinkFiles/Health_Situation_book.pdf
5. Wongwetsawat S. Epidemiology of blindness and low vision in thailand 1994. Thai Journal of Public Health Ophthalmology 2003, 17: 39-45.
6. Saw SM, Husain R, Gazzard GM, Koh D, et al. Causes of low vision and blindness in rural Indonesia. Br J Ophthalmol 2003; 87: 1075-8.
7. Saw SM, Foster PJ, Gazzard G, Seah S. Causes of blindness, low vision, and questionnaire assessed poor visual function in Singaporean Chinese adults: The Tanjong Pagar Survey. Ophthalmology 2004; 111: 1161-8.
8. Country Profile on Disability, Kingdom of Thailand; Japan International Cooperating Agency; March 2002. Available at http://www.jica.go.jp/english/global/dis/profile.html
9. Jenchitr WT, Tansirikongkol V. Ophthalmology in Thailand 1997. J Med Assoc Thai 2000; 83: 107-16.
10. Singalavanija A, Metheetrairut A, Ruangvaravate N, Tuchinda R, et al. Ocular diseases and blindness in elderly Thais. J Med Assoc Thai 2001; 84:1383-8.
11. Nussenblatt RB, Whitcup SM. Uveitis: fundamentals and clinical practice. 3rd edn. Mosby: Philadelphia, 2004.
12. Chang JHM, Wakefield D. Uveitis: a global perspective. Ocul Immunol Inflamm 2002; 10: 263–79.
13. Wakefield D, Chang JHM. Epidemiology of uveitis. Int Ophthalmol Clin 2005; 45: 1–13.
14. Rothova A, Buitenhuis HJ, Meenken C, Brinkman CJ, et al. Uveitis and systemic disease. Br J Ophthalmol 1992; 76: 137-41.
15. Rodriguez A, Claonge M, Pedroza- Seres M, Akova YA, et al. Referral patterns of uveitis in a tertiary eye care center. Arch Ophthalmol 1996; 114: 593–99.
16. Yang P, Zhang Z, Zhou H, Li B, Huang X, et al. Clinical patterns and characteristics of uveitis in a tertiary center for uveitis in China. Curr Eye Res 2005; 30: 943-8.
17. Singh R, Gupta V, Gupta A. Pattern of uveitis in a referral eye clinic in north India. Indian J Ophthalmol 2004; 52: 121-5.
18. Goto H, Mochizuki M, Yamaki K, Kotake S, et al. Epidemiological survey of intraocular inflammation in Japan. Jpn J Ophthalmol 2007; 51: 41-4.
19. Wakabayashi T, Morimura Y, Miyamoto Y, Okada AA. Changing patterns of intraocular inflammatory disease in Japan. Ocul Immunol Inflamm 2003; 11: 277-86.

20. Kotake S, Furudate N, Sasamoto Y, Yoshikawa K, et al. Characteristics of endogenous uveitis in Hokkaido, Japan. Graefe's Arch Clin Exp Ophthalmol 1997; 235: 5–9.
21. Rathinam SR, Namperumalsamy P. Global variation and pattern changes in epidemiology of uveitis. Indian J Ophthalmol 2007; 55: 173–83.
22. Dunn JP. Review of immunosuppressive drug therapy in uveitis. Curr Opin Ophthalmol 2004; 15: 293–8.
23. Gritz DC, Wong IG. Incidence and prevalence of uveitis in Northern California: the Northern California epidemiology of uveitis study. Ophthalmology 2004; 111: 491–500.
24. Khairallah M, Yahia SB, Ladjimi A, Messaoud R, et al . Pattern of uveitis in a referral centre in Tunisia, North Africa. Eye 2007; 21: 33-9.
25. Ronday MJ, Stilma JS, Barbe RF, McElroy WJ, et al. Aetiology of uveitis in Sierra Leone, West Africa. Br J Ophthalmol 1996; 80: 956-61.
26. Ausayakhun S, Watananikorn S, Ittipunkul N, et al. Epidemiology of the ocular complications of HIV infection in Chiang Mai. J Med Assoc Thai 2003; 86: 399–406.
27. Pathanapitoon K, Ausayakhun S, Kunavisarut P, et al. Blindness and low vision in a tertiary ophthalmologic center in Thailand: the importance of cytomegalovirus retinitis. Retina 2007; 27: 635–40.
28. Annual epidemiological surveillance report 2006: AIDs. Bureau of Epidemiology, Department of Disease Control, Ministry of Public Health, Thailand. Available at http://203.157.15.4/Annual/Annual49/Part1/25_AIDS.DOC
29. Pathanapitoon K, Ausayakhun S, Kunavisarut P, Pungrasame A, Sirirungsi W. Detection of cytomegalovirus in vitreous, aqueous and conjunctiva by polymerase chain reaction. J Med Assoc Thai 2005; 88: 228-32.
30. Jabs DA, Van Natta ML, Holbrook JT, Kempen JH, et al. Studies of the ocular complications of AIDS research group. Longitudinal study of the ocular complications of AIDS: 1. Ocular diagnoses at enrollment. Ophthalmology 2007; 114: 780-6.
31. De Groot-Mijnes JD, Rothova A, Van Loon AM, Schuller M, et al. Polymerase chain reaction and Goldmann-Witmer coefficient analysis are complimentary for the diagnosis of infectious uveitis. Am J Ophthalmol 2006; 141: 313-8.
32. Westeneng AC, Rothova A, de Boer JH, de Groot-Mijnes JD. Infectious uveitis in immunocompromised patients and the diagnostic value of polymerase chain reaction and Goldmann-Witmer coefficient in aqueous analysis. Am J Ophthalmol 2007; 144: 781-5.
33. Rothova A, de Boer JH, Ten Dam-van Loon NH, Postma G, et al. Usefulness of aqueous humor analysis for the diagnosis of posterior uveitis. Ophthalmology 2008; 115: 306-11.
34. Pierson T, McArthur J, Siliciano RF. Reservoirs for HIV-1: mechanisms for viral persistence in the presence of antiviral immune responses and antiretroviral therapy. Annu Rev Immunol. 2000; 18: 665-708.
35. Shepard RN, Schock J, Robertson K, Shugars DC, et al. Quantitation of human immunodeficiency virus type 1 RNA in different biological compartments. J Clin Microbiol. 2000; 38: 1414-8.
36. Persidsky Y, Poluektova L. Immune privilege and HIV-1 persistence in the CNS. Immunol Rev. 2006; 213: 180-94.

37. De Luca A, Ciancio BC, Larussa D, Murri R, et al. Correlates of independent HIV-1 replication in the CNS and of its control by antiretrovirals. Neurology. 2002; 59: 342-7.
38. García F, Niebla G, Romeu J, Vidal C, et al. Cerebrospinal fluid HIV-1 RNA levels in asymptomatic patients with early stage chronic HIV-1 infection: support for the hypothesis of local virus replication. AIDS. 1999; 13: 1491-6.
39. Strain MC, Letendre S, Pillai SK, Russell T, et al. Genetic composition of human immunodeficiency virus type 1 in cerebrospinal fluid and blood without treatment and during failing antiretroviral therapy. J Virol. 2005; 79: 1772-88.
40. Gisslén M, Fuchs D, Svennerholm B, Hagberg L. Cerebrospinal fluid viral load, intrathecal immunoactivation, and cerebrospinal fluid monocytic cell count in HIV-1 infection. J Acquir Immune Defic Syndr. 1999; 21: 271-6.
41. Stingele K, Haas J, Zimmermann T, Stingele R, et al. Independent HIV replication in paired CSF and blood viral isolates during antiretroviral therapy. Neurology. 2001; 56: 355-61.
42. Cinque P, Vago L, Ceresa D, Mainini F, et al. Cerebrospinal fluid HIV-1 RNA levels: correlation with HIV encephalitis. AIDS. 1998; 12: 389-94.
43. Rothova A, Schneider M, de Groot-Mijnes JD. Human immunodeficiency virus-induced uveitis: intraocular and plasma human immunodeficiency virus-1 RNA loads. Ophthalmology. 2008; 115: 2062-4.
44. Chang JH, McCluskey PJ, Wakefield D. Acute anterior uveitis and HLA-B27. Surv Ophthalmol 2005; 50: 364–88.
45. McCannel CA, Holland GN, Helm CJ, Cornell PJ, et al. Causes of uveitis in the general practice of ophthalmology. UCLA Community-Based uveitis study group. Am J Ophthalmol 1996; 121: 35–46.
46. Sasaki T, Kusaba Y, Yamamoto T. HLA-B27 and acute anterior uveitis in the Japanese population. Jpn J Ophthalmol 1979; 23: 374–7.
47. Mehta S. Pattern of uveitis in a referral eye clinic in North India. Indian J Ophthalmol 2004; 52: 121–5.
48. Huhtinen M, Karma A. HLA-B27 typing in the categorization of uveitis in a HLA-B27 rich population. Br J Ophthalmol 2000; 84: 413–16.
49. Pathanapitoon K, Preechawai P, Visaetsilapanonta S, Kunavisarut P, et al. Prevalence of HLA-B27 in acute anterior uveitis patients. Thai J Ophthalmol 2002; 16: 91–7.
50. Michael C. Sarcoidosis. N Engl J Med 2007; 357: 2153-65.
51. Sharma OP. Sarcoidosis around the world. Clin Chest Med 2008; 29: 357-63.
52. Chung YM, Lin YC, Liu YT, Chang SC, et al. Uveitis with biopsy-proven sarcoidosis in Chinese-A study of 60 patients in uveitis clinic over a period of 20 years. J Chin Med Assoc 2007; 70: 492-96.
53. Pathanapitoon K, Kunavisarut P, Ausayakhun S, Sirirungsi W, Rothova A. Uveitis in a tertiary ophthalmology centre in Thailand. Br J Ophthalmol 2008; 92: 474-8.
54. Read RW, Holland GN, Rao NA, Tabbara KF et al. Revised diagnostic criteria for Vogt-Koyanagi-Harada disease: report of an international committee on nomenclature. Am J Ophthalmol 2001; 131: 647–52.

CHAPTER 2

Blindness and low vision in a tertiary ophthalmologic centre in Thailand: the importance of cytomegalovirus retinitis

Pathanapitoon K, Ausayakhun S, Kunavisarut P, Wattananikorn S, Ausayakhun S, Leeungurastien T, Yodprom R, Narongjunchai D, Rothova A.

Retina 2007; 27: 635–40.

Reprinted with permission

ABSTRACT

Purpose: To determine the causes of blindness and low vision in patients consulting a tertiary ophthalmologic center in northern Thailand.

Methods: The study population included 2,951 new consecutive patients from the department of ophthalmology at university hospital in Chiang-Mai, Thailand. Main outcome measures were blindness and low vision, which were defined according to World Health Organization criteria.

Results: Of 2,951 patients, 369 (12.5%) had blindness and/or low vision (bilateral blindness in 73, unilateral blindness in 129, bilateral low vision in 77, and unilateral low vision in 90). Of the etiological causes of visual loss, age-related ocular disease was the most frequent (128 patients [35%]) followed by infections (66 patients [18%]) and trauma (43 patients [12%]). Although infections and trauma were the predominant causes of blindness, age-related disorders were frequently found in patients with low vision. Of anatomical sites, the lens (134 patients [36%]) was the main location of visual loss, closely followed by disorders of the retina and/or uvea (126 patients [34%]). Blindness and low vision were considered avoidable in 70% of cases. Of 73 patients with bilateral blindness, 14 had active cytomegalovirus retinitis, accounting for 19% of all patients with bilateral blindness.

Conclusion: The most common causes of blindness and low vision in a tertiary center in northern Thailand were age-related ocular disorders and infections, which were predominantly cases of cytomegalovirus retinitis in human immunodeficiency virus–infected patients.

Key words: blindness and visual impairment, cytomegalovirus retinitis, Thailand.

INTRODUCTION

Visual impairment is one of the most common disabilities: an estimated 45 million people worldwide are blind, and 200 million people have low vision.[1-3] Population-based studies have shown that the main causes of blindness worldwide (in addition to refraction abnormalities) are age-related cataract and glaucoma.[1,3] The prevalence of blindness and low vision due to infectious diseases, including human immunodeficiency virus (HIV)–related visual impairment, is not known. Cytomegalovirus (CMV) retinitis has not yet been recognized as an important cause of blindness, despite the huge burden of HIV infection in various areas of the world such as Southeast Asia and Africa. In Thailand during the last decades, ophthalmologic care has been aimed at the major causes of blindness (cataract and glaucoma surgery along with prevention of nutritional diseases), and the prevalence of bilateral blindness has decreased dramatically from 1.1% to 0.31%.[4,5] Nothing is known about the causes of blindness and low vision in patients consulting the tertiary centers in Southeast Asia, nor have the most frequent disorders and needs of patients consulting these centers been identified. In this study, we investigated the causes of blindness and low vision in a large university teaching hospital serving the whole of northern Thailand and paid specific attention to intraocular infections, specifically CMV retinitis.

PATIENTS AND METHODS

Of 2,951 new consecutive patients consulting the department of ophthalmology at university hospital in Chiang Mai, Thailand, from February 1 to June 31, 2005, 369 (12.5%) with bilateral or unilateral blindness and/or low vision were prospectively selected for the study. The World Health Organization's definitions of blindness (corresponding to corrected visual acuity of < 0.05 or 20/400) and low vision (corresponding to corrected visual acuity of ≤ 0.30 or 20/60) were used in this study.[6,7] Patients were assessed according to anatomical location and etiology of the main cause of blindness. In addition, preventability and curability of blindness were also registered. These data were analyzed for both patients (n = 369) and affected eyes (n = 519). When a combination of diverse causes leading to blindness and/or low vision was noted, the most direct cause of visual loss was selected. If this selection was not possible, the cases were classified separately as combinations (n = 12). The refraction abnormalities were not represented among the causes of visual loss, except where there was still subnormal vision with optimal correction (e.g., amblyopia). Anatomical location of the cause of blindness in glaucoma was classified as optic disk blindness. The etiology of primary open-angle glaucoma was classified as unknown, as was that of rhegmatogenous retinal detachment (with the exception of myopic and traumatic cases). Reversible blindness and low vision

were considered in patients with cataract (possibility of surgery). The preventable category included trauma and glaucoma (except congenital glaucoma) as well as CMV retinitis in HIV-infected patients (possibility of highly active antiretroviral therapy [HAART]). In intraocular inflammations, treatable loss of vision was considered in identified infectious causes, whereas the treatability of visual loss in uveitis of unknown cause was considered unknown. Blindness due to retinal vascular occlusions, age-related macular degeneration, and optic atrophy was considered not preventable and not reversible. Data for patients were computerized and statistically analyzed by means of the Chi-square test and Fisher exact test. $P < 0.05$ was considered statistically significant.

Table 1. Blindness and low vision in 2,951 new consecutive patients consulting the department of ophthalmology at university hospital in Chiang Mai, Thailand

Finding	Blindness*	Low vision†	Total
Bilateral loss of vision	73	77	150
Unilateral loss of vision	129	90	219
Total	202	167	369

* One or both eyes matching the World Health Organization's definition of blindness (corresponding to corrected visual acuity of < 0.05 or 20/400).
† One or both eyes matching the World Health Organization's definition of low vision (corresponding to corrected visual acuity of ≤ 0.30 or 20/60 and ≥ 0.05 or 20/400).

RESULTS

Our study included 369 patients with blindness or low vision, of whom 150 (41%) had bilateral loss and 219 (59%) had unilateral loss (519 affected eyes) (Table 1). The male-to-female ratio was 1:1, and the mean age of the patients was 52 years (median, 54 years; range, 4–87 years).

The main causes of visual impairment and blindness are listed in Table 2. The most common etiological causes were age-related ocular diseases (128 patients [35%], of whom 123 had cataract and the remainder had other age-related ocular disorders), followed by infections (66 patients [18%]) and trauma (43 patients [12%]). After age-related causes, infections were the second major cause of both blindness and low vision (21% and 14%, respectively). Although trauma was predominantly encountered in blindness ($P = 0.006$), age-related disorders were more frequently noted in low vision ($P < 0.001$).

The classification of patients according to severity and laterality of visual loss is listed in Table 3. For all 369 patients, the main anatomical location of visual loss was the lens with cataract (134 [36%]/369, including age-related cataract and cataract from other causes such as traumatic, juvenile, or others), which was followed by disorders of the retina

and/or uvea (126 [35%]/369). Active CMV retinitis was the cause of visual loss in 11% (42) of 369 patients, while age-related macular degeneration was the cause only in 0.8% (3) of 369 patients. Of 369 patients, 45 (12% [42 with active CMV retinitis, 2 with retinal detachment due to CMV retinitis, and 1 with herpetic retinopathy]) had loss of visual acuity due to HIV infection. Visual loss was caused by glaucoma in 11 (3%), non-HIV uveitis in 19 (5%),

Table 2. Causes of bilateral and unilateral blindness and bilateral and unilateral low vision

Cause	No. (%) of All Patients	No. (%) With Bilateral Blindness	No. (%) With Unilateral Blindness	P, Unilateral vs. Bilateral Blindness	No. (%) With Bilateral Low Vision	No. (%) With Unilateral Low Vision	P, Unilateral vs. Bilateral Low Vision
Age-related	128 (35)	32 (44)	23 (18)	< 0.001	36 (47)	37 (41)	NS
Infections*	66 (18)	17 (23)	25 (19)	NS	10 (13)	14 (16)	NS
Trauma**	43 (11.7)	1 (1)	31 (24)	< 0.001	0	11 (12)	< 0.002
Hereditary and developmental ocular diseases†	30 (8.1)	5 (7)	7 (5)	NS	11(14)	7 (8)	NS
Diabetes mellitus	12 (3.3)	3 (4)	3 (2)	NS	5 (6.5)	1 (1)	NS
Ocular vascular occlusions	5 (1.4)	1 (1)	2 (2)	NS	0	2 (2)	NS
Retinopathy of prematurity	2 (0.5)	1 (1)	0	NS	1(1)	0	NS
Non-malignant tumors	1 (0.3)	1 (1)	0	NS	0	0	NS
Malignancy	0	0	0	NS	0	0	NS
Nutritional	0	0	0	NS	0	0	NS
Other	6 (1.6)	0	3 (2)	NS	2 (3)	1 (1)	NS
Unknown†	64 (17.3)	7 (10)	35 (27)	< 0.004	5 (6.5)	17 (19)	< 0.018
Combinations of causes	12 (3.3)	5 (7)	0	NA	7 (9)	0	NA
TOTAL	369 (100)	73 (100)	129 (100)		77 (100)	90 (100)	

* Includes 45 patients with human immunodeficiency virus–related visual loss (42 with active cytomegalovirus retinitis, 2 with retinal detachment due to cytomegalovirus retinitis, and 1 with herpetic retinopathy).
† Includes also surgical trauma (3 patients with bullous keratopathy after cataract surgery).
‡ Hereditary and developmental causes also include amblyopia and myopia. For unknown causes and further details, see Methods.
NS, not significant; NA, not applicable.

and retinal detachment in 18 (5%) of 369 patients. Bilateral blindness and/or low vision was more frequently caused by cataract than was unilateral blindness and/or low vision (69 [46%]/150 vs. 65 [30%]/219 patients, respectively; $P < 0.001$). In contrast, the most frequent cause of unilateral visual loss was retinal and/or uveal disorders (81 [37%]/219 patients). Disorders affecting the cornea and whole globe were more frequently noted in unilateral visual loss than in bilateral visual loss (25/ 219 vs. 6/150 patients, respectively [$P < 0.012$]; and 19/219 vs. 0/150 patients, respectively [$P < 0.001$]). The prevalence of retinal diseases,

and specifically of CMV retinitis, did not differ between unilaterally and bilaterally affected patients (20 [9.1%]/219 vs. 22 [14.7%]/150, respectively; $P = 0.1$). Age-related ocular disorders were more frequent in bilateral blindness and low vision than in unilateral blindness and low vision (68 [45.3%]/150 vs. 60 [27.4%]/ 219 patients, respectively; $P < 0.001$), while trauma-induced blindness and low vision were more frequent in unilateral visual loss than in bilateral visual loss (1 [0.7%]/150 vs. 42 [19.2%]/219 patients, respectively; $P < 0.001$). Of 150 patients with bilateral blindness and/or low vision, 23 (22 with CMV retinitis and 1 with progressive herpetic outer retinal necrosis) had HIV-related visual loss, thus accounting for 15% of all patients with bilateral blindness or low vision.

Patients with blindness and low vision were further subdivided according to four separate categories (bilateral blindness, unilateral blindness, bilateral low vision, and unilateral low vision; Table 4). Retinal and/or uveal disorders were a major cause of blindness (76 [38%]/202 patients), whereas cataract was the main cause of low vision (76 [46%]/167 patients). The association of unilateral blindness with trauma and a marginally higher prevalence of bilateral blindness with CMV retinitis were also observed. When 150 patients with bilateral visual loss were subdivided into blindness (n = 73) and low vision (n = 77) groups and compared, no differences emerged in terms of location or etiological cause of visual loss (all locations and causes, $P = 0.1$).

When the data were evaluated for affected eyes (n = 519), a higher percentage of blindness and low vision due to bilateral diseases was noted (e.g., cataract). No other differences were observed. Blindness and low vision were considered preventable (n = 126) and/or reversible (n = 134) in 260 (70%) of 369 cases. Of 73 patients with bilateral blindness, 30 (42%) had cataract and 26 (36%) had retinal/uveal disorders, of whom 14 (19%) had active CMV retinitis.

DISCUSSION

The leading causes of blindness and low vision at a large tertiary center in northern Thailand were age related diseases and ocular infections, which were predominantly cases of CMV retinitis in HIV-infected patients. Bilateral active CMV retinitis was present in 19% of all patients with bilateral blindness. Loss of vision was considered preventable and/or reversible in most patients.

The very high percentage of CMV retinitis among blind patients at a tertiary center is striking. Immunosuppression due to acquired immunodeficiency syndrome is a serious public health problem in Southeast Asia. Recent data indicate that the prevalence of HIV infection among the whole population of Thailand is 1.5%.[8, 9] Before the era of HAART, ~ 30% of patients with acquired immunodeficiency syndrome in industrialized countries developed CMV retinitis.[10,11] The combination of HAART and effective anti-CMV drugs has improved the visual

Table 3. Anatomical location of ocular disorders causing loss of visual acuity classified according to laterality and severity of visual loss

Anatomical location	No. (%) of All Patients	No. (%) With Bilateral Blindness and Low Vision	No. (%) With Unilateral Blindness and Low Vision	P, Unilateral vs. Bilateral Blindness and Low Vision	No. (%) With Blindness (Unilateral and Bilateral)	No. (%) With Low Vision (Unilateral and Bilateral)	P, Blindness vs. Low Vision
Cornea	31 (8.4)	6 (4)	25 (11.4)		19 (9.4)	12 (7.2)	
Bullous keratopathy	3 (0.8)	1 (0.7)	2 (0.9)		3 (1.5)	0	
Corneal scar	13 (3.5)	2 (1.3)	11 (5)	0.012	9 (4.5)	4 (2.4)	0.44
Corneal ulcer	9 (2.4)	2 (1.3)	7 (3.2)		7 (3.5)	2 (1.2)	
Miscellaneous	6 (1.6)	1 (0.7)	5 (2.3)		0	6 (3.6)	
Lens	134 (36.3)	69 (46)	65 (29.7)	< 0.001	58 (28.7)	76 (45.5)	0.001
Retina/ uvea	126 (34.6)	45 (30)	81 (37)		76 (37.6)	50 (29.9)	
CMV retinitis	42 (11.4)	22 (14.7)	20 (9.1)		24 (11.9)	18 (10.8)	
Uveitis (non-CMV)	19 (5.1)	4 (2.7)	15 (6.9)		14 (6.9)	5 (3)	
Retinal detachment*	18 (4.9)	3 (2)	15 (6.9)	0.17	14 (6.9)	4 (2.4)	0.12
Vascular occlusions	4 (1.1)	0	4 (1.8)		2 (1)	2 (1.2)	
Diabetic retinopathy	9 (2.4)	6 (4)	3 (1.4)		5 (2.5)	4 (2.4)	
ARMD	3 (0.8)	2 (1.3)	1 (0.5)		3 (1.5)	0	
Tapetoretinal degenerations	4 (1.1)	4 (2.7)	0		2 (1)	2 (1.2)	
Miscellaneous	27 (7.9)	4 (2.7)	23 (10.5)		12 (5.9)	15 (9)	
Optic disc	28 (7.6)	9 (6)	19 (8.7)		20 (10)	8 (4.8)	
Glaucoma	11 (3)	4 (2.7)	7 (3.2)	0.34	8 (4.0)	3 (1.8)	0.07
Atrophy	13 (3.5)	4 (2.7)	9 (4.1)		10 (5)	3 (1.8)	
Neuritis	4 (1.1)	1 (0.7)	3 (1.4)		2 (1)	2 (1.2)	
Whole globe	19 (5.1)		19 (8.7)		19 (9.4)		
Endophthalmitis	5 (1.4)		5 (2.3)		5 (2.5)		
Phthisis	5 (1.4)	0	5 (2.3)	< 0.001	5 (2.5)	0	< 0.001
Rupture	5 (1.4)		5 (2.3)		5 (2.5)		
Contusion	2 (0.5)		2 (0.9)		2 (1)		
Microphthalmos	2 (0.5)		2 (0.9)		2 (1)		
Amblyopia	19 (5.1)	9 (6)	10 (4.6)		5 (2.5)	14 (8.4)	
Myopic amblyopia†	10 (2.7)	5 (3.3)	5 (2.3)	0.54	4 (2)	6 (3.6)	0.01
Non-myopic amblyopia	9 (2.4)	4 (2.7)	5 (2.3)		1 (0.5)	8 (4.8)	
Combinations of diverse disorders	12 (3.2)	12 (8)	NA	NA	5 (2.5)	7 (4.2)	0.36
Total	369 (100)	150 (100)	219 (100)		202 (100)	167 (100)	

* Included patients with bilateral retinal detachment due to CMV retinitis (n = 2), diabetic retinopathy (n = 3), myopia (n = 1), trauma (n = 1), and non–human immunodeficiency virus uveitis (n = 1) and 10 patients with idiopathic bilateral retinal detachment.

† In the group with myopic amblyopia, some patients with a combination of amblyopia and myopic retinal changes might have been included. The exact distinction between pure myopic amblyopia and myopic retinal degenerations and combinations was not feasible in this study.

CMV, cytomegalovirus; AMD, age-related macular degeneration; NA, not applicable

Table 4. Anatomical location of ocular disorders causing bilateral and unilateral blindness and low vision

Anatomical location	No. (%) of All Patients	No. (%) With Bilateral Blindness	No. (%) With Unilateral Blindness	P, Unilateral vs. Bilateral Blindness	No. (%) With Bilateral Low Vision	No. (%) With Unilateral Low Vision	P, Unilateral vs. Bilateral Blindness
Cornea	31 (8.4)	4 (6)	15 (12)	NS	2 (3)	10 (11)	NS
Lens	134 (36.3)	30 (41)	28 (22)	0.003	39 (51)	37 (41)	NS
Retina/ uvea	126 (34.6)	27 (37)	50 (39)	NS	18 (23)	31 (34.5)	NS
- *CMV retinitis*	42 (11.4)	14 (19)	12 (9)	0.044	9 (11.5)	10 (11)	NS
Optic disc	28 (7.6)	6 (8)	14 (11)	NS	3 (4)	5 (5.5)	NS
- *Glaucoma*	11 (3)	2 (3)	6 (5)	NS	2 (3)	1 (1)	NS
Whole globe	19 (5.1)	0	18 (14)	< 0.001	0	1 (1)	NS
Combinations of diverse disorders	12 (3.2)	5 (7)	NA		7 (10)	NA	
Non ocular	19 (5.1)	1 (1)	0	NS	8 (10)	6 (7)	NS
Total	369 (100)	73 (100)	129 (100)		77 (100)	90 (100)	

CMV cytomegalovirus; NS, not significant; NA, not applicable.

prognosis for patients with CMV retinitis and has dramatically reduced the risk of developing bilateral blinding disease in the industrialized world. The rates of vision loss among eyes with CMV retinitis observed in the era of HAART are approximately eightfold less than the rates reported in pre-HAART studies.[12,13] In Thailand, the prevalence of CMV retinitis was reported to be 21% (42/200 patients) in a cross-sectional study in 1996[14] and 33% in a prospective study of newly diagnosed HIV-positive patients in 2003.[15] Therefore, 33% of HIV-infected persons in Thailand can expect to develop CMV retinitis at some point during the course of their illness. Thailand's national program for access to antiretroviral treatment began in 2002, in which cheaper generic antiretroviral drugs are locally produced and distributed in all government hospitals through the Universal Coverage Project. Unfortunately, so far, not all HIV-positive patients receive HAART.[16] It is expected that locally produced HAART will be increasingly used and that the life expectancy of HIV-infected persons will increase. Hopefully, after implementation of HAART for HIV infected persons, CMV as a cause of blindness (with yet undefined magnitude in Southeast Asia) will decrease. However, some investigators have warned that the initial increase of CMV-induced blindness (related to a longer life for those who already have CMV retinitis) might be tremendous.[17]

Our study illustrates that the causes of blindness encountered in a tertiary center differ from those reported for the whole population, which is not surprising. Our study did not attempt to replace an epidemiologic survey on blindness but aimed to analyze the specific

disorders that the tertiary ophthalmologic centers are confronted with and, consequently, to determine which specific demands these centers should anticipate. We attempted to reveal the answers to questions entirely different from those of epidemiologic studies. The pattern of diseases encountered in a tertiary center undoubtedly reflects the local diagnostic and therapeutic possibilities; in addition, the referral patterns might also be influenced by age, availability of the center, and socioeconomic position of the patients. The patients attending tertiary centers are in part (self-selected) from a local area and referred from a much larger area. Although the basic medical coverage in Thailand also includes the elderly and the very poor, it is possible that these groups of patients might be underrepresented. The referral bias in the number of patients with acquired immunodeficiency syndrome and CMV retinitis is very high because tertiary centers are the only centers for treatment of CMV retinitis. In our population, the roles of glaucoma and age-related macular degeneration in blindness and low vision were negligible. The recent availability of therapeutic and surgical options for glaucoma might also have influenced the low number of cases of glaucoma-induced blindness in this university hospital. Future population studies will allow further analyses of visual impairment and blindness according to age and further distinctions, topics out of the scope of the present study.

The expertise of treating CMV retinitis in Southeast Asia is localized in tertiary centers. Our results indicate that precise data on the prevalence and incidence of blinding complications of HIV infection in Southeast Asia are urgently needed. Our study emphasizes the very important role of tertiary ophthalmologic centers in caring for HIV-infected persons with ocular disease in Thailand and probably all of Southeast Asia. Our results point out that retinal disorders, especially infections, play a major role in the cause of blindness encountered in tertiary centers in Thailand. Clearly, such institutions should anticipate the expected needs of their patient populations and prepare for taking care of large numbers of HIV-infected patients.

REFERENCES

1. Resnikoff S, Pascolini D, Etya'ale D, et al. Global data on visual impairment in the year 2002. Bull World Health Organ 2004; 82: 844–851. [Epub 2004].
2. Resnikoff S, Pararajasegaram R. Blindness prevention programmes: past, present, and future. Bull World Health Organ 2004; 82: 222–226. [Epub 2004].
3. Pascolini D, Mariotti SP, Pokharel GP, et al. 2002 Global update of available data on visual impairment: a compilation of population-based prevalence studies. Ophthalmic Epidemiol 2004; 11: 67–115.
4. World Health Organization. Status of Eye Health and Eye Care in the SEA Region, by Country, 2000 Annex 7 Health Situation in the South-East Asia Region, 1998-2000. Available at: http://w3.whosea.org/eip/tab61.htm
5. Jenchitr WT, Tansirikongkol VJ. Ophthalmology in Thailand: 1997. Med Assoc Thai 2000 ;83: 107–116.
6. World Health Organization. International statistical classification of diseases and related health problems. 10th Revision. Available at: http://www.who.int/classifications/en/
7. World Health Organization. Consultation on development of standards for characterization of vision loss and visual functioning. WHO report 2003. Available at: http://whqlibdoc. who.int/hq/2003/WHO_PBL_03.91.pdf
8. World Health Organization: Regional office for South-East Asia. Facts about HIV/AIDS in the South-East Asia Region. Available at: http://w3.whosea.org/en/Section10/Section18/Section348_9917.htm
9. UNAIDS. 2004 Report on the global AIDS epidemic. Available at: http://www.unaids.org/bangkok2004/GAR2004_html/ GAR2004_00_en.htm
10. Holbrook JT, Jabs DA, Weinberg DV, et al. Studies of Ocular Complications of AIDS (SOCA) Research Group. Visual loss in patients with cytomegalovirus retinitis and acquired immunodeficiency syndrome before widespread availability of highly active antiretroviral therapy. Arch Ophthalmol 2003; 121:99–107.
11. Goldberg DE, Smithen LM, Angelilli A, Freeman WR. Human immunodeficiency virus–associated retinopathy in the highly active antiretroviral therapy era. Retina 2005; 25: 633–649.
12. Holbrook TA, Jabs AD, Weinberg VD, et al. Visual loss in patients with cytomegalovirus retinitis and acquired immunodeficiency syndrome before widespread availability of highly active antiretroviral therapy. Arch Ophthalmol 2003; 121:99–107.
13. Thorne EJ, Jabs AD, Kempen HJ, et al. Studies of Ocular Complications of AIDS Research Group. Incidence of and risk factors for visual acuity loss among patients with AIDS and cytomegalovirus retinitis in the era of highly active antiretroviral therapy. Ophthalmology 2006; 113: 1432–1440.
14. Tanterdtam J, Suwannagool S, Namatra C, et al. A study of ocular manifestations in HIV patients. Thai Journal of Ophthalmology 1996; 10: 11–20.
15. Ausayakhun S, Watananikorn S, Ittipunkul N, et al. Epidemiology of the ocular complications of HIV infection in Chiang Mai. J Med Assoc Thai 2003; 86: 399–406.
16. Kitajima T, Kobayashi Y, Chaipah W, et al. Access to antiretroviral therapy among HIV/AIDS patients in Khon Kaen Province, Thailand. IDS Care 2005; 17: 359–366.
17. Guex-Crosier Y, Telenti A. An epidemic of blindness: a consequence of improved HIV care? Bull World Health Organ 2004; 82: 181. [Epub 2004].

CHAPTER 3

Uveitis in a tertiary ophthalmology centre in Thailand

Pathanapitoon K, Kunavisarut P, Ausayakhun S, Sirirungsi W, Rothova A.

British Journal of Ophthalmology 2008; 92: 474-8.

ABSTRACT

Purpose: To determine the aetiology and clinical characteristics of patients with uveitis in a tertiary ophthalmology centre in northern Thailand.

Methods: Standard ophthalmological examination and laboratory screening blood tests were performed in 200 consecutive new patients with uveitis. Patients were classified according to the location and aetiology of the uveitis. Specific clinical characteristics were recorded.

Design: Prospective case series.

Results: The case series included 106 male and 94 female patients with a mean age of 38 years. HIV associated uveitis was noted in 31% (62/200), and included mostly patients with cytomegalovirus retinitis (85%, 53/62). In the non-HIV group, the most common anatomical type was anterior uveitis (34%, 47/138). Infectious uveitis was diagnosed in 22% (30/138) of non-HIV patients, and toxoplasmosis was the most common infection (12/138, 8.7%). The most common noninfectious clinical entities were Vogt–Koyanagi–Harada disease (20%, 22/108) and HLA-B27-associated acute anterior uveitis (9%, 10/108).

Conclusions: The spectrum of uveitis in northern Thailand included 27% of HIV-infected patients with cytomegalovirus retinitis. Causes of non-HIV uveitis were similar to those often observed in the Far East, but the specific prevalences of these disorders were distinct from that found in India and Japan.

INTRODUCTION

Uveitis is a major cause of severe visual impairment throughout the world. It comprises a large group of diverse diseases affecting not only the uvea but also the retina, optic nerve and vitreous. Changes in the aetiology of uveitis over time have been brought about by effective treatment of specific infectious entities, such as tuberculosis and syphilis, and the emergence of new intraocular infections, such as cytomegalovirus (CMV) retinitis, related to HIV.[1] In addition, combinations of geographical, environmental, nutritional, socioeconomic, ethnic and genetic factors influence the causes of uveitis, and different patterns of uveitis are encountered around the world. The pattern of uveitis observed in the Far East differs considerably from that found in the West.[2,3] However, large differences with in Asian countries are also encountered, including differences in the prevalence of ocular toxoplasmosis and Leptospira infections.[4] In Thailand, data on the prevalence of different types of uveitis are lacking. We undertook a prospective study of 200 consecutive new patients with uveitis and determined their specific diagnoses on the basis of clinical manifestations and laboratory results.

PATIENTS AND METHODS

This prospective study of 200 consecutive new patients with uveitis was conducted at the Department of Ophthalmology, University Hospital in Chiang-Mai from January 2005 to November 2006. Each patient had a full ophthalmic examination, including slit-lamp biomicroscopy, tonometry, indirect ophthalmoscopy and medical history. Ancillary ophthalmic tests (eg, fluorescein angiography, visual field testing) were performed depending on the clinical characteristics of the patient. The terminology and classification of uveitis used, including anatomical criteria, are those given by the International Uveitis Study Group.[5]

Depending on the anatomical classification, all HIV-negative patients underwent the standard screening protocol for uveitis, which included erythrocyte sedimentation rate, red and white blood cell counts, serological tests for HIV, syphilis and Toxoplasma, urine analysis, and radiological chest examination. Human leucocyte antigen (HLA)-B27 typing was also performed in all patients with anterior uveitis. Laboratory and radiological analyses were not performed in patients with a first attack of anterior uveitis that reacted well to treatment (except patients with hypopyon or with uveitis persisting for longer than 3 months).[5] For several reasons (mainly logistical and financial), not all examinations could be performed on all patients. Ocular toxoplasmosis was diagnosed in patients with evidence of focal retinitis and positive Toxoplasma serology and in an additional two non-HIV patients who presented with focal active chorioretintis located adjacent to a chorioretinal scar, which is typical of ocular toxoplasmosis, and who also reacted well to treatment with antiparasitic drugs. These two pa-

tients with typical features of ocular toxoplasmosis had negative serological results at a dilution 1:231, which was the cut-off for positive Toxoplasma serology in our laboratory. However, low positive titres are regularly observed in patients with ocular toxoplasmosis, and therefore our serological findings do not exclude the possibility of ocular toxoplasmosis in these two patients. A diagnosis of ocular toxocarosis was based on a clinical finding of localised retinal granuloma. A diagnosis of Vogt–Koyanagi–Harada (VKH) disease was based on the revised diagnostic criteria.[6] A diagnosis of Behcet's disease was based on the International Study Group Classification criteria for Behcet's disease.[1] Selected patients (depending on their medical history, clinical presentation and ocular disease activity, as well as the outcomes of laboratory and radiographic screening procedures) underwent special tests and diagnostic procedures (''tailored approach''). Mantoux testing was not used as a screening test for the diagnosis of tuberculosis, as the prevalence of positives is, 80% in the normal Thai population, because of high exposure to tuberculosis and BCG vaccinations.[7] Mantoux testing, in addition to radiological chest examinations, was only performed in patients with clinical features suggesting a diagnosis of ocular tuberculosis (serpinginous chorioretinopathy and granulomatous uveitis).

At that time, HIV-positive patients did not undergo general screening for uveitis. HIV-positive patients who presented with ocular features compatible with a diagnosis of CMV retinitis were referred to a specialist in HIV disease for further examination and treatment. Therefore, only a limited number of HIV-positive patients underwent the additional tests in our department (erythrocyte sedimentation rate, red and white blood cell counts in 17 HIV-positive patients with symptoms of fatigue and other systemic symptoms; radiological chest examination in 12 HIV-positive patients with a chronic cough or dyspnoea). Syphilis and Toxoplasma serology were not systematically assessed in HIV patients with retinitis; the results were available for 40 and 39 patients, respectively. CMV retinitis was diagnosed by ophthalmological examination based on its characteristic presentation.[8] Immune recovery uveitis was diagnosed in eyes with previous and inactive CMV retinitis, which developed ocular inflammatory signs during highly active antiretroviral therapy (HAART)-induced immune recovery.[9]

We recorded the general, clinical and laboratory data. Chi-square test and Fisher's exact tests were used for statistical analysis, and $p < 0.05$ was considered significant.

RESULTS

Table 1 presents general data of the patients. All patients were Thai, although a few may have had (partial) Chinese ancestry. Of the 200 patients with uveitis, 62 (31%) were HIV infected (mean age 35.5 years; male/female ratio 1.8:1). In non-HIV patients with uveitis, the anterior location was the most common, followed by panuveitis and posterior uveitis, in contrast with HIV-positive patients, in whom the posterior location was most common (table 1; $p< 0.001$).

Table 2 gives the results of the diagnostic tests. Five of 62 HIV-positive patients were unaware of their HIV status at the onset of their ocular disease and were identified during the screening for the cause of their uveitis; the remainder asked for medical help for a combination of systemic and visual symptoms. None of the patients with active CMV retinitis was already receiving HAART medication when they consulted an ophthalmologist for the first time. The most informative test in non-HIV patients was HLA-B27 typing in those with anterior uveitis (tables 2 and 3); 12/25 (48%) of anterior uveitis cases were HLA-B27-positive. In the non-HIV group, syphilis serology was positive in 5/136 (4% and Toxoplasma serology was positive (titre above 1:231) in 10/136 (7%). Of the non-HIV patients with uveitis, abnormal results from the general blood examination were found in 7% (10/135), and none had an abnormal radiological chest examination.

In the HIV-positive group, 9/39 (23%) patients tested had positive syphilis serology and 9/40 (23%) had positive Toxoplasma serology results. Radiological chest examination was abnormal in 7/12 HIV-positive patients with a chronic cough or dyspnoea; this led to a diagnosis of tuberculosis in all seven.

Table 3 gives the results of various tests in relation to the anatomical type of uveitis. Positive HIV serology was more common with posterior uveitis, as well as abnormal general laboratory results and radiological chest examination (12/49 vs 9/103 ($p < 0.009$) and 6/47 vs 1/101 ($p=0.002$), respectively). In addition, syphilis and Toxoplasma serology were mostly positive in posterior uveitis (9/72 vs 5/103 ($p=0.067$) and 15/72 vs 4/104 ($p< 0.001$), respectively).

Table 4 gives the specific diagnoses and associations with systemic diseases for non-HIV patients with uveitis. The most commonly diagnosed infection was toxoplasmosis (12/138, 9% of all; 12/30, 40% of infections; 12/36, 33% of posterior uveitis) followed by toxocarosis (6/138, 4.3% of all; 6/30, 20% of infections) and herpetic viral infections (4/138, 2.9% of all; 4/30, 13% of infections). VKH disease was the most common disorder in non-infectious uveitis (22/138, 16% of all; 22/108, 18.5% of all with non-infectious uveitis). Idiopathic intermediate uveitis (pars planitis) was documented in 13/138 (9.5%) of all and 13/108 (12%) of all with non-infectious uveitis. HLA-B27-associated anterior uveitis was found in 10/138 (7.2%) of all, 10/108 (9.3%) of all with non-infectious uveitis, and in 10/47 (21%) of all with anterior uveitis. None of the patients had clinical features of birdshot chorioretinopathy. In all our masquerade cases, multiple chronic serous retinopathy (also named diffuse retinal pigment epitheliopathy) associated with serous retinal detachments was present, but with no involvement of the optic disc and no intraocular inflammatory signs; these cases were initially mistaken for VKH disease.[10]

The most common clinical diagnosis in HIV-positive patients was CMV retinitis (53/62, 85%) (table 5). Other infections included acute retinal necrosis (3/62, 5%) and ocular tuberculosis (2/62, 3%). Immune recovery uveitis was found in 3/62 (5%) of patients. HIV-positive

patients with uveitis had a higher prevalence of positive serological results for syphilis (9/39 (23%) vs 5/139 (4%), p< 0.001) and toxoplasmosis (9/40 (23%) vs 10/135, p=0.007) than non-HIV patients. However, the clinical ocular features of all these patients were compatible with the diagnosis of CMV retinitis.

Table 1. General characteristics of 200 consecutive new patients with uveitis in northern Thailand

	Total		Non-HIV uveitis		HIV-positive uveitis	
	N	%	N	%	N	%
Number of patients	200	100	138	100	62	100
Uni-to-bilateral ratio	1.1:1	n.a.	1.3:1	n.a.	1:1.2	n.a.
Male-to-female ratio	1:0.9	n.a.	1:1.1	n.a.	1.8:1	n.a.
Average age in years	38.4	(range 8-85)	39.6	(range 8-85)	35.5	(range 22-48)
Anterior uveitis	49	24.5	47	34	2	3
Intermediate uveitis	17	8.5	17	12	0	0
Posterior uveitis	92	46	36	26	56	90
Panuveitis	42	21	38	28	4	7

HIV: human immunodeficiency virus, n.a. not applicable

Table 2. HIV positivity in uveitis and results of screening

	Total		*HIV*		non *HIV*	
	N=200	%	N=62	%	N=138	%
Serology positive for *HIV*	62/200	31	62/62	100	0/138	0
Serology positive for *Treponema pallidum*	14/175	8	9/39	23	5/136	4
Serology positive for *Toxoplasma gondii*	19/176	7	9/40	23	10/136	7
HLA-B27 positive	13 /33	39	1/1	100	12/32	38
Abnormal general laboratory examination*	21 /152	14	11/17	65	10/135	7
Abnormal radiological chest examination	7/148	5	7/12	60	0/136	0

HIV: human immunodeficiency virus, HLA: human leukocyte antigen
Includes: erythrocyte sedimentation rate, red and white blood cell counts, urine analysis

Table 3. Anatomical types of uveitis and results of screening (all patients)

Examinations	Total		Anterior uveitis		Intermediate uveitis		Posterior uveitis		Pan-uveitis	
	N =200	%	N=49	%	N=17	%	N=92	%	N=42	%
Serology positive for *HIV*	62/200	31	2/49	4	0/17	0	56/92	61	4/42	9.5
Serology positive for *Treponema pallidum**	14/175	8	3/47	6	0/ 17	0	9/72	13	2/39	5
Serology positive for *Toxoplasma gondii**	19/176	11	0/47	0	0/17	0	15/72	21	4/40	10
HLA-B27 typing*	13 /33	40	12/25	48	0/3	0	1/3	33	0/2	0
General laboratory examination*	21/152	14	3/46	7	4/17	24	12/49	24	2/40	5
Radiological chest examination*	7/148	5	0/44	0	0/17	0	6/47	13	1/40	3

HIV: human immunodeficiency virus, HLA: human leukocyte antigen
*Due to logistic problems not all patients could be examined in all diagnostic tests

DISCUSSION

Our study shows that 31% (62/200) of new patients with uveitis in northern Thailand were HIV positive, of whom 53 had CMV retinitis. Therefore CMV retinitis was the most common diagnosis in the whole group (53/200, 26.5%). Infectious uveitis was present in 88/200 (44%) of all patients (58/62 (96%) in the HIV-positive group and 30/138 (22%) in the non-HIV group). The most common diagnoses in the non-HIV group were VKH disease, ocular toxoplasmosis, idiopathic intermediate uveitis and HLA-B27-associated anterior uveitis.

Anterior is the most common location of uveitis, particularly in Western countries, accounting for 50–60% of all cases in most tertiary referral centres and 90% in primary care.[2,3,11,12] In contrast, anterior uveitis is less prevalent (29–50%) in Asian countries, particularly Japan and Korea.[13,14] This may, in part, be explained by the high prevalence of Behcet's and VKH disease and the low frequency of the HLA-B27 haplotype in these areas.[13–15] In our study, posterior uveitis was the most common type, which is certainly due to the high number of HIV-positive patients with CMV retinitis. Many studies did not include CMV retinitis in their series on uveitis. In China, anterior uveitis was noted in 45% of patients, followed by panuveitis (41%), whereas intermediate and posterior uveitis were seldom seen.[16] A series from India found anterior uveitis to be the most common type (49%) followed by posterior uveitis (20%).[4,17] Among our non-HIV patients, anterior uveitis was present in 34%, and panuveitis and posterior uveitis in 28% and 26%, respectively; these percentages are similar to those in Western Europe and contrast with those in Japan and China.

Table 4. Diagnoses of 138 new consecutive patients with non-HIV uveitis in northern Thailand

Diagnosis	N	%
TOTAL	138	100
INFECTIOUS ORIGIN	30	22
Toxoplasmosis*	12	8.7
Toxocarosis	6	4.3
Herpes virus associated ARN	4	2.9
Presumed Ocular Tuberculosis	3	2.2
Syphilis	2	1.5
Endogenous endophthalmitis	2	1.5
CMV retinitis**	1	0.7
ASSOCIATION WITH SYSTEMIC DISEASE	36	26
Vogt Koyanagi Harada's disease	22	16
Behcet's disease	8	5.8
Psoriatic arthritis, HLA B27-	2	1.5
Ankylosing spondylitis, HLA B27+	1	0.7
Systemic lupus erythematodes	1	0.7
Relapsing polychondritis	1	0.7
Multiple sclerosis	1	0.7
ESTABLISHED CLINICAL ENTITIES	46	33
Idiopathic intermediate uveitis	13	9.5
HLA B27 associated AAU	10	7.2
Fuchs heterochromic uveitis	5	3.6
Neuroretinitis	4	2.9
Posner Schlossman's syndrome	3	2.2
Eales disease	2	1.5
Serpiginous chorioretinopathy***	2	1.5
Sympathetic ophthalmia	1	0.7
Multifocal chorioretinitis	1	0.7
Ocular masquerade syndromes	5	3.6
UNDETERMINED	26	19

HIV: human immunodeficiency virus, ARN: acute retinal necrosis, HLA: human leukocyte antigen, *CMV: cytomegalovirus,* AAU: acute anterior uveitis

* There were in total 12 patients with the diagnosis of ocular toxoplasmosis. All had focal chorioretinitis and 10 were also positive for *Toxoplasma* serology in a titer higher than 1:231. Two remaining patients tested negative in a dilution 1:231, however presented with focal active chorioretintis located adjacent to chorioretinal scar, which is typical of ocular toxoplasmosis and reacted well to the treatment with antiparasitic drugs and were therefore considered as having ocular toxoplasmosis.

** *CMV* retinitis in an immunosuppressed patient with acute myeloid leukemia.

*** These 2 patients had normal results of radiological chest examinations and showed no sighs or symptoms of systemic tuberculosis.

Table 5. Diagnoses of 62 new consecutive HIV positive patients in northern Thailand

Diagnosis	N	%
TOTAL	62	100
INFECTIOUS ORIGIN	58	94
CMV	53	91
HSV, VZV	3	5
Tuberculosis	2	4
NON –INFECTIOUS ORIGIN	4	6
IRU	3	5
HLA-B27 AAU	1	1.6

HIV: human immunodeficiency virus, HLA: human leukocyte antigen, *CMV: cytomegalovirus,* AAU: acute anterior uveitis, IRU: immune recovery uveitis

In this prospective study, infectious uveitis was present in 88/200 (44% of all patients studied). This relatively high percentage of infectious uveitis was mainly due to the large amount of CMV retinitis in HIV-infected patients The prevalence of HIV-associated uveitis (30% of all with uveitis) is higher than in the West and is influenced by the high number of HIV-positive people in the Thai population and also by the fact that ophthalmological treatment is concentrated in tertiary centres such as ours.[11,12,18] However, this percentage is similar to those in a study from the USA in the pre-HAART era (11–30%).[11,19,20] Recent data indicate that the prevalence of HIV infection in the whole population of Thailand is 1.5%,[21] and the prevalence of CMV retinitis in the HIV-infected population has been reported to vary from 21% to 33%.[22,23] A survey of blindness and poor vision in a tertiary centre in northern Thailand in 2005 noted that 19% of bilateral blindness was due to CMV retinitis in HIV-infected patients, which illustrates the high number of patients with HIV-associated ocular disease encountered in tertiary centres.[18] However, with the advent of antiretroviral treatment, the incidence of CMV retinitis has decreased dramatically in the areas in which the treatment is available, and it is probable that widely applied treatment in Thailand will have a similar effect in the coming years.[24]

Ocular infections were diagnosed in 22% of non-HIV patients, with ocular toxoplasmosis representing the most common infection. The prevalence of antibody to Toxoplasma in the Thai population has been reported to be 5–15%.[25,26] It was 7% (10/136) in our non-HIV uveitis series, increasing in patients with posterior uveitis to 28% (10/36; $p < 0.001$). In our study, presumed toxoplasmic retinochoroiditis was the leading cause of infectious uveitis (12/30 of infectious origin; 12/36 of posterior uveitis), which is similar to results from studies in the West and India, but higher than in Japan and China. The reason for the lower frequency of toxoplasmosis observed in some Asian countries is not known, but it may, in part, be due

to the lower rate of domestication of cats in these areas and alimentary factors.[2] The lack of diagnoses of ocular tuberculosis, herpetic infections and other more uncommon infections in our study may be explained by their absence or by our inadequate means of diagnosis. It is hoped that the expected introduction of PCR technology and Goldmann–Witmer coefficient in our laboratory will allow us to diagnose ocular infections more precisely in the future.

The high prevalence of VKH disease (16% of non-HIV positive patients) resembles the results on uveitis from Japan and China, but is higher than found in India.[4,14–17,27] In our study, none of the patients were diagnosed with ocular sarcoidosis. The prevalence of ocular sarcoidosis in uveitis patients has been reported to be low in most countries in Asia (none in Korea, 0.1% in China, 0.4% in Taiwan and 3.8% in India), in contrast with Japan, where it is similar (9.5%) to the prevalence in Western series.[2,3,11,13–17,28,29] In Thailand, sporadic cases of systemic sarcoidosis have been reported, but so far none with ocular involvement.[30] However, we cannot exclude the possibility that the diagnosis of ocular sarcoidosis was missed because of the limited availability of various diagnostic tests. The absence of typical lesions on chest radiography endorses the low frequency of this disease in northern Thailand. Several other uveitis entities observed regularly in the USA and/or Europe—specifically, presumed ocular histoplasmosis and birdshot retinochoroidopathy[11,12,28,29]—were not observed in our series. Furthermore, in multiple reports from Asia, no patients with HLA-A29-associated birdshot retinochoroidopathy were noted.[4,13–16] Data on the prevalence of HLA-A29 in the Thai population are not available. The exact pathogenesis of these entities is not yet known, nor are the reasons for the different prevalences.

In our study, the determination of HLA-B27 in anterior uveitis was a valuable test, as 48% (12/25) of patients with anterior uveitis were positive. This is higher than the prevalence of HLA-B27-associated anterior uveitis in India (~13%)[17] and Japan (< 35%),[2,3] but lower than in China (68%).[16] None of our non-HIV patients with uveitis had an abnormal radiological chest examination, and it could be argued that this examination is not a sensible test for the routine testing of all patients with uveitis in Thailand. Although we made a diagnosis of presumed ocular tuberculosis in three patients in the non-HIV group, their diagnoses were based on the clinical features of iris and/or choroidal granuloma and positive response to treatment with antituberculous drugs. None of these patients had an abnormal chest radiograph.

Obviously, there are several drawbacks to our study. Firstly, a selection bias is introduced by the referral pattern, which is similar to other series from tertiary centres. Further, the extent of diagnostic workup and availability of various investigations were limited and undoubtedly affected the pattern of uveitis observed by us. However, these results are the first step in recognising the causes of uveitis in Thailand, reflecting the demographic and causative pattern.

In conclusion, our study shows that the pattern of uveitis in a tertiary centre in northern Thailand is characterised by the high prevalence of CMV retinitis in HIV-infected, HAART-naïve patients. In addition, presumed ocular toxoplasmosis was the most commonly diagnosed infection and VKH disease the most common non-infectious type of uveitis in HIV-negative patients. It is hoped that future investigations of intraocular fluids will find additional causes of infectious uveitis and set up the possibility of targeted treatment.

REFERENCES

1. Nussenblatt RB, Whitcup SM. Uveitis: fundamentals and clinical practice. 3rd edn. Mosby: Philadelphia, 2004.
2. Chang JHM, Wakefield D. Uveitis: a global perspective. Ocul Immunol Inflamm 2002;10:263–79.
3. Wakefield D, Chang JHM. Epidemiology of uveitis. Int Ophthalmol Clin 2005;45:1–13.
4. Rathinam SR, Namperumalsamy P. Global variation and pattern changes in epidemiology of uveitis. Indian J Ophthalmol 2007;55:173–83.
5. Standardization Of Uveitis Nomenclature Working Group. Standardization of uveitis nomenclature for reporting clinical data. Results of the First International Workshop. Am J Ophthalmol 2005;140:509–16.
6. Read RW, Holland GN, Rao NA, et al. Revised diagnostic criteria for Vogt-Koyanagi-Harada disease: report of an international committee on nomenclature. Am J Ophthalmol 2001;131:647–52.
7. Thongngarm T, Valyasevi A, Pattanapanyasat K, et al. Delayed hypersensitivity skin testing in Thai adult population. J Med Assoc Thai 2006;89:195–200.
8. Studies of Ocular Complications of AIDS (SOCA) Research Group, AIDS Clinical Trials Group (ACTG). Studies of ocular complications of AIDS Foscarnet-Ganciclovir Cytomegalovirus Retinitis Trial. 1. Rationale, design and methods. Control Clin Trials 1992;13:22–39.
9. Nguyen QD, Kempen JH, Bolton SG, et al. Immune recovery uveitis in patients with AIDS and cytomegalovirus retinitis after highly active antiretroviral therapy. Am J Ophthalmol 2000;129:634–9.
10. Gass JD, Little H. Bilateral bullous exudative retinal detachment complicating idiopathic central serous chorioretinopathy during systemic corticosteroid therapy. Ophthalmology 1995;102:737–47.
11. Rodriguez A, Claonge M, Pedroza- Seres M, et al. Referral patterns of uveitis in a tertiary eye care center. Arch Ophthalmol 1996;114:593–9.
12. McCannel CA, Holland GN, Helm CJ, et al. Causes of uveitis in the general practice of ophthalmology. UCLA Community-Based Uveitis Study Group. Am J Ophthalmol 1996;121:35–46.
13. Chung H, Choi DG. Clinical analysis of uveitis. Korean J Ophthalmol 1989;3:33–7.
14. Yokoi H, Goto H, Sakai J, et al. Incidence of uveitis at Tokyo Medical College hospital. Nippon Ganka Gakkai Zasshi 1995;99:710–14.
15. Kotake S, Furudate N, Sasamoto Y, et al. Characteristics of endogenous uveitis in Hokkaido, Japan. Graefe's Arch Clin Exp Ophthalmol 1997;235:5–9.

CHAPTER 4

Infectious uveitis in Thailand: serologic analyses and clinical features

Sirirungsi W, Pathanapitoon K, Kongyai N, Weersink A, de Groot-Mijnes J, Leechanachai P, Ausayakhun S, Rothova A.

Ocular Immunology and Inflammation 2009; 17: 17–22.

ABSTRACT

Purpose: To determine the seroprevalence of various infectious agents in Thai patients with uveitis.

Methods: Prospective study of 101 consecutive patients with uveitis, 100 HIV-infected retinitis patients, and 100 nonuveitis controls.

Results: Antibodies against T. gondii were detected in 31/101 non-HIV patients, mostly with posterior uveitis and focal retinitis, and were significantly higher than in other groups examined. Antibodies for T. pallidum and Leptospira were observed more frequently in patients with HIV-infected retinitis. Active tuberculosis in non-HIV patients was not found.

Conclusions: Seroprevalence of T. gondii antibodies in patients with non-HIV posterior uveitis was higher than in nonuveitis controls and HIV patients with retinitis.

INTRODUCTION

Uveitis is a major cause of severe visual impairment in the world.[1,2] It is crucial to discriminate infectious from noninfectious causes of uveitis since their prognoses and treatment regimens are entirely different. Treatment of infectious uveitis with antibiotics may lead to improvement or even a cure of ocular disease and might even prevent further systemic involvement. The immunosuppressive treatment of infectious uveitis can be harmful, especially in areas where acquired immune deficiency syndrome (AIDS), tuberculosis, and other infectious diseases are endemic.[3] Infectious etiology was documented in at least 20–30% of all uveitis cases.[4] Therefore, it is essential that the diagnosis of intraocular infection is made before the immunosuppressive treatment is instituted.

In Thailand, cytomegalovirus (CMV) retinitis is a well known cause of retinitis in human immunodeficiency virus (HIV)-infected patients.[5,6] However, other causes of infectious uveitis in Thailand have not yet been systematically studied. Here we determine the seroprevalence of various infections known to cause uveitis in other parts of the world in 101 consecutive Thai patients with non-HIV uveitis and in 100 HIV-infected patients with retinitis and compare the results with 100 nonuveitis controls.

PATIENTS AND METHODS

We performed a prospective study on sera of 101 new consecutive patients with non-HIV uveitis and 100 consecutive HIV-infected patients with retinitis consulting the ophthalmology department of Chiang Mai University Hospital, which represents a large tertiary teaching center serving the whole of northern Thailand. In addition, 100 nonuveitis control subjects from the same geographical areas as the patients were included. The anonymous serum samples of controls were selected by age and gender using quota sampling from the remainder of the community health services samples at the Faculty of Associated Medical Sciences in Chiang Mai. Clinical data of the patients were registered, including gender, age at onset of uveitis, duration and laterality of uveitis, location and activity of uveitis, and associated systemic complaints and diseases. Whenever possible, fundus photographs of the eyes of the patients with uveitis were taken. The HIV-infected patients all had posterior uveitis with clinical features consistent with the diagnosis of cytomegalovirus retinitis and all were treated by administration of intravitreal ganciclovir injections. Seventy HIV-positive patients were sampled during the active stage of their ocular disease and 30 during the quiet phase following the local treatment with ganciclovir.

The patients who originally enrolled as having uveitis and nonuveitis controls were all tested for HIV by using Vironostika HIV Uni-Form II Ag/Ab (Biomerieux, France). The positive

samples were tested by gelatin particle agglutination test (Serodia.HIV, Japan). The samples positive in both tests were excluded from further analysis (1 in the uveitis group and 1 in the control group). The HIV status of patients with retinitis was examined at Maharaj Nakorn Chiang Mai University Hospital. Three methods were used to confirm HIV infection, including two ELISA-based methods, Enzygnost Anti-HIV 1/2 Plus (Dade Behring, Germany) and Elecsys HIV AG (Roche Diagnostics, Germany). The samples that gave positive results in both tests were tested by the immunochromatographic-based method, the Determine HIV -1/2 assay (Abbott, USA). Toxoplasma gondii (T. gondii)-specific IgG antibody in serum was investigated by enzyme-immuno-assay (EIA) with Enzygnost* Toxoplasmosis/IgG (Dade Behring) using the manufacturer's conditions. Optical density generated by any sample that was higher than the negative cutoff but lower than the positive value (equivocal result) was considered negative. All sera were screened for antibody specific to Treponema pallidum (T. pallidum) by using the immunochromatographic test BIOLINE Syphilis 3.0 (Pacific Biotech, Thailand). The borderline and positive samples were retested by using the ELISA-based method ICE* Syphilis (Abbott-Murex, USA). Only the samples reacting in both methods were considered positive and were tested with the Venereal Disease Research Laboratory test (VDRL: Becton Dickinson, USA), except for one specimen where insufficient volume precluded the VDRL testing. Leptospira antibody was determined in all samples by the fluorescent antibody test or by the SD Bioline Leptospira IgG test (Standard Diagnostics, Korea). The fluorescent antibody test was performed with leptospira-coated slides provided by the Department of Medical Science, Ministry of Public Health, Thailand, reacting with IgG to common leptospiral serotypes found in Thailand. Screening test-positive sera (titer ≥ 1:50) were confirmed using the microagglutination test (MAT) at the Department of Medical Science, Ministry of Public Health, Thailand. A MAT titer of ≥1: 400 was interpreted as current infection.[7] All commercial tests were performed according to the manufacturers' instructions. In addition, all non-HIV infected patients with uveitis were evaluated for the presence of tuberculosis using chest X-ray and sputum cultures.

The approval of the institutional review board was received and the study was completed with an anonymized data set of the controls. The data were computerized and statistically analyzed by Fisher's exact test; a value of $p < .05$ was considered significant.

RESULTS

The mean age of patients with non-HIV uveitis was 39 years (range, 9–85), of patients with HIV 37 years (range, 24–55), and of controls 40 years (range, 20–60). General characteristics of patients and controls are shown in Table 1.

The results of Toxoplasma, T. pallidum, and Leptospira serology are given in Table 2. Non-HIV uveitis patients had more frequently positive Toxoplasma serology than the con-

trols (31/101 versus 17/100, p = .023) and the HIV-positive retinitis patients (31/101 versus 19/100, p = .055). The non-HIV patients with posterior uveitis had the highest prevalence of positive Toxoplasma serology results compared to other anatomical types of uveitis (15/35, 43% versus 16/66, 24%; p=.05). Six out of 15 patients (40%) with posterior uveitis and positive Toxoplasma serology had focal retinal lesions compatible with the standard features of ocular toxoplasmosis

The rate of T. pallidum infection was significantly higher in HIV-positive patients with retinitis (13/100) compared to HIV-negative uveitis patients (5/101, p = .046) and nonuveitis controls (2/100, p = .003), however, it was not distinct from that of the non-HIV patients with posterior uveitis (13/100 versus 1/35, p = .09). Out of 13 patients with HIV-positive retinitis and positive EIA results, 5 were VDRL positive and considered to have a current infection. Positive T. pallidum serology was most common in HIV patients with active retinitis (12/70 compared to 1/30 inactive cases, p = .06). Antibodies against both T. gondii and T. pallidum were found in 5 HIV-positive patients (4 with active and 1 with inactive retinitis), but only 1 case with active retinitis was VDRL-positive. In 101 non-HIV uveitis patients, 5 patients showed the presence of anti-Treponema antibodies but only 4 samples were available for VDRL testing, 2 of which were positive. In nonuveitis controls, 2 of 100 samples were positive for both the anti-Treponema test and the VDRL.

A higher prevalence of Leptospira antibody in the HIV-positive retinitis group (9/100, 9%) was observed compared to the nonuveitis controls (2/100, 2%, p = .03). No differences in Leptospira serology were found between HIV patients with active and inactive retinitis (7/70 versus 2/30; p = .594) and between the non-HIV uveitis group and the controls (3/101 versus 2/100; p = .505). The MAT test was positive in only 1 non-HIV patient with panuveitis, in whom a positive current infection with Leptospira interrogans serovar australis was confirmed by a MAT titer of 1:400. None of the patients with non-HIV uveitis had evidence of active tuberculosis.

Table 1. General characteristics of patients with uveitis and nonuveitis controls

Characteristic	Non-HIV uveitis	HIV-positive uveitis	Nonuveitis controls
Number of patients	101	100	100
Average age in years (range)	39 (9-85)	37 (24-55)	40 (20-60)
Male-to-female ratio	52:49	46:54	50:50

Table 2. Seroprevalence of T. gondii, T. pallidum, and leptospira in HIV-negative uveitis, HIV positive uveitis, and nonuveitis controls

	No.	T. gondii		T. pallidum (N)		Leptospira b (N)	
		Number	%	EIA	VDRL	IFA or Bioline	MAT
HIV-negative uveitis	101	31	31	5	2[a]	3	1
Anterior	27	5	19	2	0	0	
Intermediate	14	4	29	0	0	0	
Posterior	35	15	43	1	1	1	0
Panuveitis	25	7	28	2	1[a]	2	1[b]
HIV-positive uveitis	100	19	19	13	5	9	0
Active	70	15	21	12	5	7	0
Inactive	30	4	13	1	0	2	0
Non-uveitis controls	100	17	17	2	2	2	0

Note: EIA, enzyme-immuno-assay; VDRL, Venereal Disease Research Laboratory test; IFA, immunofluorescence antibody test; MAT, microagglutination test.

[a] Insufficient volume precluded VDRL testing in one sample.

[b] MAT antibody titer was positive in a titre 1:400 to *Leptospira australiensis*

DISCUSSION

Our results point out the higher prevalence of positive Toxoplasma antibodies in patients with non-HIV uveitis (specifically in those with posterior uveitis) compared to non-uveitic controls or HIV-positive patients with retinitis. Our findings suggest that in Thailand, infection with Toxoplasma may play a role in the pathogenesis of non-HIV posterior uveitis. Although we demonstrated a higher frequency of positive T. pallidum and Leptospira antibodies in HIV-positive patients with retinitis compared to non-HIV uveitis and controls, the question of whether these microorganisms are really involved in the pathogenesis of their ocular diseases cannot be answered by the present study. The definitive proof of ocular infections would require the examination of intraocular fluids.

Infectious causes of uveitis differ largely around the world.[8] Mycobacterium tuberculosis and Toxoplasma were common infective causes of uveitis in India and Indonesia,[9,10] whereas Toxoplasma uveitis was strikingly low in Japan and China.[11,12] In Thailand, seroprevalence of toxoplasmic antibodies was investigated in pregnant women, blood donors, and kidney recipients, and seropositive rates of these groups varied from 10 to 15%,[13–15] which is lower than in Western Europe and similar to U.S. seroprevalence.[16,17] In China, where Toxoplasma uveitis is not observed, the seropositivity in the population is between 2 and 9%, with the exception of one study, which reported that Toxoplasma IgG prevalence among pregnant

women in Chengdu was 39%.[18] In previous reports, a marked difference in Toxoplasma IgG levels between Thai and Austrian pregnant women was also noted. These differences were attributed either to the different strain of virulences of T. gondii around the world or to the possibility that Thai women were infected at a younger age than Austrian women, which would also explain their lower IgG levels.[19] In addition, many other factors may be responsible for these differences, including the eating habits and presence of T. gondii in potential sources of human infections, and, not least, different definitions of positive and negative serologic cutoff points. Our study confirms the presence of the Toxoplasma antibody in 17% of the general population in northern Thailand. In Bangkok in 1995, toxoplasmic IgG was detected in 3.2% of healthy persons, in 12.5% of patients with ocular disease, and in 42.5% in HIV positive patients.[20] However, we found no significant difference between the prevalence of Toxoplasma antibody in HIV-infected retinitis patients (19%) and in the general population (17%), which is similar to a report of a Bangkok survey in HIV-positive and HIV-negative persons.[21] In contrast, 43% (15/35) of HIV-negative patients with posterior uveitis were positive for Toxoplasma IgG, which is higher than in other uveitis locations and indicates a high probability of toxoplasmic uveitis. A previous study from Singapore [22] revealed a similar distribution of Toxoplasma serology in patients with uveitis and controls. In addition, clinical features typical of ocular toxoplasmosis were noted in 40% of seropositive patients with posterior uveitis. The unusual forms of toxoplasmic chorioretinitis are increasingly being reported.[23] In addition, the combination of seropositivity for toxoplasma and uveitis in atypical cases could also be caused by the coincidence as 17% of controls were also positive. In addition, ocular toxoplasmosis in Thailand might have different characteristics and should be further studied.

Syphilis and leptospirosis are considered important causes of uveitis in developing countries. In our study, the evidence of prior infection with T. pallidum, Leptospira, and Mycobacterium tuberculosis was not common in non-HIV uveitis patients, which demonstrates that these microorganisms probably do not frequently cause uveitis in northern Thailand. In northern Thailand, the prevalence of T. pallidum antibodies seems to be stable according to a report of 2.7% in men and 2.1% in women in a 1998–2001 survey,[24] which is similar to our result of 2%. However, in our series, the prevalence of T. pallidum antibody was higher in HIV-infected retinitis patients (13/100 HIV) and could indicate that these patients might have ocular disease related to their syphilis. Although the design of our study precluded the confirmation or exclusion of syphilitic uveitis, our data show that HIV-infected patients with retinitis were more frequently exposed to T. pallidum infection than other groups studied. This finding is supported by a report conducted in the northern Thai population showing that persons having syphilis were 3 times more likely to have HIV.[24] Also in reports from the United States of America and Europe, the prevalence of ocular syphilis among HIV-infected patients in the HAART era is rising.[25–27] It might be possible that the association between HIV and syphilis

arises from the fact that they share common risk factors or that the ocular syphilis might be misdiagnosed in HIV-infected patients from Thailand. It is our opinion that the possibility of concurrent syphilis should be considered in all HIV-positive patients with intraocular inflammation. Further investigations into syphilitic uveitis in Thai HIV-infected patients are needed.

Data observed in this study showed that Leptospira is not a major cause of uveitis in the northern Thai population. Although leptospirosis was reported as an emerging infectious disease in Thailand, our data showed that only 2% of the general population was positive in the screening for Leptospira antibody. This low percentage might be explained by geographical difference, since the most cases (90% of all Thai infections) were reported in the northeastern region of Thailand.[28] So far, Leptospira uveitis was predominantly observed during the chronic stage of systemic Leptospira infection.[29] Only one HIV-negative patient with chronic bilateral panuveitis was proven to have a current infection with Leptospira australis, a strain being most commonly found in a survey of suspected leptospirosis cases in northeast Thailand.[7] His eye examination revealed panuveitis characterized by anterior chamber and vitreous cells, fine keratic precipitates, but no retinal lesions, clinical features that are compatible with the diagnosis of ocular leptospirosis.[30] The higher prevalence of Leptospira serology in HIV-positive patients is a surprising finding as Leptospira infection is not previously reported to be common in the HIV population.[31]

The majority of patients with uveitis are of working age and significant percentages suffer from associated systemic diseases, including poverty-associated infections.[32] Many infections can be treated and, therefore, it is important to identify the causative agents. Our finding of positive Toxoplasma serology in 43% of patients with posterior uveitis indicates that Toxoplasma maybe responsible for a significant number of posterior uveitis cases in non-HIV-infected patients in Thailand and those infections with T. pallidum and Leptospira are less common causes of uveitis in the non-HIV-infected population. However, occasional uveitis patients might suffer from these infections, as in our series 1 HIV negative patient had syphilis and 1 additional patient suffered from leptospirosis.

The best assessment of the definitive diagnosis of infectious uveitis currently lies in the analysis of intraocular fluid samples for DNA/RNA and antibodies by PCR and Goldmann-Witmer coefficient (GWC) analysis, respectively.[33] These diagnostic possibilities (and consequent treatments) are, however, not widely available, even in so-called developed countries, and their widespread implementation is challenging. Our data provide an insight into the etiology of infectious uveitis in Thailand and form a firm basis for future laboratory investigations for potentially causative microorganisms by the molecular and serological analyses of intraocular fluids.

REFERENCES

1. Durrani OM, Meads CA, Murray PI. Uveitis: a potentially blinding disease. Ophthalmologica. 2004; 218: 223–236.
2. Rothova A, Suttorp-van Schulten MS, Frits Treffers W, et al. Causes and frequency of blindness in patients with intraocular inflammatory disease. Br J Ophthalmol. 1996; 80: 332–336.
3. Dunn JP. Review of immunosuppressive drug therapy in uveitis. Curr Opin Ophthalmol. 2004; 15: 293–298.
4. Gritz DC, Wong IG. Incidence and prevalence of uveitis in Northern California: the Northern California Epidemiology of Uveitis Study. Ophthalmology. 2004; 111: 491–500.
5. Ausayakhun S,Watananikorn S, Ittipunkul N, et al. Epidemiology of the ocular complications of HIV infection in Chiang Mai. J Med Assoc Thai. 2003; 86: 399–406.
6. Pathanapitoon K, Ausayakhun S, Kunavisarut P, et al. Blindness and low vision in a tertiary ophthalmologic center in Thailand: the importance of cytomegalovirus retinitis. Retina. 2007; 27: 635–640.
7. Kusum M, Boonsarthorn N, Biaklang M, et al. Comparison of leptospira serovars identification by serology and cultivation in northeastern region, Thailand. J Med Assoc Thai. 2005; 88: 1098–1102.
8. Rathinam SR, Namperumalsamy P. Global variation and pattern changes in epidemiology of uveitis. Indian J Ophthalmol. 2007; 55: 173–183.
9. Salman A, Parmar P. Pattern of uveitis in a referral eye clinic in North India. Indian J Ophthalmol. 2004; 52: 345–346.
10. Suhardjo HN, Utomo PT, Agni AN. Clinical manifestations of ocular toxoplasmosis in Yogyakarta, Indonesia: a clinical review of 173 cases. Southeast Asian J Trop Med Public Health. 2003; 34: 291–297.
11. Goto H, MochizukiM, Yamaki K, et al. Epidemiological survey of intraocular inflammation in Japan. Jpn J Ophthalmol. 2007; 51: 41–44.
12. Yang P, Zhang Z, Zhou H, et al. Clinical patterns and characteristics of uveitis in a tertiary center for uveitis in China. Curr Eye Res. 2005; 30: 943–948.
13. Taechowisan T, SutthentR, Louisirirotchanakul S, et al. Immune status in congenital infections by TORCH agents in pregnant Thais. Asian Pac J Allergy Immunol. 1997; 15: 93–97.
14. Pinlaor S, Ieamviteevanich K, Pinlaor P, et al. Seroprevalence of specific total immunoglobulin (Ig), IgG and IgM antibodies to Toxoplasma gondii in blood donors from Loei Province, Northeast Thailand. Southeast Asian J Trop Med Public Health. 2000; 31: 123–127.
15. Sukthana Y, Chintana T, Damrongkitchaiporn S, et al. Serological study of Toxoplasma gondii in kidney recipients. J Med Assoc Thai. 2001; 84: 1137–1141.
16. Kortbeek LM, DeMelker HE, Veldhuijzen IK, et al. Population-based Toxoplasma seroprevalence study in The Netherlands. Epidemiol Infect. 2004; 132: 839–845.
17. Jones JL, Kruszon-Moran D, Wilson M. Toxoplasma gondii infection in the United States, 1999–2000. Emerg Infect Dis.2003; 9: 1371–1374.
18. Sun RG, Liu ZL, Wang DC. The prevalence of Toxoplasma infection among pregnant women and their newborn infants in Chengdu. Zhonghua Liu Xing Bing Xue Za Zhi. 1995; 16: 98–100.

19. Sukthana Y. Difference of Toxoplasma gondii antibodies between Thai and Austrian pregnant women. Southeast Asian J Trop Med Public Health. 1999; 30: 38–41.
20. Wongkamchai S, Rungpitaransi B, Wongbunnate S, et al. Toxoplasma infection in healthy persons and in patients with HIV or ocular disease. Southeast Asian J Trop Med Public Health. 1995; 26: 655–658.
21. Sukthana Y, Chintana T, Lekkla A. Toxoplasma gondii antibody in HIV-infected persons. J Med Assoc Thai. 2000; 83: 681–684.
22. Chee SP, Seah S, Ong SG, et al. Anti-toxoplasma serotitres in uveitis and ocular toxoplasmosis. Ann Acad Med Singapore.1992; 21: 226–229.
23. Smith JR, Cunningham ET. Atypical presentations of ocular toxoplasmosis. Curr Opin Ophthalmol. 2002; 13: 387–392.
24. Beyrer C, Kawichai S, Hyder JA, et al. Patterns of HIV and syphilis infection in Northern Thailand 1998–2001. Int J STD AIDS. 2007; 18: 179–183.
25. Gaudio PA. Update on ocular syphilis. Curr Opin Ophthalmol. 2006; 17: 562–566.
26. Balba GP, Kumar PN, James AN, et al. Ocular syphilis in HIVpositive patients receiving highly active antiretroviral therapy. Am J Med. 2006; 119: 448.e21–e25.
27. Westeneng AC, Rothova A, De Boer JH, et al. Infectious uveitis in immunocompromised patients and the diagnostic value of polymerase chain reaction and Goldmann-Witmer coefficient in aqueous analysis. Am J Ophthalmol. 2007; 144: 781–785.
28. Wuthiekanun V, Sirisukkarn N, Daengsupa P, et al. Clinical diagnosis and geographic distribution of leptospirosis, Thailand. Emerg Infect Dis. 2007; 13: 124– 126.
29. Rathinam SR. Ocular leptospirosis. Curr Opin Ophthalmol. 2002;13:381–386.
30. Rathinam SR. Ocular manifestations of leptospirosis. J Postgrad Med. 2005; 51: 189–194.
31. NevesES, PereiraMM,GalhardoMC, et al.Leptospirosis patient with AIDS: the first case reported. Rev Soc Bras Med Trop. 1994; 27: 39–42.
32. Suttorp-Schulten MS, Rothova A. The possible impact of uveitis in blindness: a literature survey. Br J Ophthalmol. 1996; 80: 844–848.
33. Groot-Mijnes DJ, Rothova A, Loon AV, et al. Polymerase chain reaction and Goldmann-Witmer coefficient analysis are complimentary for the diagnosis of infectious uveitis. Am J Ophthalmol. 2006; 141: 313–318.

CHAPTER 5

Infectious uveitis in Thailand: Intraocular fluid analysis by Polymerase Chain Reaction

Pathanapitoon K, Kongyai N, Sirirungsi W, de Groot-Mijnes J, Leechanachai P, Choovuthayakorn J, Kunavisarut P, Rothova A.

Submitted

ABSTRACT

Purpose: To evaluate the results of polymerase chain reaction analysis (PCR) for herpes simplex virus (HSV), varicella zoster virus (VZV), cytomegalovirus (CMV) and *Toxoplasma gondii* in intraocular fluid samples from patients with uveitis of unknown cause in Thailand.
Study design: Prospective cohort study.
Methods: One hundred human immunodeficiency virus (HIV)-negative patients and 47 HIV-positive patients with uveitis who consulted the Uveitis Clinic of Chiang Mai University Hospital, Thailand between May 2006 and October 2009. All patients underwent real time PCR analysis for CMV, HSV, VZV and *T. gondii* in their intraocular fluid samples and clinical features were registered.
Results: Positive PCR results in intraocular fluids were found in 66/147 (45%) of all patients with uveitis. The PCR positive results were lower in the HIV-negative patients (33/100; 33%) than in HIV-positive patients (33/47; 70%). CMV was the most common agent found in both HIV-negative and HIV-positive patients with uveitis (49% and 91%, respectively). In HIV-negative patients, CMV was predominantly associated with anterior uveitis while in HIV-positive patients CMV was mostly identified in posterior and panuveitis. Infections with *T. gondii* had a high prevalence of unusual clinical features.
Conclusions: PCR analysis of intraocular samples in uveitis was a valuable diagnostic assay and revealed positive results in 45% of patients with uveitis of unknown cause in Thailand. CMV was identified as the most frequent cause of infectious uveitis in both, HIV-negative and HIV-positive patients.

INTRODUCTION

Uveitis is an inflammatory eye disease which can be initiated by various infectious and non-infectious causes. Early recognition of specific infections is important as the treatment with antibiotics might stop the progression or even cure the eye disease and possibly also prevent involvement of other organs. Medical history and clinical manifestations are useful for making of the presumed diagnoses; however appropriate diagnostic tests are required for the definitive conclusions. The blood examinations in patients with infectious uveitis do not discriminate whether antibodies or DNA found are the result of a (previous) systemic infection or indicate a cause of active intraocular disease. Therefore, for the definitive diagnosis of infectious uveitis, the analysis of intraocular fluid is required. [1-3] In Thailand, analysis of intraocular fluids was not readily available; the lack of these novel diagnostic tests in local laboratories and the costs involved prevented systematic research on these ocular infections in this country.

The value of polymerase chain reaction (PCR) for the analysis of intraocular fluids has repeatedly been reported and was especially successful for the detection of viruses.[1-4] In this report, we evaluate the results of PCR analysis for cytomegalovirus (CMV), herpes simplex virus (HSV), varicella zoster virus (VZV) and *Toxoplasma gondii (T.gondii)* in intraocular fluid of 100 human immunodeficiency virus (HIV)-negative patients and 47 HIV-positive patients with uveitis of unknown origin examined in our center from May 2006 until October 2009 and assess the usefulness of PCR for diagnosis of uveitis in Thailand.

PATIENTS AND METHODS

We performed a prospective study in 100 new HIV-negative patients with uveitis and negative results of screening protocol and in 47 new HIV-positive patients with uveitis between May 2006 and October 2009 at the uveitis clinic of Chiang Mai university hospital in Thailand. All patients with uveitis underwent a tailored screening protocol which included chest X-ray (CXR) and various laboratory tests, including erythrocyte sedimentation rate (ESR) and blood counts, serology for HIV, *Treponema pallidum, T. gondii;* when appropriate, tuberculosis skin test and HLA-B27 typing were also determined. This study was performed with the approval of the local medical ethical committee. Uveitis was classified according the anatomic localization recommended by the SUN working group.[5]

Sample collection and processing:

Aqueous sampling in uveitis patients was performed as a subsequent diagnostic step in patients with negative initial screening. Aqueous tap was performed with the patient lying down and using lid speculum. Before the procedure, povidone iodine was used in all patients

and after the sampling, antibiotic eye drops were applied and the eye was patched for several hours. Vitreous analysis was performed in patients who required pars plana vitrectomy for different indications such as severe vitritis or retinal detachment. Control samples consisted of 22 intraocular fluid samples from patients without uveitis collected during the intraocular surgery for cataract and retinal detachment.

Intraocular fluid samples were stored at -80 °C in tubes within 12 hours of collection. All samples were analyzed for the presence of CMV, HSV, VZV and *T.gondii* by real-time PCR analysis. Nucleic acid was extracted from the ocular fluid by using the QIAamp DNA blood mini kit as recommended by the manufacturer (QIAGEN, Inc., Valencia, CA, USA). In addition, a standard amount of low concentration seal herpes virus type 1 (PhHV-1) was added to each sample at the beginning of the extraction procedure as an internal control. Since this virus does not infect humans, it does not interfere with the diagnostic assays.[6] By analyzing for PhHV-1, both extraction efficiency and real-time PCR inhibition could be monitored. Moreover, it enabled us to be more confident on negative result decisions.[6] Samples with an inhibited PhHV test result would be excluded. All extracted DNAs were kept at -20 °C until real-time PCR analysis. After nucleic acid extraction, each sample was separately tested by real-time PCR for the four pathogens of interest and PhHV-1. Briefly, 10 µl of extracted nucleic acid was added to 15 µl of real-time PCR reaction mixture (DyNAmo™ Probe qPCR kit, New England Biolabs Inc.) containing specific primers and probes.[1] Real-time PCR was performed in Chromo4™-Real-time PCR Detector Machine (DNA Engine®, BIO-RAD, USA) with repeated cycling program as previously described.[1] In each run, positive control DNA and negative controls were analyzed in parallel to the clinical samples.

RESULTS

Positive PCR results for HSV, VZV, CMV and/or *T.gondii* were found in 66/147 (45%) of all patients with uveitis. None of the samples were inhibited as determined by PhHV-1 analyses. The percentages of positive results was higher in the HIV-positive patients than in the HIV-negative patients ((33/47, 70% versus 33/100, 33%; P< 0.001). The results of PCR and clinical examinations are given in Table 1 and 2. All control samples were negative in PCR analyses for all microorganisms tested.

HIV-negative patients

In the HIV-negative group, anterior uveitis (AU) was the most common anatomical entity (34/100; 34%), followed by panuveitis (26/100; 26%), posterior uveitis (PU, 25/100; 25%) and intermediate uveitis (IU, 15/100; 15%; Table 1). A positive PCR result was found in 33/100 patients. Of these, the most common identified agent was CMV (16/33; 49%; 16/100,

16% of all), followed by *T.gondii* (7/33; 21%; 7/100, 7% of all). HSV and VZV were responsible for 10/33 (30%) of infections (10/100, 10% of all). CMV was the most common agent in all anatomical locations of uveitis except panuveitis where HSV was the most frequent. *T.gondii* infections were characterized by atypical findings (other than focal chorioretinitis) in 4/7 (57%) patients.

In HIV-negative AU, the most common agent was CMV (8/34; 24%); HSV-positive AU was not observed. Five out of the 8 (63%) CMV-positive AU cases were associated with ocular hypertension and/or glaucoma. Clinical features of 8 patients with CMV-positive AU included unilateral corneal endotheliitis (n=5) and unilateral chronic anterior uveitis (n=3) one of which had clinical characteristics of Fuchs heterochromic uveitis syndrome (FHUS). Clinical features of 3 AU patients who tested positive for *T.gondii* included unilateral AU of 4 months duration in 2 patients (Figure 1) and unilateral anterior uveitis with keratitis of 3 weeks duration in the remaining patient. All patients showed some of the clinical features compatible with the diagnosis of FHUS. All *T.gondii* positive patients with AU had keratic precipitates, but no synechiae; two patients had associated high IOP and cataract and one patient exhibited iris atrophy. All patients reacted well to symptomatic treatment with topical corticosteroids and disease subsided in all. The aqueous samples of the patients with 3 *T.gondii*-positive AU were retested with the same PCR results.

In patients with IU, the only infectious agent found was CMV (2/15; 13%, Table 1). Clinical features of these 2 CMV positive IU patients included bilateral vitritis without anterior segment involvement.

CMV was the most common agent found in PU (5/25; 20%), followed by *T.gondii* (3/25; 12%) and HSV (2/25; 8%; Table 1). Clinical features of 5 HIV-negative and CMV-positive PU patients included unilateral necrotic retinitis in 2 patients who received systemic steroids for non-ocular disorders (and were in consequence partially immunosuppressed; Figure 2 and 3). The other two HIV-negative PU patients who tested positive for CMV presented with unilateral retinal lesions and severe vitritis (age at onset 11 and 41 years, respectively; Figure 4); and the remaining 26-year-old patient presented with retinal vasculitis, severe vitritis and tractional retinal detachment. These three CMV-positive PU patients did not receive immunosuppressive medications and other causes or disorders associated with (partial) immunosupression were not observed. The clinical features of 3 PU patients with *T.gondii* included focal retinitis (n=2) and retinal vasculitis without any detectable retinal lesion in the remaining patient.

In patients with panuveitis, HSV was the most common agent (3/26; 11%) followed by VZV (2/26; 8%). Further, one patient tested positive for *T.gondii* and one for CMV. The clinical features of the 5 patients positive for HSV or VZV encompassed retinitis with clinical manifestations typical of ARN in all; in addition they all had vitritis and severe AU (Figure 5).

The CMV-positive but HIV-negative patient with panuveitis was partially immunosuppressed by mycofenolate mofetil treatment after renal transplantation and exhibited multiple retinal lesions and retinal vasculitis with hemorrhages. The patient positive for T. gondii manifested with primary retinal focal lesion with vitritis and anterior uveitis (Figure 6).

HIV-positive patients

In the HIV positive group, PU (22/47; 47%) and panuveitis (18/47; 38%) were the most common anatomical uveitis entities. Positive results were obtained in 33/47 (70%) of HIV-infected patients. CMV was the most frequent agent observed (30/33; 91%), followed by VZV (3/33; 9%), *T. gondii* (2/33; 6%) and HSV (1/33; 3%; Table 2). Three patients had double positive results which explains that the number of positive results (n=36) differed from the number of positive patients (n=33). Three double positive PCR results included one for CMV and HSV second for CMV and VZV, and the third for VZV and *T. gondii.*

Out of 4 HIV-positive patients with AU, one was positive for CMV. This patient was a 50-year-old, HAART-naive female with unilateral AU and secondary glaucoma. The etiology of AU in the 3 remaining HIV-positive patients remained undetermined. In PU and panuveitis, the most common infectious agent was CMV (16/22; 72% and 13/18; 72%, respectively).

Table 1. Polymerase chain reaction results of 100 HIV-negative patients with uveitis

Anatomical location of uveitis	Any positive PCR result		PCR positive for HSV		PCR positive for VZV		PCR positive for CMV		PCR positive for *T.gondii*	
	N	%	N	%	N	%	N	%	N	%
Total (n=100)	33/100	33	5/100	5	5/100	5	16/100	16	7/100	7
Anterior (n=34)	14/34	42	0/34	0	3/34	9	8/34	24	3/34	9
Intermediate (n=15)	2/15	13	0/15	0	0/15	0	2/15	13	0/15	0
Posterior (n=25)	10/25	40	2/25	8	0/25	0	5/25	20	3/25	12
Panuveitis (n=26)	7/26	27	3/26	11	2/26	8	1/26	4	1/26	4

Polymerase chain reaction (PCR); human immunodeficiency virus (HIV); herpes simplex virus (HSV), varicella zoster virus (VZV), cytomegalovirus (CMV) and *Toxoplasma gondii (T.gondii)*

Table 2. Polymerase chain reaction results of 47 HIV-positive patients with uveitis

Anatomical location of uveitis	Any positive PCR result*		PCR positive for HSV		PCR positive for VZV		PCR positive for CMV		PCR positive for *T.gondii*	
	N	%	N	%	N	%	N	%	N	%
Total (n=47)	33/47	70	1/47	2	3/47	6	30/47	64	2/47	4
Anterior (n=4)	1/4	25	0/4	0	0/4	0	1/4	25	0/4	0
Intermediate (n= 3)	0/3	0	0/3	0	0/3	0	0/3	0	0/3	0
Posterior (n = 22)	16/22	72	0/22	0	0/22	0	16/22	72	0/22	0
Panuveitis (n =18)	16/18	89	1/18	5	2/18	10	13/18	72	2/18	10

Polymerase chain reaction (PCR); human immunodeficiency virus (HIV); herpes simplex virus (HSV), varicella zoster virus (VZV), cytomegalovirus (CMV) and *Toxoplasma gondii (T.gondii)*
*Three patients had double positive PCR results which explains that the number of patients with any positive result (n=33) is different from the total number of positive PCR results (n=36)

DISCUSSION

Real time PCR on intraocular fluid samples of Thai patients with uveitis revealed positive results for HSV, VZV, CMV and/or *T. gondii* in 45% (66/147). CMV was the most common infectious agent found, specifically in 49% (16/33) of HIV-negative patients and in 91% (30/33) of HIV-positive patients with positive PCR results and was observed in all anatomical entities of uveitis. In HIV-negative patients, CMV was predominantly associated with AU while in HIV-positive patients, CMV was mostly identified in PU and panuveitis. The unexpected findings of this study include CMV-associated posterior uveitis in 3 immunocompetent HIV-negative patients and the frequent occurrence of atypical features in *T.gondii* infections.

The results of PCR studies in uveitis are extremely difficult to compare since the published series usually included selected populations of patients (e.g. with clinical features suggesting a specific cause of uveitis) and incorporated different combinations of studied agents as well as different PCR methods. Systematic PCR studies from Asia have, to our knowledge, not been published. In the West, an infectious etiology was documented in at least 20–30% of all uveitis cases [7-9], however clinical studies implied much higher percentages of infectious uveitis in Asia and Africa.[10-14] In our series, at least 45% of all patients with uveitis were of infectious origin (33% in HIV-negative and 70% of HIV-positive patients). A combined PCR and Goldmann-Witmer coefficient (GWC; intraocular antibody production) study from Europe demonstrated positive results by any assay in 23% (54/230) of immunocompetent patients; PCR solely was positive in 12% (28/230).[1] These results are much lower compared to 33% (33/100) found in our series of non-HIV infected patients. The explanation of this discrepancy might include the different selection criteria and definitions of the groups involved. However,

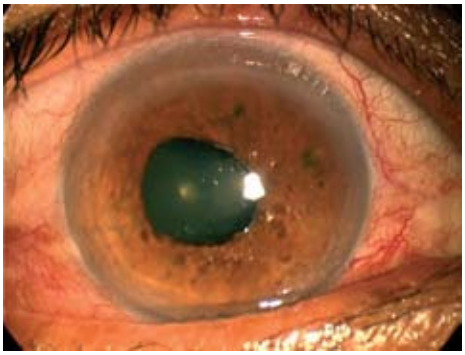


Figure 1: Patient with anterior uveitis and iris atrophy but without retinal lesions; aqueous analysis by polymerase chain reaction was positive for *T.gondii*, and negative for herpes simplex virus, varicella zoster virus and cytomegalovirus.

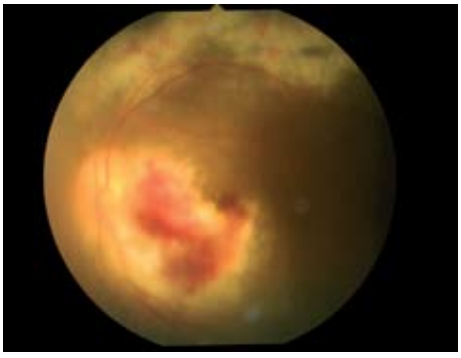


Figure 2: Unilateral necrotic retinitis in 39-year-old female who received systemic steroids for nephrotic syndrome and tested positive for cytomegalovirus in her intraocular fluid sample.

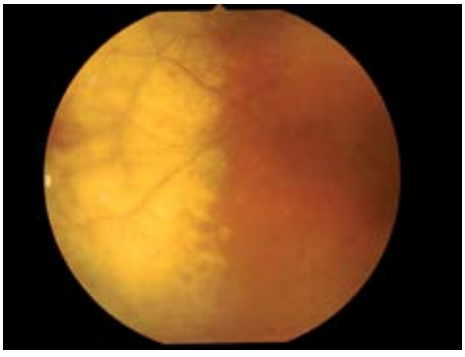


Figure 3: Unilateral acute retinal necrosis in a 59-year-old female who received systemic steroids for her non-ocular disease and tested positive for cytomegalovirus in her intraocular fluid sample.

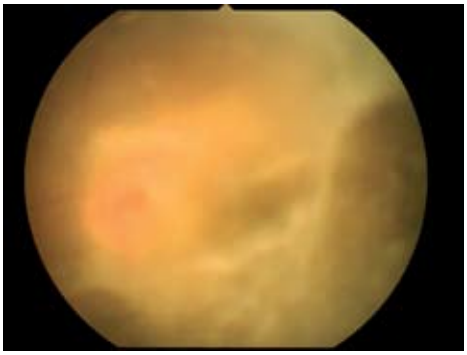


Figure 4: Unilateral posterior uveitis with retinal lesions and severe vitritis in a HIV-negative 41-year-old male who was not immunosuppressed and tested positive for cytomegalovirus in intraocular fluid.

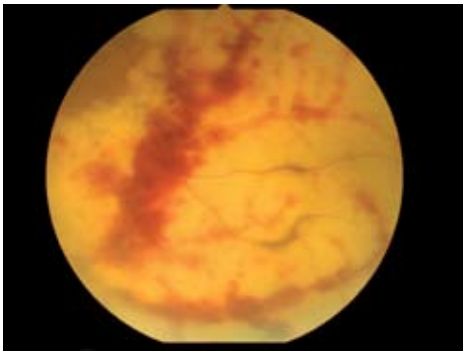


Figure 5. Acute retinal necrosis with associated vitritis and anterior uveitis in a human immunodeficiency virus-negative 42-year-old female who tested positive for varicella zoster virus in her intraocular fluid sample.

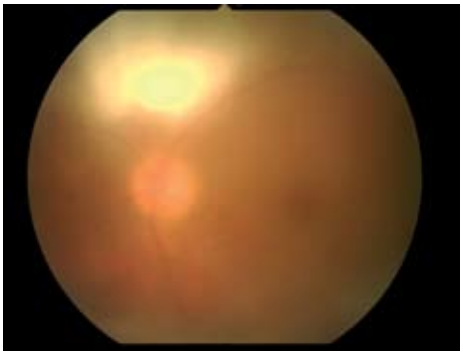


Figure 6. Focal chorioretinitis with vitritis and anterior uveitis but without old retinal scars in a human immunodeficiency virus-negative 50-year-old male who tested positive for *Toxoplasma gondii* in intraocular fluid sample.

genuine higher prevalence of infectious diseases in Thailand is also a possibility. A study from the USA of 133 immunocompetent and immunocompromised patients with posterior segment involvement has shown a prevalence of positive PCR results in 57% (76/133) which is in agreement with our findings of 54% (49/91) in our patients with posterior and panuveitis (both, HIV-negative and -positive).[4] In a similar study from Europe, positive PCR findings in posterior uveitis were found in only 10% (15/152).[3]

The most common agents causing uveitis in the West include HSV type1 and 2, VZV and Toxoplasma.[1,4] Clinical studies from Asia and Africa reported that the most common agents in uveitis were *T.gondii,* herpes viruses and *M.tuberculosis.*[10-14] Our previous study documented that tuberculosis was not a frequent cause of uveitis in Thailand.[15] Our present results indicate that CMV is the most frequent agent involved in both HIV-negative and HIV-positive uveitis patients in Thailand. Various studies suggested the higher incidence of ocular CMV infections in Asia and the high prevalence of CMV in AU (23%, 24/105) was previously reported in a study from Singapore.[16,17] CMV was the most frequent infectious agent found in all anatomical locations of uveitis with the exception of panuveitis. CMV was a cause of AU in 25% in HIV-negative patients, which is very similar to the 23% observed in Singapore.[17] Similar systematic PCR studies in AU patients from the USA and Europe have to our knowledge not yet been published, but the most cases of infectious AU are attributed to HSV.[17] The frequency of CMV as a cause of AU in the West remains to be determined.[18] CMV-associated AU was reported to cause a spectrum of ocular manifestations, varying in severity from a mild iritis to the more chronic and severe form AU. Patients had frequently associated corneal endotheliitis, diverse forms of iris atrophy and a high intraocular pressure.[17,18] Similar clinical manifestations were also observed in our patients. However, we also documented CMV-associated posterior segment involvement in 3 immunocompetent HIV-negative patients. The occurrence of CMV-retinitis in immunocompetent individuals was already occasionally reported.[19-21] Previous studies suggested that CMV retinitis might preferentially manifest in patients with insufficient immune response (e.g. in elderly patients, those with diabetes mellitus and/ or after splenectomy).[19,21] CMV retinitis was also observed in immunocompetent patients following intraocular triamcinolone application.[20] We did not identify any factors affecting the immune functions in our three HIV-negative patients with CMV-associated posterior segment involvement.

HSV and VZV were agents regularly involved in uveitis in many studies from different parts of the world.[4,11] In our series, HSV and VZV together were responsible for 30% of all infections in HIV-negative patients (15% each; Table 1), which is lower than 49% caused by CMV. Surprisingly, in our series, none of the AU patients was positive for HSV. This absence might be also due to the selection bias as the patients with typical clinical features of HSV-positive AU might not have been sampled. However, a low frequency of HSV-associated

uveitis was observed in Singapore (3/105 patients with AU) and does, like our results, suggest that HSV might be a less frequent cause of AU in Asia.[17]

Our findings demonstrate that *T.gondii* was the second most common pathogen in HIV-negative uveitis patients. Discrepancies on the prevalence of toxoplasmic uveitis were reported from Asia.[15] While *T.gondii* was a frequent cause of uveitis in Indonesia, it was virtually absent in China.[22,23] Low seroprevalence (5-15%) was previously reported from blood donors in Thailand.[24,25] In our previous study, we found that the seroprevalence of anti-toxoplasmic IgG in uveitis patients was higher (31%) than in non-uveitic controls (17%).[26] Noteworthy is also the high prevalence of atypical clinical features in our patients with ocular toxoplasmosis, but the classical forms of focal chorioretinitis also occurred. Less common presentations of ocular toxoplasmosis have been previously described and a possible involvement of *T.gondii* in the pathogenesis of FHUS was also reported.[27] *T. gondii* may proliferate in other parts of the eye and produce anterior uveitis, vitritis and occlusive retinal vasculitis.[28-31] Our series comprised 3 AU patients positive for toxoplasma PCR and one additional patient with retinal vasculitis without retinal lesions. AU in ocular toxoplasmosis was observed in a HIV-positive patient [28], however, all of our *T.gondii* positive AU patients were HIV negative and were not immunocompromised; in addition they all had some of the features compatible with the diagnosis of FHUS.[32] The uncommon manifestations of ocular toxoplasmosis might be more prevalent in the Asian population and therefore might not have been previously recognized as ocular toxoplasmosis.

PCR-positive results were more common in the HIV-positive patients (70% 33/47), which is consistent with previous findings.[1,2] In immunocompromised patients, especially in those with HIV infection, CMV was the most common pathogen involved in ocular disease, which is consistent with the findings from all over the world.[33] However, PCR examination of intraocular fluid revealed that 6/33 (18%) of HIV-positive patients were negative for CMV but positive for other pathogens. Diagnosis of CMV retinitis in Thailand is usually based on clinical findings solely, but overlapping funduscopic findings may occur. Furthermore, simultaneous infection of the retina by more than one pathogen underlines the usefulness of PCR analysis in HIV positive patients.

Unfortunately, all studies (including ours) performing the analyses of intraocular fluids have a strong selection bias. Obviously, less severe cases or patients with easily clinically recognized uveitis entities would not be sampled. In addition, European studies documented a superior diagnostic value of combined determination of intraocular antibodies (Goldmann-Witmer coefficient, GWC) together with PCR.[1] Unfortunately, GWC is a more difficult procedure to perform and is in many centers, including ours, not (yet) available. It was reported that PCR was usually positive in the early stage of the disease and predominantly in viral infections while GWC was more frequently positive in the later stages and in the non-viral causes. In

addition, novel causes of uveitis are regularly being reported and our study concerns only the four tested pathogens. The additional use of GWC and testing for more pathogens might reveal additional causes of infectious uveitis and change the relative percentage of causes of infectious uveitis in our population.

In conclusion, PCR examination of intraocular fluids was highly valuable in the diagnosing process of Thai patients with uveitis and demonstrated a high prevalence of positive results in HIV-negative as well as in HIV-positive uveitis population. Our study further revealed that CMV was the most frequent cause of infectious uveitis in both immunocompetent and immunosuppressed patients in Thailand and that ocular toxoplasmosis exhibited a high prevalence of atypical features. Further exploration of infectious causes of uveitis in Asia and their clinical manifestations seems to be warranted.

REFERENCES

1. De Groot-Mijnes JDF, Rothova A, Van Loon AM, et al. Polymerase chain reaction and Goldmann-Witmer coefficient analysis are complimentary for the diagnosis of infectious uveitis. Am J Ophthalmol 2006; 141: 313-8.
2. Westeneng AC, Rothova A, de Boer JH, de Groot-Mijnes JDF. Infectious uveitis in immunocompromised patients and the diagnostic value of polymerase chain reaction and Goldmann-Witmer coefficient in aqueous analysis. Am J Ophthalmol 2007; 144: 781-5.
3. Rothova A, de Boer JH, Ten Dam-van Loon NH, et al. Usefulness of aqueous humor analysis for the diagnosis of posterior uveitis. Ophthalmology 2008; 115: 306-11.
4. Harper TW, Miller D, Schiffman JC, Davis JL. Polymerase chain reaction analysis of aqueous and vitreous specimens in the diagnosis of posterior segment infectious uveitis. Am J Ophthalmol 2009; 147: 140-7.
5. Standardization of uveitis nomenclature (SUN) working group. Standardization of uveitis nomenclature for reporting clinical data. Results of the first international workshop. Am J Ophthalmol 2005; 140: 509-16.
6. Niesters HG. Quantitation of viral load using real-time amplification techniques. Methods 2001; 25: 419-29.
7. McCannel CA, Holland GN, Helm CJ, et al. Causes of uveitis in the general practice of ophthalmology. UCLA community-based uveitis study group. Am J Ophthalmol 1996; 121: 35–46.
8. Merrill PT, Kim J, Cox TA, et al. Uveitis in the southeastern United States. Curr Eye Res 1997; 16: 865-74.
9. Mercanti A, Parolini B, Bonora A, et al. Epidemiology of endogenous uveitis in north-eastern Italy. Analysis of 655 new cases. Acta Ophthalmol Scand 2001; 79: 64-8.
10. Ronday MJ, Stilma JS, Barbe RF, et al. Aetiology of uveitis in Sierra Leone, West Africa. Br J Ophthalmol 1996; 80: 956-61.
11. Wakabayashi T, Morimura Y, Miyamoto Y, Okada AA. Changing patterns of intraocular inflammatory disease in Japan. Ocul Immunol Inflamm 2003; 11: 277-86.
12. Singh R, Gupta V, Gupta A. Pattern of uveitis in a referral eye clinic in north India. Indian J Ophthalmol 2004; 52: 121-5.
13. Khairallah M, Yahia SB, Ladjimi A, et al . Pattern of uveitis in a referral centre in Tunisia, North Africa. Eye 2007; 21: 33-9.
14. Rathinam SR, Namperumalsamy P. Global variation and pattern changes in epidemiology of uveitis. Indian J Ophthalmol 2007; 55: 173–83.
15. Pathanapitoon K, Kunavisarut P, Ausayakhun S, et al. Uveitis in a tertiary ophthalmology centre in Thailand. Br J Ophthalmol 2008; 92: 474-8.
16. Madhavan HN, Priya K.The diagnostic significance of enzyme linked immuno-sorbent assay for herpes simplex, varicella zoster and cytomegalovirus retinitis. Indian J Ophthalmol 2003; 51: 71-5.
17. Chee SP, Bacsal K, Jap A, et al. Clinical features of cytomegalovirus anterior uveitis in immunocompetent patients. Am J Ophthalmol 2008; 145: 834-40.

18. Van Boxtel LA, van der Lelij A, van der Meer J, Los LI. Cytomegalovirus as a cause of anterior uveitis in immunocompetent patients. Ophthalmology 2007; 114: 1358-62.
19. Stewart MW, Bolling JP, Mendez JC. Cytomegalovirus retinitis in an immunocompetent patient. Arch Ophthalmol 2005; 123: 572-4.
20. Saidel MA, Berreen J, Margolis TP. Cytomegalovirus retinitis after intravitreous triamcinolone in an immunocompetent patient. Am J Ophthalmol 2005; 140: 1141-3.
21. Yoshinaga W, Mizushima Y, Abematsu N, et al. Cytomegalovirus retinitis in immunocompetent patients. Nippon Ganka Gakkai Zasshi 2008; 112: 684-7.
22. Suhardjo HN, Utomo PT, Agni AN. Clinical manifestations of ocular toxoplasmosis in Yogyakarta, Indonesia: a clinical review of 173 cases. Southeast Asian J Trop Med Public Health 2003; 34: 291–7.
23. Yang P, Zhang Z, Zhou H, et al. Clinical patterns and characteristics of uveitis in a tertiary center for uveitis in China. Curr Eye Res 2005; 30: 943–8.
24. Taechowisan T, Sutthent R, Louisirirotchanakul S, et al. Immune status in congenital infections by TORCH agents in pregnant Thais. Asian Pac J Allergy Immunol 1997; 15: 93–7.
25. Pinlaor S, Ieamviteevanich K, Pinlaor P, et al. Seroprevalence of specific total immunoglobulin (Ig), IgG and IgM antibodies to Toxoplasma gondii in blood donors from Loei Province, Northeast Thailand. Southeast Asian J Trop Med Public Health 2000; 31: 123–7.
26. Sirirungsi W, Pathanapitoon K, Kongyai N, et al. Infectious uveitis in Thailand: serologic analyses and clinical features. Ocul Immunol Inflamm 2009; 17: 17–22.
27. Smith JR, Cunningham ET. Atypical presentations of ocular toxoplasmosis. Curr Opin Ophthalmol 2002, 13: 387–92.
28. Rehder JR, Burnier Jr, MB Pavesio CE, et al. Acute unilateral toxoplasmic iridocyclitis in an AIDS patient. Am J Ophthalmol 1988, 106: 740–1.
29. Holland GN, Muccioli C, Silveira C, et al: Intraocular inflammatory reactions without focal necrotizing retinochoroiditis in patients with acquired systemic toxoplasmosis. Am J Ophthalmol 1999, 128: 413–20.
30. Braunstein RA, Gass JD: Branch artery obstruction caused by acute toxoplasmosis. Arch Ophthalmol 1980, 98: 512–3.
31. Theodossiadis P, Kokolakis S, Ladas I, et al: Retinal vascular involvement in acute toxoplasmic retinochoroiditis. Int Ophthalmol 1995, 19: 19–24.
32. Mohamed Q, Zamir E. Update on Fuchs' uveitis syndrome. Curr Opin Ophthalmol 2005; 16: 356–63.
33. Holland GN. AIDS and ophthalmology: the first quarter century Am J Ophthalmol 2008; 145: 397-408.

CHAPTER 6

Intraocular and plasma HIV-1 RNA loads and exploration of HIV-induced uveitis

Pathanapitoon K, Riemens A, Kongyai N, Sirirungsi W, Leechanachai P, Ausayakhun S, Ayuso VK, Kunavisarut P, de Groot-Mijnes J, Rothova A.

Submitted

ABSTRACT

Objective: To analyze human immunodeficiency virus (HIV) dynamics across the blood-retinal barrier and to determine whether the high levels of HIV in the eye are associated with any ocular disorders in HIV-infected patients.

Design: Prospective case series.

Participants: 40 HIV-positive patients with uveitis.

Methods: Clinical and laboratory examinations including plasma and intraocular HIV-1 RNA loads, polymerase chain reaction (PCR) for Cytomegalovirus (CMV), Herpes Simplex Virus (HSV), Varicella Zoster Virus (VZV) and *Toxoplasma gondii* and CD4 counts as well as the clinical manifestations of uveitis.

Main Outcome Measures: Results of aqueous analysis and ophthalmologic features and the correlations between the results of aqueous testing and clinical features.

Results: Intraocular HIV-1 RNA was detected in 33% (13/40) of HIV-positive patients with uveitis. Intraocular HIV-1 RNA loads were associated with high HIV-1 RNA plasma loads and not being on HAART therapy (P=0.000 for both, Mann-Whitney test). In addition, detectable intraocular HIV-1 RNA levels were associated with an inflammatory reaction within the eye (P=0.000, Fisher exact test) and with the absence of retinal lesions (P=0.000, Mann-Whitney test). In three patients, the HIV load in the eye largely exceeded that of plasma. These three patients had all bilateral anterior uveitis and/or vitritis without retinal lesions and exhibited no evidence of other intraocular infectious agents causing uveitis than HIV itself.

Conclusions: HIV can enter the eye and might cause an intraocular inflammatory reaction.

INTRODUCTION

In the past, ophthalmic disease in patients with human immunodeficiency virus (HIV) infection and acquired immunodeficiency syndrome (AIDS) represented primarily a result of progressive immune dysfunction and was mostly caused by opportunistic infections and malignancies common to HIV. Since the introduction of highly active anti-retroviral therapy (HAART), immune recovery uveitis (IRU) and ocular toxic and allergic drug reactions increased, while the prevalence of opportunistic infections decreased.

There is a growing concern about sanctuaries of local HIV replication as it has already been documented for the central nervous system (CNS).[1-3] HIV might persist in these sanctuaries during antiretroviral treatment and theoretically cause the generation and dissemination of drug-resistant viruses.

An ocular condition that is believed to be directly caused by HIV infection is retinal microvasculopathy.[4] In addition, HIV is a suspected cause of intraocular inflammation. Several cases of suspected HIV-associated uveitis have been presented, including an HIV-positive patient with anterior uveitis and an intraocular HIV-1 RNA load largely exceeding that of plasma and no evidence of other intraocular infectious agents causing uveitis other than HIV itself.[5-6]

We assess the plasma and intraocular loads of HIV in 40 HIV-infected patients and attempt to analyze HIV dynamics across the blood-retinal barrier and to determine whether the high levels of HIV in the eye are associated with any ocular disorders.

MATERIALS AND METHODS

We performed a prospective study in 40 HIV-positive patients with uveitis between December 2007 and October 2009 at the uveitis clinic of Chiang Mai university hospital in Thailand in which concurrent plasma and intraocular HIV-1 RNA loads were determined. We had samples from both eyes of 6 patients with bilateral uveitis, which resulted in total of 46 samples from 46 different affected eyes. We reviewed ophthalmologic data from all the patients and registered all ocular abnormalities, systemic and laboratory features at the time of aqueous sampling including the use of HAART and CD4 counts. The recent details on HAART medication and CD4 counts were not always available since the majority of patients were treated for their HIV infection outside our centre in peripheral hospitals. All patients were screened for syphilis using the TPHA (*Treponema pallidum* Haemagglutination test) and underwent a chest X-ray and sputum cultures when indicated. In addition, we determined intraocular loads of the 3 HIV-positive patients without uveitis.

In all patients with uveitis, paired intraocular fluid and plasma samples were taken for diagnostic purposes. In three of the HIV-positive patients without uveitis, the aqueous and

plasma samples, were collected during cataract surgery (n=2) and during glaucoma surgery (n=1).

In ocular samples, real-time polymerase chain reaction (PCR) for cytomegalovirus (CMV), herpes simplex virus (HSV), varicella zoster virus (VZV) and *Toxoplasma gondii* tests were performed. Prior to laboratory analyses, intraocular fluids were within 5 hours stored at - 80 C in sterile screw-cap tubes. Nucleic acid was extracted from 25 µl of ocular fluid with the QIAamp® DNA blood mini-kit (QIAGEN, Inc., Valencia, CA, USA). To monitor the quality of the extraction and the amplification procedure, a standard dose of Seal herpes virus type 1 (PhHV-1) was added to each sample as an internal control before extraction. The real-time PCR assays were performed on an automated thermal cycler (GeneAmp® PCR system 2700, Applied Biosystems, California, USA). Samples for which the internal control PCR was inhibited were excluded.

HIV loads in plasma and aqueous were assessed in the remainders of the diagnostic samples. Plasma and ocular fluid samples were extracted and analyzed by using the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test (Roche Molecular Systems, Inc., Branchburg, NJ, USA). To determine the respective HIV loads of the samples, the following methods were used. For the plasma samples 1050 µL was used. If less plasma was available the sample was diluted with negative plasma to reach the volume of 1050 µl. For each ocular fluid sample 25 µl was used and diluted with 1025 µL negative plasma. Further determinations and calculations were performed according to the instructions of the manufacturer. The detection limit of the plasma load was therefore 40 cp/ml and for the ocular fluid 1680 cp/ml (dilution factor being 42).

Multiple variables per patient and various groups of patients were compared, including the intraocular fluid and plasma HIV-1 RNA loads, age, gender, use of HAART, CD4+ T-lymphocyte levels and ocular characteristics. Differences in proportions among groups were compared by means of the chi-square and Fisher exact tests. The Mann-Whitney U test was used for comparison of the means of groups. P values < 0.05 were considered statistically significant. Our study was conducted with the approval of the medical ethical committee of Chiang Mai university hospital, Thailand.

RESULTS

Our series included 40 HIV-positive patients with uveitis (21 males and 19 females) with an average age of 41 years. Bilateral involvement was present in 22 patients. Ocular and laboratory data of the patients are shown in Table 1 and 2. The screening results for uveitis were within the normal limits except for two patients (one tested positive for syphilis serology, but had ocular features compatible with the diagnosis of CMV retinitis and positive CMV PCR

in aqueous; the other patient was diagnosed with pulmonary tuberculosis and presented with neuroretinitis).

Table 1. General characteristics of 40 HIV-positive patients with uveitis

	Total		Male-to-female ratio	Uni-bilateral uveitis ratio	HAART > 2 months		HIV-1 RNA plasma load > 40,000 copies/ml		CD4+ cells < 200 cells/µl	
	N	%			N*	%	N	%	N**	%
Total uveitis	40	100	1.1 :1	18:22	16/33	48	16/40	40	17/30	57
Intraocular HIV load not detectable	27/40	68	0.8 :1	11:16	14/20	70	4/27	15	9/18	50
Intraocular HIV load positive	13/40	32	2.25:1	7:6	2/13	15	12/13	92	8/12	67
Intraocular HIV load > plasma HIV load	4/40	10	3 :1	3:1	1/4	25	3 / 4	75	2/4	50

HIV: human immunodeficiency virus; HAART: highly active antiretroviral therapy
*Unknown in 7 patients
**Unknown in 10 patients

Table 2. Ocular features of 46 eyes with positive and negative HIV-1 RNA ocular loads

	Total		Absent retinal lesions		Intraocular inflammatory reaction		Intraocular PCR positive for opportunistic infection*	
	N	%	N	%	N	%	N	%
Total uveitis	46	100	6/46	13	29 /46	63	35/46	76
Intraocular HIV load not detectable	32/46	70	2/32	6	16/32	50	27/32	84
Intraocular HIV load positive	14/46	30	4/14	29	13/14	93	8/14	57
Intraocular HIV load > plasma HIV load	4/46	9	3/4	75	4/4	100	1/ 4	25

HIV: human immunodeficiency virus; PCR: polymerase chain reaction
*PCR was performed for cytomegalovirus, herpes simplex virus, varicella zoster virus and *Toxoplasma gondii*

Table 3. HIV-positive patients with uveitis and HIV-1 RNA ocular loads exceeding their HIV-1 RNA plasma loads

Patient Number	Age (Yr)	Sex	HAART > 2 months	HIV-1 RNA plasma load (copies/ml)	HIV-1 RNA ocular load (copies/ml)	Location	Inflammatory reaction	Retinal lesions	Opportunistic infections	CD4 count (cells /µl)	Intraocular PCR positive for infection*
1	40	F	no	169,000	89,800,000	Anterior chamber	yes	no	Not detected	342	negative
2	40	M	no	92,400	9,370,000	Anterior chamber, vitreous	yes	no	Not detected	247	negative
3	50	M	Yes**	540,000	2,460,000	Anterior chamber, vitreous	yes	no	Not detected	5	negative
4	35	M	no	512	2,200	Anterior chamber, vitreous, retina	yes	yes	CMV retinitis	100	CMV

HIV: human immunodeficiency virus; HAART: highly active antiretroviral therapy; PCR: polymerase chain reaction

*PCR was performed for cytomegalovirus (CMV), herpes simplex virus, varicella zoster virus and *Toxoplasma gondii*

**The laboratory data suggest an insufficient effect of the treatment or not using the medication. After the patient received new antiretroviral regimen (GPO-vir®), his HIV plasma load gradually decreased to non -detectable levels (the concurrent ocular load decreased to 1940 copies/ml), CD4+ cells increased to 165 cells/µl and uveitis disappeared.

Intraocular HIV-1 RNA loads were detected in 13/40 patients (32%; 14/46; 30% eyes). None of these 13 patients had systemic vascular diseases such diabetes mellitus. Detectable ocular loads were associated with positive plasma loads (eyes with detectable HIV-1 RNA ocular loads had median plasma loads of 169,000 copies/ml (cp/ml) and eyes with negative HIV-1 RNA ocular loads had median plasma loads of 233 cp/ml, P=0.000, Mann-Whitney; Table 1). Being on HAART for longer than 2 months was associated with lower intraocular loads levels (median intraocular HIV-1 RNA load was 3,820 cp/ml in HAART-negative patients and 0 in HAART-positive patients, P=0.000 Mann Whitney test; Table 1). Detectable intraocular loads were not related to gender or age of the patients (P =0.901 for both, Mann-Whitney test), nor to their CD4+ counts (P= 0.372, Mann-Whitney test). Intraocular HIV-1 RNA levels were positively related to the presence of intraocular inflammation (P= 0.000, Mann Whitney test) and were more frequently noted in eyes with absent retinal lesions P= 0.000, Mann Whitney test; Table 1). Positive association between the detectable intraocular HIV-1 RNA loads

and the presence of CMV retinitis (P=0.143, Fisher exact test) or the presence of intraocular opportunistic infections (P=0.06, Fisher exact test) were not identified.

Four out of 40 (10%) patients had ocular HIV-1 RNA loads higher than in their concurrent plasma samples and exhibited therefore a positive ocular-to-plasma ratio (Table 3). Three out of these four patients had uveitis located in the anterior segment and/or vitreous solely and had no retinal lesions or scars (Figure1 and 2). In these three patients, HIV was the only infectious agent detected within the eye and no other cause for uveitis could be identified. The remaining patient had retinal lesions consistent with the diagnosis of CMV retinitis and his aqueous PCR tested double positive for CMV and HIV-1 RNA. This patient had been on HAART for less than 2 months and his inflammatory ocular signs were clinically attributed to his recent immune recovery.

The 3 HIV-positive patients without uveitis were all on HAART and their levels in plasma and aqueous were below the detection limit.

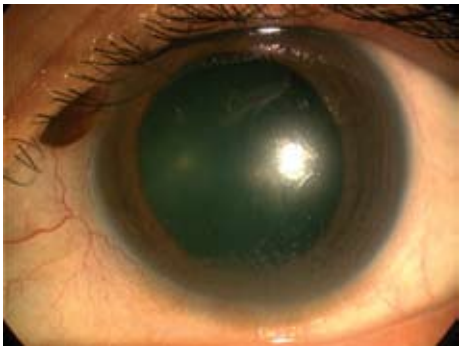


Figure 1. A 40-year-old HAART-naïve female with a newly diagnosed HIV infection presented with anterior uveitis and keratic precipitates in the left eye without vitreous and retinal lesions (Table 3, case no 1). The ocular HIV load largely exceeded that of the concurrent plasma HIV load. Anterior segment photograph of presumed unilateral HIV-induced uveitis demonstrates anterior chamber cells with moderate keratic precipitates.

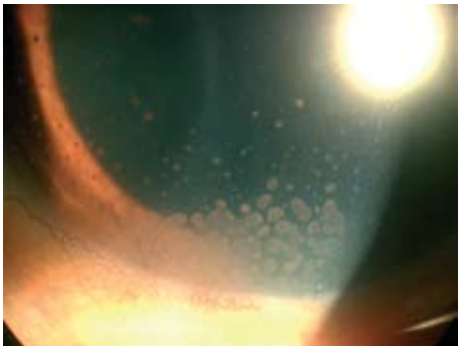


Figure 2. A 50-year-old HIV-positive male presented with bilateral anterior uveitis and vitritis without retinal lesions (Table 3, case no 3). The ocular HIV load largely exceeded that of the concurrent plasma HIV load. Anterior segment photographs of presumed HIV-induced uveitis exhibit inflammatory cells in anterior chamber with moderate-sized pigmented keratic precipitates. After the patient received HAART medication, his HIV plasma load gradually decreased to the not detectable levels (his concurrent ocular load decreased to 1,940 copies/ml), his CD4+ cells increased to 165 cells/μl and his uveitis disappeared.

DISCUSSION

We documented the presence of intraocular HIV in 32% of consecutive HIV-positive patients with uveitis in a tertiary center in Thailand. The presence of HIV in the eye showed a positive association with high HIV loads in the plasma and not receiving HAART. Furthermore, positive intraocular HIV-1RNA loads were associated with intraocular inflammation signs and the absence of retinal lesions. In 3 patients, the HIV load in the eye largely exceeded that of plasma. These 3 patients had all bilateral anterior uveitis and/or vitritis without retinal lesions and exhibited no evidence of other intraocular infectious agents causing uveitis than HIV itself.

HIV entrance into the eye was repeatedly reported and was attributed to the entrance of infected lymphocytes.[7] The virus has been demonstrated in retina, vitreous humour, iris, cornea, conjunctiva and tears.[8-14] Plasma viral loads and breakdown of the blood-retinal barrier were suggested as determinants of the positive intraocular HIV load.[15,16] Detectable intraocular HIV-1 RNA loads were generally observed in HIV-positive patients with infectious retinitis, who were not on HAART and had plasma HIV-1 RNA loads exceeding 100,000 copies/ml.[15,16] Being on HAART had a significant impact on aqueous humour HIV-1 RNA levels.[15,16] However, previous studies reported on intraocular HIV loads which were consistently lower than concurrent HIV-1 RNA loads in plasma. Although lower than in plasma, the intraocular HIV-1 RNA levels were reported to be higher in patients with opportunistic ocular infections than in patients with HIV retinopathy.[15] In our study, the intraocular HIV loads were also associated with higher HIV plasma loads and not being on HAART, which is entirely consistent with the previous reports. The absence of intraocular HIV-1 RNA in 27/40 (68%) patients might be explained by their low plasma viral load in combination with a well functioning blood-retina barrier; in addition the presence of low HIV-1 levels below the detection limit cannot entirely be excluded.

The striking finding in our series is that 4 of our patients had intraocular HIV-1 RNA levels higher than their corresponding HIV-1 RNA plasma levels. In 3 patients this concerned largely elevated ocular-to-plasma HIV ratio; all three had anterior uveitis and/or vitritis of unknown cause without associated retinal lesions. Of these 3 patients, only one was on HAART, but his high viral loads together with his CD4 positive count of 5 cells per µl document either an insufficient effect of the treatment or not using his medication. After the medication was changed and patient was urged to take his drugs, his plasma and ocular HIV-1 RNA loads decreased, CD4 count increased to 165 cells per ul and his intraocular inflammation subsided (Table 3, Figure 2). The high ocular-to-plasma HIV ratio and similar clinical manifestations in our three patients suggest that HIV can replicate itself within the eye and cause uveitis.

It has been already previously suggested that HIV of itself can cause uveitis.[5,6] A strong argument for HIV-induced uveitis rested on the positive ocular-to-plasma HIV ratio, and the fact that no other explanation could be given for ocular inflammation and the quick response of uveitis to treatment with antiretroviral therapy (HAART).[6] But it should be borne in mind that HAART might equally well halt the progression of opportunistic infection. However, our patients with elevated ocular-to-plasma HIV ratio had similar ocular manifestations (anterior uveitis, vitritis) which are not typical of opportunistic infection and further their aqueous analyses for opportunistic infections exhibited negative results.

Our study did not allow the comparison between the ocular HIV-1 RNA levels and the presence and severity of HIV vasculopathy. Future studies might clarify whether there are any associations between the ocular HIV-1 RNA loads and these vascular changes. Due to incomplete laboratory and clinical data, we did not find any positive correlation between the ocular HIV-1 RNA loads and the CD4 positive cell counts; however with more patients included this association might emerge.

The target cells for HIV-1 infection in the eye have not yet been identified. Possible candidates are retinal vascular endothelium and multinucleated giant cells of glial origin.[11,12] It has also been reported that human retinal pigment epithelial (RPE) cells may be an HIV-1 target and that in these cells, the HIV viral life cycle can be completed.[12] Further studies are needed to recognize the ocular cells in which HIV can replicate.

In summary, our results indicate that HIV may enter the inner eye and support the existence of HIV-induced uveitis. This entity should be suspected in HIV-positive patients with high HIV loads in plasma who present with uveitis without associated retinal lesions and test negative for opportunistic infections. Our study suggests that the eye can form a sanctuary where HIV might replicate itself and theoretically cause the dissemination of drug-resistant viruses. Further studies are needed to determine the clinical role and importance of the presence and replication within the eye of HIV.

REFERENCES

1. Persidsky Y, Poluektova L. Immune privilege and HIV-1 persistence in the CNS. Immunol Rev 2006; 213: 180-94.
2. De Luca A, Ciancio BC, Larussa D, et al. Correlates of independent HIV-1 replication in the CNS and of its control by antiretrovirals. Neurology 2002; 59: 342-7.
3. García F, Niebla G, Romeu J, et al. Cerebrospinal fluid HIV-1 RNA levels in asymptomatic patients with early stage chronic HIV-1 infection: support for the hypothesis of local virus replication. AIDS 1999; 13: 1491-6.
4. Pomerantz RJ, Kuritzkes DR, de la Monte SM, et al. Infection of the retina by human immunodeficiency virus type I. New Engl J of Med 1987; 317: 1643–47.
5. Rosberger DF, Heinemann MH, Friedberg DN, Holland GN. Uveitis associated with human immunodeficiency virus infection. Am J Ophthalmol 1998; 125: 301-5.
6. Rothova A, Schneider M, de Groot-Mijnes JD. Human immunodeficiency virus-induced uveitis: intraocular and plasma human immunodeficiency virus-1 RNA loads. Ophthalmology 2008; 115: 2062-4.
7. Pierson T, McArthur J, Siliciano RF. Reservoirs for HIV-1: mechanisms for viral persistence in the presence of antiviral immune responses and antiretroviral therapy. Annual Rev Immunol 2000; 18: 665-708.
8. Cantrill HL, Henry K, Jackson B, et al. Recovery of human immunodeficiency virus from ocular tissues in patients with acquired immune deficiency syndrome. Ophthalmology 1988; 95: 158-62.
9. Pham VT, Wen L, McCluskey P, et al. Human retinal microglia express candidate receptors for HIV-1 infection. Br J Ophthalmol 2005; 89: 753-7.
10. Rummelt V, Rummelt C, Jahn G, et al. Triple retinal infection with human immunodeficiency virus type 1, cytomegalovirus, and herpes simplex virus type 1. Light and electron microscopy, immunohistochemistry, and in situ hybridization. Ophthalmology 1994; 101; 270-9.
11. Reux I, Fillet AM, Fournier JG, et al. In situ hybridization of HIV-1 RNA in retinal vascular wall. Am J Path 1993; 143; 1275-79.
12. Canki M, Sparrow JR, Chao W, et al. Human immunodeficiency virus type 1 can infect human retinal pigment epithelial cells in culture and alter the ability of the cells to phagocytose rod outer segment membranes. AIDS Res Hum Retroviruses 2000; 16: 453-63.
13. Mueller AJ, Geier S, Gürtler L, Klauss V. HIV-1 and tears. Results of virus isolation and polymerase chain reaction (PCR). Ophthalmologe 1994; 91: 663-7.
14. Peng CH, Chen SJ, Ho CK, et al. Detection of HIV RNA levels in intraocular and cerebrospinal fluids in patients with AIDS-related cryptococcosis. Ophthalmologica 2005; 219: 101-6.
15. Ciulla TA, Schnizlein-Bick CT, Danis RP, et al. Comparison of intraocular to plasma HIV-1 viral burden in patients with cytomegalovirus retinitis. Am J Ophthalmol 1999; 127: 221–3.
16. Hsu WM, Chiou SH, Chen SS, et al. The HIV RNA Levels of plasma and ocular fluids in AIDS patients with ophthalmic infections. Ophthalmologica 2004; 218: 328-32.

CHAPTER 7

HLA-B27-associated acute anterior uveitis in the university referral centre in North Thailand: clinical presentation and visual prognosis

Pathanapitoon K, Suksomboon S, Kunavisarut P, Ausayakhun S, Wattananikorn S, Leetrakool N, Rothova A

British Journal of Ophthalmology 2006; 90: 1448–50.

ABSTRACT

Background: Acute anterior uveitis (AAU) is the most frequent type of uveitis encountered in the west. Although human leucocyte antigen (HLA)-B27-associated ankylosing spondylitis was reported in South East Asia, it is not known whether HLA-B27-associated ocular disease is prevalent in Thailand.

Methods: A prospective study of 100 unrelated blood donors and 121 consecutive patients with AAU was carried out. All people underwent HLA-B27 typing and full ocular examination. Radiological examination of the sacroiliac joints was conducted in patients with low back pain or arthralgias.

Results: The prevalence of HLA-B27 was 10% among the blood donors in contrast with 44% in the AAU group ($p< 0.001$). The clinical characteristics of HLA-B27-associated AAU were similar to those published throughout the world (unilaterality in 74%, hypopyon in 31%, recurrent AAU in 64%). However, the increased intraocular pressure (IOP) was more common in the HLA-B-27-negative group ($p = 0.03$) than in their HLA-B27-positive counterparts. At least 15% of the HLA B27-positive group had radiological signs of ankylosing spondylitis.

Conclusion: The prevalence of HLA-B27 in the population without uveitis in Thailand is about 10% and clinical characteristics of HLA-B27-positive AAU are similar to those reported in the west. In contrast with earlier reports, HLA-B27-negative AAU in Thailand was associated with increased IOP and should be further studied.

INTRODUCTION

Acute anterior uveitis (AAU) is the most frequent type of uveitis encountered in western Europe and the US, accounting for about 50% of all cases of uveitis.[1,2] In Asia, similar findings were reported for the Indian subcontinent, but by contrast, in Japan the frequency of AAU was reported to be low.[3,4] In most uveitis series, the prevalence of HLA-B27 in patients with AAU was about 50%, ranging from 19% to 82% across the different racial groups.[1,3,5,6] In Thailand, the presence and role, if any, of HLA-B27- associated AAU in patients with uveitis is not known. The purpose of this study was to assess the frequency of HLA-B27 in healthy people without AAU and in patients with AAU in northern Thailand and to describe the clinical features of HLA-B27-positive AAU in this particular population.

PATIENTS AND METHODS

We studied 100 unrelated Thai blood donors without history and clinical presentation of anterior uveitis, and 121 consecutive Thai patients with AAU who were examined at the ophthalmologic department of the University Maharaj Nakorn Hospital, Chiang Mai, Thailand, from January 2004 to December 2005. AAU was defined as anterior chamber inflammation of any cause that completely healed within 3 months. Patients in whom anterior uveitis was initially diagnosed, but which later become chronic, were not included in the study. Increased intraocular pressure (IOP) was considered in cases with measurements > 21 mm Hg. The complete ophthalmological examination was carried out in all blood donors and patients with AAU, and all patients were questioned about systemic complaints indicating the presence of human leucocyte antigen (HLA)-B27-associated systemic diseases. During the course of follow-up, all ocular complications were recorded, including the development of cataract, ocular hypertension or glaucoma, and posterior segment abnormalities. Clinical data including sex, age, clinical features at onset, management, follow-up time, ocular complications and visual outcomes were also registered, as well as results of HLA-B27 typing. HLA-B27 allele typing was carried out in all patients by a standard genotyping of DNA extracted from peripheral blood.[7] Radiological examination of sacroiliac joints was carried out in all patients with AAU with low back pain or arthralgias. The mean (range) follow-up duration was 20 (0–108) months. The data were analysed by calculating the descriptive statistics for the prevalence of HLA-B27 in blood donors without a history or presence of uveitis, and in the HLA-B27- positive and HLA-B27-negative AAU groups. The differences were assessed using Chi-square test and Student's t test where appropriate, and p values < 0.05 were considered significant.

RESULTS

The prevalence of HLA-B27 among the blood donors was 10% (10/100). The prevalence of HLA-B27 in the AAU group was 44% (53/121; p< 0.001) and the average age at onset of the uveitis was 38 years (median (range) 38 (4–67) years). The male:female ratio was 1.94:1 among the 53 patients with HLA-B27-positive uveitis and 0.94:1 among the 68 patients with HLA-B27-negative uveitis (p=0.05). HLA-B27-positive patients with AAU were slightly younger than HLAB27-negative patients (average age 37 v 39.5 years, not significant). The number of patients < 40 years was similar in both groups (28/53 v 34/68; p=0.75).

Table 1 shows the general and clinical features of HLAB27-positive and HLA-B27-negative patients with AAU at onset of uveitis. The HLA-B27-positive and HLA-B27-negative patients with AAU did not differ in initial visual acuity and eye involvement. However, the number of patients with severe AAU complicated by hypopyon was higher in HLAB27- positive patients (p=0.001). Increased IOP (> 21 mm Hg) was more common in the HLA-B27-negative patients than in their HLA-B27-positive counterparts (table 2). In our limited follow-up, the recurrent episodes were slightly more frequent in the HLA-B27-positive group (p=0.04). Development of glaucoma was observed in 10 of 53 (19%) HLA-B27-positive patients and in 21 of 68 (31%) HLA-B27-negative patients (p=0.133).

All patients were treated with topical corticosteroids according to the severity of the intraocular inflammation. Periocular corticosteroids were used for patients who developed posterior segment complications. A short course of systemic corticosteroids was given to patients resistant to local treatments (2 in the HLA-B27-negative v 0 in HLA-B27-positive group, p=0.1). The number of HLA-B27-positive patients with low back pain was [21], and radiological examinations showed signs characteristic of ankylosing spondylitis in 8 patients (8/53, 15% of patients in the HLA B27-positive group; 8/21, 38% of patients with low back pain in whom radiological examination was carried out). The diagnosis of psoriatic uveitis was made in two patients in the HLA-B27-negative group, but none of the HLA B27-negative patients and none of the blood donors had symptoms characteristic of ankylosing spondylitis.

Table 1. Clinical characteristics of HLA-B27-positive versus HLA-B27-negative patients at the onset of acute anterior uveitis

Characteristic at onset	HLA-B27 Positive AAU (N=53)		HLA-B27 Negative AAU (N=68)		P-value
	N	%	N	%	
Acuity less than 0.05	8	15%	10	15%	0.952
Unilateral AAU	39	74%	50	74%	0.995
Hypopyon	17	32%	5	7%	0.001

AAU, acute anterior uveitis.

Table 2. Complications and visual outcome of HLA-B27-positive and HLA-B27-negative patients with acute anterior uveitis

Characteristis	HLA-B27 POSITIVE AAU (N=53)		HLA-B27 NEGATIVE AAU (N=68)		P-value
Average duration of follow-up	19 months		22 months		
Visual outcome less than 0.05	1/50	(2%)	6/ 64	(10%)	0.104
Significant visual improvement*	12/50	(24%)	13/64	(20%)	0.637
More than one attack of AAU	23/36	(64%)	24/57	(42%)	0.041
Intraocular pressure above 21 mmHg	11/53	(21%)	27/68	(40%)	0.026
Glaucoma	10/53	(19%)	21/68	(31%)	0.133
Cataract	7/53	(13%)	12/68	(18%)	0.505
Posterior synechiae	5/53	(9%)	5/68	(7%)	0.680
Cystoid macular oedema**	1/53	(5%)	1/68	(3%)	0.859

AAU, acute anterior uveitis.
*Clinically significant visual improvement was defined as an improvement of more than 2 lines on the Snellen chart.
**The actual number of patients with (slight) CME might in reality be higher since not every patient could be extensively assessed due to the presence of synechiae and/or cataract.

DISCUSSION

Our study detected the prevalence of HLA-B27 in 10% of the northern Thai population. Further, we found that the HLAB27-associated AAU group formed about 43% of all patients with AAU, which is compatible with most published surveys on uveitis.[1,4,8–10] In addition, the clinical characteristics of HLA-B27 AAU in Thailand were similar to those published worldwide.[1,8–15] HLA-B27 exhibits considerable variation in prevalence in different populations, ranging from 1–6% in Japan to 50% among the Haida Indians of North America.[16–18] In Caucasian populations, the prevalence of HLA-B27 is about 8–10%. In northern Thailand, the frequency of HLA-B27 was previously reported to be about 4%, which approximates to our findings of 10%.[7,16] The HLA-B27-associated systemic diseases such as ankylosing spondylitis, reactive arthritis, and other disorders such as Reiter's syndrome and psoriatic arthropathy are prevalent in Thailand; however, their exact prevalence is as yet unidentified. In our study, at least 15% of HLA-B27-positive patients with AAU had ankylosing spondylitis, which is lower than in most previous studies.[1,8–15,19] The low numbers in our study may be (in part) explained by the fact that the radiological examinations were carried out only in patients with back pain or arthralgia and were not examined by a rheumatologist, and the real numbers of patients with ankylosing spondylitis might in fact be much higher. This was also the case with the HLA-B27-positive blood donors. The prevalence of HLA-B27-associated ocular disease in Thailand

has so far not been studied. Because the frequency of HLA-B27 found in this study was similar to those in Europe and the US, it is reasonable to expect that the prevalence of HLA-B27-associated diseases might be similar.

The prevalence of HLA-B27-associated ocular disease seems indeed to be similar to that of HLA-B27-associated AAU in the Thai population as almost half of all patients with AAU were HLA-B27 positive, a finding that is consistent with the findings from Europe and the US.[1,5,7–15] However, HLA-B27 represents a family of > 20 closely related alleles that differ at diverse amino acid positions. The prevalence of various subtypes of HLA-B27 varies in different races, and some subtypes play a stronger disease-predisposing part than other polymorphic positions.[18] The presence of the specific HLA-B27 subtypes that are only weakly associated with ankylosing spondylitis might also have contributed to the low prevalence of ankylosing spondylitis in our patients with HLA-B27 AAU. Other additional various factors such as infections with Gram-negative bacteria and their interaction with HLA antigens might also be involved in the pathogenesis of HLA-B27-associated systemic and ocular diseases.[20–22] It may be important to investigate further whether certain HLA-B27 subtypes show any preferential association with ocular disease, and vary between the various ethnic/racial populations and geographical regions of the world. Further, our patients were not assessed for the existence of additional HLA-B27-related systemic diseases such as reactive arthritis, inflammatory bowel disease and undifferentiated spondyloarthropathy. Financial and organisational causes precluded the addition of extensive rheumatological assessment to this initial study. However, we decided to incorporate the radiological examinations as these objectively show the presence of ankylosing spondylitis. Future studies on the HLA-B27 subtypes and extensive rheumatological analysis could further clarify the pathogenesis of HLA-B27-associated disorders.

The clinical characteristics of HLA-B27-associated AAU have been extensively reviewed in the literature and include mainly unilateral ocular involvement, sudden onset, marked fibrinous reaction and younger age of onset.[1,8–15,23] In addition, some authors indicated a poor visual prognosis, specifically when compared with the HLA-B27-negative patients with AAU.[12,13] For example, legal blindness was observed in HLA-B27-negative patients with AAU in only 2% versus 11% in HLA-B27-positive patients with AAU, and systemic treatment was required more frequently in HLAB27-positive patients.[12] By contrast, other authors observed different outcomes.[9,24] Undoubtedly, the results of the comparison also depend on the nature and outcome of HLA-B27-negative uveitis. The HLA-B27-negative AAU is an accumulation of known and unknown disorders and seems to be, at least in Europe and the US, associated with a good outcome. In our study from South East Asia, the observed clinical features of HLA-B27-associated AAU accord well with previous findings, including a male predominance and a high number of patients with severe acute features such as hypopyon. The HLA-B27-negative AAU, by contrast, seems to be characterised by more frequent complications, especially by raised

IOP. The number of patients with glaucoma in the B27-negative group exceeded slightly that with B27-positive AAU, but the difference was not significant. Definite conclusions concerning the development of glaucoma, however, cannot be made on the basis of this study owing to the limited follow-up, and difficult differentiation between the glaucoma and ocular hypertension because of the frequent presence of cataract or posterior synechiae. Outcomes of visual acuity cannot be exactly compared owing to the different criteria used by various studies. Our findings suggest that HLA-B27-negative patients in Asia might have a pathogenesis different from those in Europe and the US. The aetiology of HLA-B27-negative AAU is essentially undetermined, and may include various viral infections (often associated with increased IOP) and certainly requires further investigation.

In conclusion, we observed that the prevalence of HLA-B27 in a population without uveitis in Thailand, as well as the frequency and clinical characteristics of HLA-B27-positive AAU, is similar to that in populations in the US and Europe. Our findings indicate that the pathogenesis and causes of HLA-B27-negative uveitis in Thailand differ from those in the US and Europe and should be further studied.

REFERENCES

1. Chang JH, McCluskey PJ, Wakefield D. Acute anterior uveitis and HLA-B27. Surv Ophthalmol 2005;50:364–88.
2. McCannel CA, Holland GN, Helm CJ, et al. Causes of uveitis in the general practice of ophthalmology. UCLA community-based uveitis study group. Am J Ophthalmol 1996;121:35–46.
3. Sasaki T, Kusaba Y, Yamamoto T. HLA-B27 and acute anterior uveitis in the Japanese population. Jpn J Ophthalmol 1979;23:374–7.
4. Mehta S. Pattern of uveitis in a referral eye clinic in North India. Indian J Ophthalmol 2004; 52:121–5.
5. Huhtinen M, Karma A. HLA-B27 typing in the categorisation of uveitis in a HLA-B27 rich population. Br J Ophthalmol 2000;84:413–16.
6. Pathanapitoon K, Preechawai P, Visaetsilapanonta S, et al. Prevalence of HLA-B27 in acute anterior uveitis patients. Thai J Ophthalmol 2002;16:91–7.
7. Fongsatitkul L, Nantachit N, Kamtorn N, et al. HLA gene frequencies of northern Thais. J Med Assoc Thai 1997;80:38–42.
8. Wakefield D, Easter J, Penny R. Clinical features of HLA-B27 anterior uveitis. Aust J Ophthalmol 1984;12:191–6.
9. Rothova A, van Veenendaal GW, Linssen A, et al. Clinical features of acute anterior uveitis. Am J Ophthalmol 1987;103:137–45.
10. Tuncer S, Adam YS, Urgancioglu M, et al. Clinical features and outcomes of HLA-b27-positive and HLA-B27-negative acute anterior uveitis in a Turkish patient population. Ocul Immunol Inflamm 2005;13:367–73.
11. Wakefield D. Montanaro A, McCluskey P. Acute anterior uveitis and HLAB27. Surv Ophthalmol 1991;36:223–32.
12. Power JW, Rodriguez A, Pedroza-Seres M, et al. Outcomes in anterior uveitis associated with the HLA-B27 haplotype. Ophthalmology 1998;105:1646–51.
13. Monnet D, Breban M, Hudry C, et al. Ophthalmic findings and frequency of extraocular manifestations in patients with HLA-B27 uveitis: a study of 175 cases. Ophthalmology 2004;111:802–9.
14. Tay-Kearney LM, Schwam LB, Lowder C, et al. Clinical features and associated systemic diseases of HLA-B27 uveitis. Am J Ophthalmol 1996;121:47–56.
15. Beckingsale AB, Guss RB, Rosenthal AR. Acute anterior uveitis associated with HLA-B27 positive tissue type. A comparative study in two populations. Trans Ophthalmol Soc UK 1982;102:168–70.
16. Khan MA. HLA-B27 and its subtypes in world populations. Curr Opin Rheumatol 1995;7:263–9.
17. Konno Y, Numaga J, Tsuchiya N, et al. HLA-B27 subtypes and HLA class II alleles in Japanese patients with anterior uveitis. Invest Ophthalmol Vis Sci 1999;40:1838–44.
18. Reveille JD, Ball EJ, Khan MA. HLA-B27 and genetic predisposing factors in spondyloarthropathies. Curr Opin Rheumatol 2001;13:265–72.

19. Khan MA. Update on spondyloarthropathies. Ann Intern Med 2002;136:896–907.
20. Rosenbaum T. HLA B27-associated diseases. In: Pepose JS, Holland GN, Wilhelmus KR, eds. Ocular infection and immunity. St Louis: Mosby-Yearbook, 1996:475–84.
21. Reveille JD, Arnett FC. Spondyloarthritis: update on pathogenesis and management. Am J Med 2005;118:592–603.
22. Kim TH, Uhm WS, Inman RD. Pathogenesis of ankylosing spondylitis and reactive arthritis. Curr Opin Rheumatol 2005;17:400–5.
23. Ramsay A, Lightman S. Hypopyon uveitis. Surv Ophthalmol 2001;46:1–18.
24. Linssen A, Meenken C. Outcomes of HLA-B27-positive and HLA-B27-negative acute anterior uveitis. Am J Ophthalmol 1995;120:351–61.

CHAPTER 8

Ocular Sarcoidosis in Thailand

Pathanapitoon K, Goossens J.H.M., Tilborg TC, Kunavisarut P, Choovuthayakorn J, Rothova A.

Accepted for publication in Eye, 2010.

ABSTRACT

Purpose: To determine the prevalence of pulmonary sarcoidosis in patients with uveitis from the University Referral Centre in Thailand and to report on their clinical characteristics.
Methods: We performed a retrospective review of results of radiological examinations of 209 consecutive new patients diagnosed with uveitis. In patients with signs of pulmonary sarcoidosis, we reviewed clinical characteristics including age, gender, laterality and anatomical location of uveitis and complications.
Results: From 209 chest x-ray (CXR) examinations, one patient exhibited radiological signs typical of stage 1 sarcoidosis. Chest CT of 3 patients with posterior multifocal chorioretinitis (PMC) revealed abnormalities suggesting the diagnosis of pulmonary sarcoidosis. All PMC patients were females older than 50 years; they had no pulmonary complaints and their CXRs were without abnormalities.
Conclusion: Ocular sarcoidosis is prevalent in Thailand and our findings suggest that possibly more patients with sarcoidosis would be identified if diagnosis was thought of and ancillary tests were performed.

Keywords: Uveitis, Sarcoidosis, Thailand

INTRODUCTION

Sarcoidosis is a multi-systemic granulomatous disease that is characterised by non-caseating granulomas. The exact cause of this disease is still unknown, although it has been suggested that genetic, environmental factors or infectious agents may act as triggers.[1,2] Organs affected most often are the lungs, thoracic lymph nodes, skin and eyes. Sarcoidosis affects people of all racial and ethnic groups and can occur at all ages, however, there is a great variety of incidence and prevalence throughout the world. In the United States, the majority of patients are black with a prevalence of 40 per 100.000, compared to 5 per 100.000 among White Europeans.[1,3] In contrast to Japan, sarcoidosis is less common in India, South-East Asia, New Zealand and mainland China.[3] So far, it is not known whether the rarity of sarcoidosis in South-East Asia is genuine or whether sarcoidosis in this geographic area remains under-diagnosed. Prevalence of sarcoidosis among patients with uveitis in the West is approximately 8-10 % [4], in Japan 13% [5], and in India and Taiwan 4 % [4,6], but virtually no patients with ocular sarcoidosis have been identified in mainland China.[6, 7]

In Thailand, systemic sarcoidosis has rarely been diagnosed and so far, no cases with ocular sarcoidosis have been reported.[8,9] In this study, we explore the possibility of pulmonary sarcoidosis in 209 new consecutive patients with uveitis and describe ocular features of 4 Thai patients with signs of associated pulmonary sarcoidosis.

METHODS

We performed a retrospective review of the radiological examinations of 209 consecutive new HIV-negative patients diagnosed with uveitis in Chiang Mai university hospital between June 2006 and December 2008.

All patients with uveitis underwent a screening protocol which included chest X-ray (CXR) and various laboratory tests which encompassed erythrocyte sedimentation rate and complete blood counts, serology for human immunodeficiency virus *(HIV), Treponema pallidum, Toxoplasma gondii,* and human leukocyte antigen (HLA-B27) typing. In addition to the initial screening, a chest CT was performed in all patients with posterior multifocal choroiditis[10] (PMC, 3/209, 1.4%) and all with chronic vitritis in patients older than 50 years (4/209; 1.9%; Figure 1). Uveitis was classified according to the anatomical localization as recommended by the SUN working group.[11]

The purified protein derivative standard (PPD test) was performed in 23 patients and included all with abnormal CXRs suggesting (old) tuberculosis (TB; n=6) or sarcoidosis (n=1), abnormal chest CT suggesting sarcoidosis (n=4), clinical presentation of choroditis (n=5) or retinal vasculitis (n=7). Tissue biopsy was performed in two patients with PMC and normal

CXR, but a chest CT suggesting sarcoidosis. The remaining two patients with signs of pulmonary sarcoidosis (one on CXR and one on CT scan) refused the tissue sampling.

We also reviewed the ocular characteristics of patients with signs of pulmonary sarcoidosis revealed by radiological and chest CT, which included age, gender, laterality, anatomical location of uveitis, ocular manifestation, visual acuity and complications.

RESULTS

A total of 209 patients with uveitis were included in this study; 102 (49%) men and 107 (51%) women with average age of 40 years (range 8-85 years). Out of the total of 209 patients, 54 had anterior uveitis; 26 intermediate uveitis; 71 posterior and 58 panuveitis (Figure 1). One out of 209 patients (0.5%) had bilateral hilar lymphadenopathy typical of sarcoidosis stage 1. This 38-year-old male patient presented with blurred vision due to acute unilateral papillitis and with inflammatory reaction in the anterior segment and vitreous (Table). He had no systemic symptoms.

A chest CT was performed in 7 patients who presented either with PMC (n=3, all females) or chronic idiopathic vitritis in patients older than 50 years (n=4, all females). Of these 7 chest CTs, 3 patients with PMC exhibited bilateral abnormalities typical of the diagnosis of sarcoidosis (Table). All had bilateral hilar adenopathy, sometimes in combination with enlarged lymph nodes in paratracheal or other thoracal locations. Four elderly females with bilateral vitritis but without PMC had no abnormalities on chest CT and their definitive diagnoses were not clarified.

In two patients with PMC and positive CT scan, tissue biopsy from paratracheal lymph nodes was performed and revealed in both, noncaseating granulomas, negative for acid-fast bacilli with peroidic acid-Schiff and Gomori methenamine stains. All four patients with signs of pulmonary sarcoidosis had negative tuberculin skin tests. All four patients with suspected sarcoidosis were seen by a pulmonologist and TB was ruled out. According to the new diagnostic criteria for ocular sarcoidosis, two of our four patients fulfilled the criteria for a definitive and other two for a presumed diagnosis of ocular sarcoidosis.[12]

All three patients with positive chest CT findings were females aged older than 50 years who suffered from chronic PMC characterised by bilateral vitritis associated with peripheral retinal punched-out lesions (Table and Figure 2). All developed cystoid macular oedema (CME) and epiretinal membranes (Table). After pars plana vitrectomy (PPV) and membrane peeling their CME improved and their visual acuities fluctuated between 20/70 and 20/20 (Table).

In addition, one out of 209 patients (0.5%) was diagnosed with active pulmonary TB and TB-associated uveitis (choroidal granuloma with exudative retinal detachment).

Figure 1. Flow chart of patients indicating performed chest examinations. Radiologic examination of chest (CXR) was performed in all 209 patients and chest CT scan was performed in 7 patients including all with chronic bilateral vitritis (n=4) and all with posterior multifocal choroiditis (n=3).

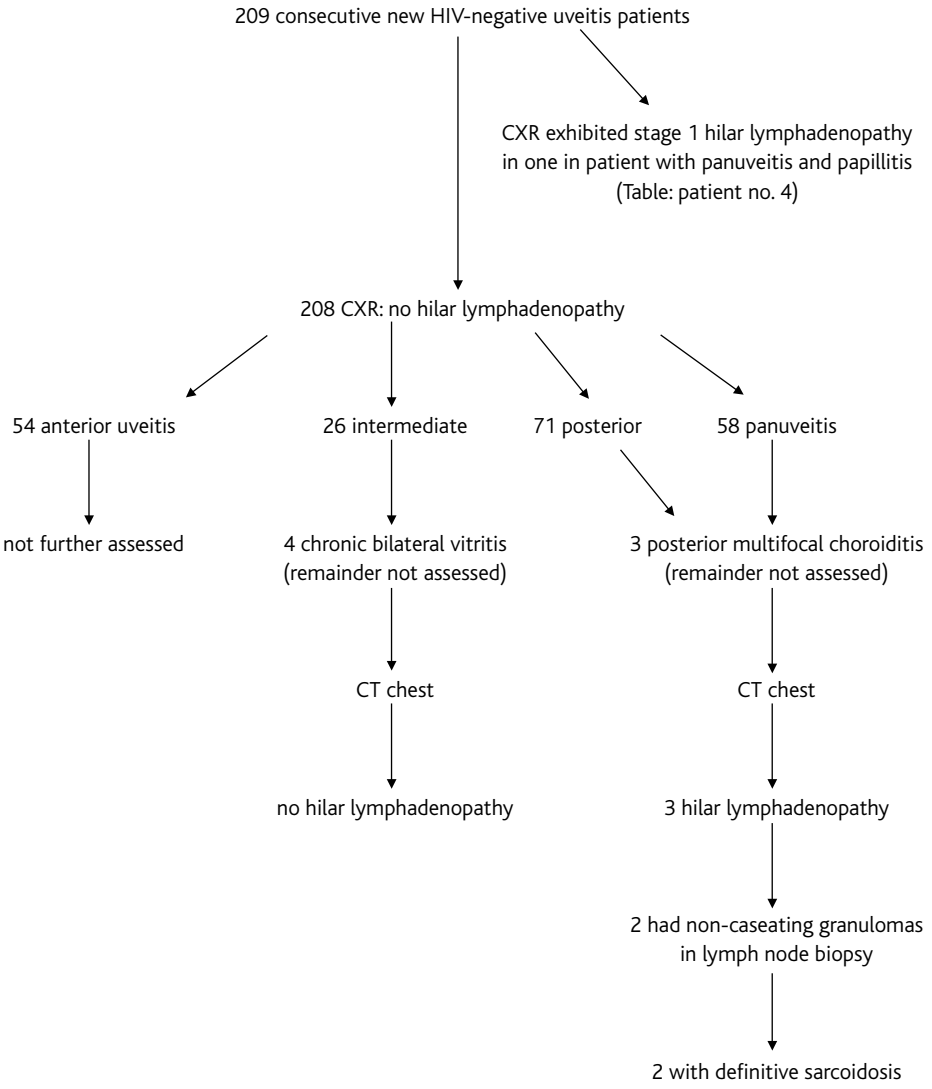


Table: Clinical characteristics of 4 Thai patients with uveitis and pulmonary signs of sarcoidosis

	Patient 1	Patient 2	Patient 3	Patient 4
Gender	Female	Female	Female	Male
Age of onset (yrs)	56	56	52	38
Chest X-Ray	No abnormalities	No abnormalities	No abnormalities	Bilateral hilar adenopathy
Chest computerized tomography	Bilateral hilar lymphadenopathy, enlargement of multiple lymph nodes in paratracheal, precarinal, subcarinal, and peribronchial regions.	Bilateral hilar lymphadenopathy, enlargement of multiple lymph nodes in paratracheal and peribronchial regions.	Bilateral hilar lymphadenopathy	Bilateral hilar lymphadenopathy and multiple lymph nodes in paratracheal, precarinal, subcarinal, and peribronchial regions.
Tissue biopsy	Noncaseating granulomas	Noncaseating granulomas	Not done	Not done
Duration of uveitis	Chronic	Chronic	Chronic	Acute
Laterality	Bilateral	Bilateral	Bilateral	Unilateral
Location [11] Keratic precipitated Clinical manifestations	Panuveitis Granulomatous Posterior multifocal choroiditis	Posterior - Posterior multifocal choroiditis	Posterior - Posterior multifocal choroiditis	Panuveitis Granulomatous Papillitis, mild vitritis
IWOS diagnosis [12]	Definite	Definite	Presumed	Presumed
Complications	Epiretinal membrane CME	Epiretinal membrane CME	Epiretinal membrane CME	None
Therapy	Periocular steroid injections PPV and membrane peeling	Systemic steroids (30 mg/d) and Azathioprine (100 mg/d) PPV and membrane peeling	Periocular steroid injections PPV and membrane peeling	Systemic steroids (60 mg/d)
Visual acuity 1 year after the onset of uveitis	20/40 ; 20/20	20/30 ; 20/30	20/20 ; 20/70	20/20 ; 20/20
Cause of decreased visual acuity	CME	CME	CME	Not applicable

CME cystoid macular edema, PPV pars plana vitrectomy

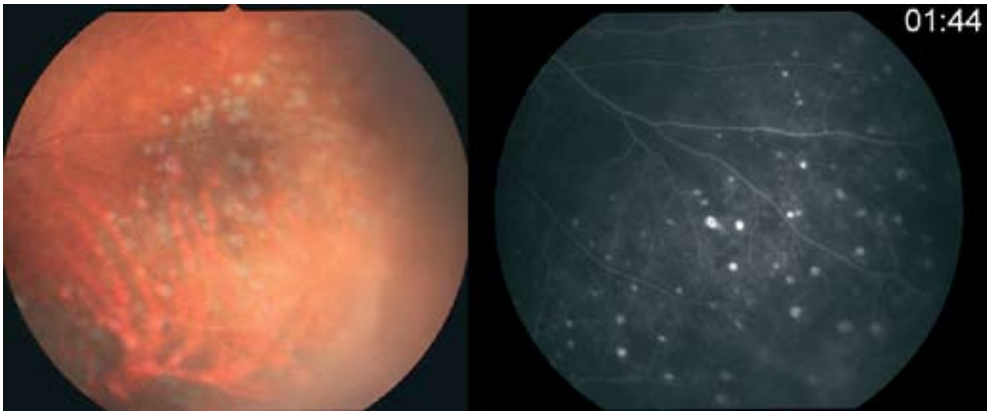


Figure 2: Fundus and fluorescein angiography photographs demonstrate multiple chorioretinal punched out lesions in the peripheral retina in elderly female Thai patients with hilar adenopathy on chest CT and histological diagnosis of sarcoidosis (Table, patient no 2).

DISCUSSION

Our study documents that pulmonary sarcoidosis is prevalent in the uveitis population in Thailand as one out of 209 (0.5%) new consecutive patients with uveitis had radiological signs of sarcoidosis and three additional patients exhibited bilateral hilar adenopathy typical of sarcoidosis on their chest CT. In two patients with an abnormal chest CT, tissue biopsy confirmed the diagnosis of definitive sarcoidosis. The patient with an abnormal CXR was a 38-year-old male with unilateral panuveitis and papillitis and three patients with negative CXR but positive chest CT were elderly females with chronic bilateral PMC.

In the West, the prevalence of sarcoidosis among the patients with uveitis is approximately 7% [3]. Our results indicate that at least 4 out of 209 (2%) uveitis patients suffered from pulmonary sarcoidosis. This percentage might be in reality even higher, if additional diagnostic tests such as chest CT were systematically applied. In Japan and China, a large increase in ocular sarcoidosis was noted over time.[5,6] It is not clear whether this increase is due to previous underdiagnosis or concerns a genuine increase of prevalence of ocular involvement. Sarcoidosis can remain subclinical for long periods of time and may also be mistaken for tuberculosis. It is possible, that sarcoidosis in Thailand might be unrecognized and/ or confused with TB; a similar phenomenon was also reported from India.[13]

The current major diagnostic test for the diagnosis of sarcoidosis is undoubtedly CXR, but for the definitive diagnosis of sarcoidosis a confirmation by tissue biopsy is required. In our study, tissue biopsy was attempted in 2 patients with PMC and confirmed the diagnosis in both. Unfortunately, due to non-medical reasons, tissue diagnosis could not be obtained

in the remaining two patients with pulmonary abnormalities and their diagnoses remained presumed.[12] The value of CXR for diagnosis of sarcoidosis in patients with uveitis was repeatedly debated, and it is currently agreed that the chest CT is more sensitive than the CXR.[2,14,15] In addition, serum ACE and lysozyme levels are useful for the screening of uveitis patients,[1,16] however, these serologic tests were not available in our institute. It is agreed that chest CT is superior and provides more details for assessing the pattern and extent of the disease compared to CXR.[2,14,15] Possibly, the systematic making of a chest CT in patients with a clinical suspicion of sarcoidosis might reveal additional cases. However, chest CT is not recommended for the initial screening of patients with uveitis for sarcoidosis, due to its high radiation and cost.[1] So far, studies systematically comparing diagnostic values of CT and CXR in uveitis are lacking and the indications for chest CT in patients with uveitis are not clear. It was reported that especially elderly females with PMC had frequently positive chest CT findings typical of sarcoidosis.[10,15] This observation is entirely in agreement with our findings.

The overall value of CXR in the initial screening for uveitis in Northern Thailand might be considered limited as only 2 out of 209 CXR (1%) were informative for the diagnosis of ocular diseases (one sarcoidosis and one active tuberculosis). The limited value of CXR in initial screening for uveitis was also reported in other series.[2,14,15] Future studies are needed to indicate the value of CXR in the initial screening of all patients with uveitis and to determine the exact role of chest CT in diagnostic work-up of patients with uveitis and presumed sarcoidosis.

The typical features of ocular sarcoidosis include uveitis (30-70%) and conjunctival nodules (40%).[17] Posterior uveitis with the involvement of the retina was most frequently seen in elderly white females while anterior uveitis was noted mainly in studies where the majority of the patients were black.[17] All of our patients had posterior segment involvement (100%), however this might reflect a selection bias as chest CT was performed only in 7 patients with bilateral PMC and/or vitritis. In Japan, posterior segment involvement was documented in more than half of the patients with ocular sarcoidosis.[18] PMC is a clinical entity primarily encountered in white elderly females. It is characterised by the presence of multiple, small, round, punched-out lesions in the peripheral retina and an associated intraocular inflammatory reaction.[10] Approximately one third of PMC patients had systemic sarcoidosis and the majority developed CME. [10,17,19] Similar manifestations were present in our three patients. Visual loss in sarcoidosis was predominantly noted in patients with chronic posterior uveitis and was mainly caused by CME (19-72%).[17] In India, however, CME was much less frequently found (7%) [13].

Differential diagnosis of ocular sarcoidosis includes mainly lymphoma and tuberculosis. The diagnosis of lymphoma was considered in all of our patients, however none had further symptoms or manifestations of lymphoma; in addition, all patients were assessed by a pul-

monologist. Two patients had definitive sarcoidosis diagnosed by tissue biopsy. No signs of lymphoma were noted in two patients with presumed sarcoidosis. The male patient (no 4) with presumed sarcoidosis had acute uveitis, which completely healed following a systemic course with corticosteroids and the hilar lymphadenopathy showed no further progression at one-year follow-up. The remaining patient with presumed sarcoidosis and PMC reacted well to periocular corticosteroid injections. No progression of ocular or systemic features was observed at one-year follow-up. Ocular tuberculosis in the wake of active pulmonary infection was diagnosed in one out of 209 (0.5%) HIV-negative uveitis patients. In our previous study, we identified presumed ocular tuberculosis in 3/138 (2%) of new uveitis patients who were HIV-negative.[8] Recently, the association between latent tuberculosis and uveitis was described.[20] The interferon gamma release assay such as the Quantiferon® test was not used in our series and therefore the number of patients with uveitis, which might have been related to latent tuberculosis, was not identified.

Our study has all the shortcomings of a retrospective study. It was not possible to perform chest CT scans in all patients with clinical features compatible with the diagnosis of sarcoidosis (e.g. all forms of chronic uveitis including vasculitis). We performed chest CTs only in 7 patients (elderly with chronic vitritis and all with PMC) and in consequence, the prevalence of sarcoidosis observed by us is probably underestimated. Due to non-medical causes, we were not able to perform the review of systems and other ancillary tests in all patients with uveitis.

In conclusion, our study confirms that sarcoidosis is prevalent in Thailand and points out that possibly more patients with sarcoidosis would be identified among the uveitis patients in the Far East, if the diagnosis was considered and ancillary tests performed. Future investigations are needed to clarify the exact prevalence, clinical spectrum and visual prognosis of ocular sarcoidosis in South-East Asia.

Acknowledgement: Juntima Euathrongchit M.D. for her advice in evaluation of radiological examinations.

REFERENCES

1) Michael C. Sarcoidosis. N Engl J Med 2007; 357: 2153-65
2) Margolis R, Lowder CY. Sarcoidosis. Curr Opin Ophthalmol 2007; 18: 470-5.
3) Sharma OP. Sarcoidosis around the world. Clin Chest Med 2008; 29: 357-63.
4) Rathinam SR. Global variation and pattern changes in epidemiology of uveitis. Indian J Ophthalmol 2007; 55: 173-83.
5) Goto H, Mochizuki M, Yamaki K, et al. Epidemiological survey of intraocular inflammation in Japan. Jpn J Ophthalmol 2007; 51: 41-4.
6) Chung YM. Uveitis with biopsy-proven sarcoidosis in Chinese-A study of 60 patients in uveitis clinic over a period of 20 years. J Chin Med Assoc 2007; 70: 492-96.
7) Yang P, Zhang Z, Zhou H, et al. Clinical patterns and characteristics of uveitis in a tertiary center for uveitis in China. Curr Eye Res 2005; 30: 943-8.
8) Pathanapitoon K, Kunavisarut P, Ausayakhun S, et al. Uveitis in a tertiary ophthalmology centre in Thailand. Br J Ophthalmol 2008; 92: 474-8.
9) Manonukul J, Wanitphakdeedecha R, Wisuthsarewong W, et al. Histopathologic aid to diagnosis of sarcoidosis: report of 8 cases. J Med Assoc Thai. 2006; 89: 864-71.
10) Lardenoye CW, Van der Lelij A, de Loos WS, et al. Peripheral multifocal chorioretinitis: a distinct clinical entity? Ophthalmology 1997; 104: 1820-6.
11) Standardization of uveitis nomenclature (SUN) working group. Standardization of uveitis nomenclature for reporting clinical data. Results of the first international workshop. Am J Ophthalmol 2005; 140: 509-16.
12) Herbort CP, Rao NA, Mochizuki M; members of Scientific Committee of First International Workshop on Ocular Sarcoidosis. International criteria for the diagnosis of ocular sarcoidosis: results of the first International Workshop On Ocular Sarcoidosis (IWOS). Ocul Immunol Inflamm 2009; 17: 160-9.
13) Khanna A, Sidhu U, Bajwa G, et al. Pattern of ocular manifestations in patients with sarcoidosis in developing countries. Acta Ophthalmol Scans 2007; 85: 609-12.
14) Lynch JP 3rd. Computed tomographic scanning in sarcoidosis. Semin Respir Crit Care Med 2003; 24: 393-418.
15) Kaiser PK, Lowder CY, Sullivan P, et al. Chest computerized tomography in the evaluation of uveitis in elderly women. Am J Ophthalmol 2002, 133: 499-505.
16) Kawaguchi T, Hanada A, Horie S, et al. Evaluation of characteristic ocular signs and systemic investigations in ocular sarcoidosis patients. Jpn J Ophthalmol 2007; 51: 121-6.
17) Rothova A. Ocular involvement in sarcoidosis. Br J Ophthalmol 2000; 84: 110-6.
18) Ohara K. Intraocular manifestations of systemic sarcoidosis. Jpn J Ophthalmol 1992; 36: 452-7.
19) Abad S, Meyssonier V, Allali J, et al. Association of peripheral multifocal choroiditis with sarcoidosis: a study of thirty-seven patients. Arthritis Rheum 2004; 51: 974-82.
20) Tabbara KF. Tuberculosis. Curr Opin Ophthalmol. 2007;18: 493-501.

CHAPTER 9

Chronic central serous chorioretinopathy associated with serous retinal detachment in a series of Asian patients

Kunavisarut P, Pathanapitoon K, Schooneveld MV, Rothova A.

Ocular Immunology and Inflammation 2009; 17: 269–77.

ABSTRACT

Purpose: To determine clinical features of patients with severe chronic central serous chorioretinopathy (diffuse retinal pigment epitheliopathy, DRPE) associated with bullous retinal detachment in Thailand.

Methods: The authors reviewed clinical and imaging characteristics, visual outcomes, and complications of 7 patients with severe DRPE associated with bullous retinal detachment.

Results: Included were 6 males and 1 female with average age at onset of 39 years (range 30–46 years) diagnosed with DRPE. Although 4 patients had unilateral complaints, retinal pigment epithelium (RPE) changes on fluorescein angiography (FA) were visible in both eyes in all patients and 10 out of 14 affected eyes exhibited large exudative bullous retinal detachments (RD) and evidence of multiple characteristic leakage points. The disease was induced by steroid medications in 3 patients and an additional 3 patients received steroid treatment after they were initially considered to have Harada disease. The administration of steroids caused worsening in all cases.

Conclusion: Chronic central serous chorioretinopathy associated with bullous retinal detachment is a severe variant of DRPE, which might be mistaken for Harada disease. The early diagnosis of DRPE might prevent the complications from harmful medications as well as unnecessary surgery and visual loss.

INTRODUCTION

Chronic central serous chorioretinopathy (CSC) represents an idiopathic disorder also called diffuse retinal pigment epitheliopathy (DRPE), and is characterized by multiple subretinal foci of leakage leading to serous retinal detachments, which are associated with widespread alterations in retinal pigment epithelium (RPE). Frequently, multiple serous detachments of RPE are also visible on fluorescein angiography (FA).[1] The course is chronic and recurrences of subretinal leakage are common. Data obtained from indocyanine green angiographies suggest that the primary abnormalities lie in the choriocapillaris and the involvement of the RPE might be secondary.[2] DRPE is a rare disorder, which can develop during any type of steroid administration [3–5] or during pregnancy, [6–9] but may also occur spontaneously.[10] DRPE associated with exudative bullous retinal detachment (RD) occurs predominantly in male adult patients and seems to occur more frequently in Asia, but whether this severe DRPE variant really has ethnic predilection remains to be elucidated.[10–14]

In this article we report on the clinical features and visual prognosis of 7 Thai patients with DRPE and bullous RD and point out the frequent development of subretinal fibrosis and similarity of presenting features with Harada disease.

METHODS

We reviewed the medical records of 7 patients with severe DRPE associated with bullous RD who had consulted the ophthalmologic department of the university hospital in Chiang-Mai during 2005–2007. All patients had to fulfill all of the following criteria: multiple leakage points at RPE level on FA, serous detachments in the absence of inflammatory signs, exudative RD with positive shifting fluid, no retinal breaks, and the absence of leakage of optic disc and/ or retinal vessels on FA. Each patient underwent a full ophthalmic examination, including slit-lamp biomicroscopy, tonometry, indirect ophthalmoscopy, FA, and optical coherence tomography (OCT) examinations. Clinical features, visual acuity changes over time, and eventual complications were registered. Mean follow-up was 11 months (range, 6–24 months).

RESULTS

The clinical characteristics of 7 patients with a final diagnosis of CSC with exudative bullous RD are given in Tables 1 and 2. There were 6 males and 1 female, with an average age at onset of 39 years (range, 30–46 years). A previous history of classical CSC was present in 3/7 cases (43%). Although 4 patients had unilateral complaints only, RPE changes on FA were

visible in both eyes in all patients and 10 out of 14 affected eyes exhibited large exudative bullous detachments (7 patients; Figure 1). In all cases retinal detachments were located in the inferior quadrants and were characterized by the accumulation of subretinal fluid shifting with changing body position. In addition, all cases exhibited white-yellow subretinal exudates in the posterior pole (Figures 2 and 3). FA showed on the early frames multiple characteristic leakage pinpoints with increased leakage with time (Figure 1B, C; Figure 2B–D). OCT through the exudative lesions demonstrated extensive detachments of thickened retina and cloudy subretinal fluid (Figure 2E). All patients had multiple serous detachments and 5 out of 7 had associated pigment epithelial detachment (PED; Figure 1D). Four out of 7 patients developed their ocular disease spontaneously; the remaining 3 cases developed their ocular disease during steroid administration for their underlying conditions (Table 1). The ocular signs and symptoms did not differ between the idiopathic cases and the steroid induced cases. Apart from the 3 steroid-induced cases, 3 idiopathic cases received steroid medication since they were incorrectly considered to have Harada disease. The administration of steroids caused worsening of the ocular pathologic features in all (Table 2).

After the diagnosis of DRPE was made, steroid medication was withdrawn in all, except 1 case (case 7) for whom the steroid medication had to be gradually tapered from 50 to 10 mg/day and azathioprine 50 mg/day was added to control myasthenia gravis. After the withdrawal of steroids, a gradual regression of the exudative RD occurred and on average in 5 months (range, 1–9 months) the retina was reattached (confirmed by OCT in all). The time needed to reattachment was longer in all 6 patients who received steroid medication than in the remaining patient who had never received steroids. Apart from the withdrawal of the steroids, additional treatment was required in 1 patient (case 3) due to the prolonged persistence of subretinal fluid and marked leakage in the papillomacular area. He was treated with a "half-dose" of PDT (photodynamic therapy; spot size 8000 μm, GLD 7000 μm, verteporfin (Visudyne) 2.6 mL, light burn 50 J/cm^2, intensity 600 nW/cm^3, 83 s), which resulted in a complete resolution of subretinal fluid in 3 months. Complications included predominantly extensive subretinal fibrosis, which was noted in 7/14 affected eyes (7/10 eyes with exudative RD; Figure 4). Sub-and juxtafoveal scarring occurred in 4/14 affected eyes (4/10 eyes with exudative RD). So far, no subretinal neovascularization has developed. Visual acuity of 0.1 or less at the 6-month follow-up was observed in 3/14 eyes (3/10 eyes with RD, 3 patients). The best-corrected visual acuity at the 6-month follow-up of 0.5 or better was found in 8/14 eyes (4/10 eyes with RD). During the follow-up, none of the patients developed systemic symptoms of Vogt-Koyanagi-Harada disease (VKH).

Table 1. Clinical characteristics of 7 patients with severe diffuse retinal pigment epitheliopathy (also termed chronic multiple central serous chorioretinopathy) associated with bullous retinal detachment

Case	Gender	Age at first presentation	Eye(s) complaints	Involvement on FA	Associated systemic disease	Initial clinical diagnoses	Steroid medication before the onset of DRPE
1	M	31	unilateral	bilateral	None	RRD,TB,VKH	None
2	M	30	bilateral	bilateral	None	VKH	None
3	M	42	bilateral	bilateral	Diabetes mellitus Systemic hypertension Hyperlipidemia	RRD, VKH	None
4	M	43	unilateral	bilateral	Psoriasis	RRD	Systemic (20mg/day) and local administration
5	M	46	bilateral	bilateral	None	VKH	None
6	M	40	unilateral	bilateral	Allergic rhinitis	VKH, DRPE	Local administration
7	F	42	unilateral	bilateral	Myasthenia gravis Systemic hypertension	VKH, DRPE	Systemic, 50mg/day

RRD rhegmatogenous retinal detachment, TB tuberculosis ,VKH Vogt-Koyanagi-Harada, FA fluorescein angiography, DRPE diffuse retinal pigment epitheliopathy

Table 2. Ocular features of 7 patients with severe diffuse retinal pigment epitheliopathy (also termed chronic multiple central serous chorioretinopathy) associated with bullous retinal detachment

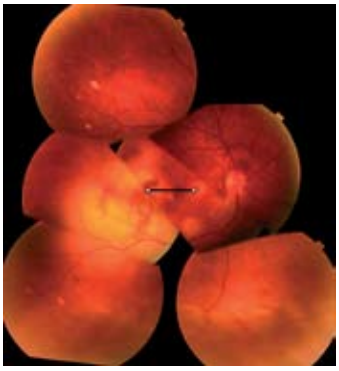
Case	Steroid medication during the course of DRPE	VA onset	VA changes following steroid medication	Retina changes following steroid medication	Total duration of RD (months)	Duration of resolution of SRF (months)	VA at 3 months follow-up	VA at 6 months follow-up	Complications at 6 months follow-up
1	None	RE 20/80	n.a.	n.a.	2	2	RE 20/40	RE 20/30	Subretinal fibrosis
		LE 20/30	n.a.		n.a.	n.a.	LE 20/30	LE 20/30	None
2	Systemic and intraocular	RE 20/80	RE FC	Increase of RD	9	7	RE FC	RE 20/80	Subretinal fibrosis
		LE 20/80	LE FC	Increase of RD	9	7	LE FC	LE FC	Subretinal fibrosis
3	Systemic and intraocular	RE 20/20	RE FC	Increase of RD	10	10	RE 20/200	RE 20/200	Subretinal fibrosis
		LE 20/200	LE FC	Increase of RD	10	8	LE 20/200	LE 20/40	Subretinal fibrosis
4	Systemic and local	RE 20/80 LE 20/20	Initiation of the disease (see VA at onset)	DRPE development	6.5 n.a.	6 n.a.	RE 20/200 LE 20/20	RE 20/40 LE 20/20	None None
5	Systemic	Not known	RE 20/200	Increase of RD	4	2	RE 20/200	RE 20/80	Subretinal fibrosis
		Not known	LE 20/40	Increase of RD	3	1	LE 20/20	LE 20/20	None
6	Local	RE FC LE 20/20	Initiation of the disease (see VA at onset)	DRPE development	8 n.a.	3 n.a.	RE FC LE 20/20	RE 20/200 LE 20/20	None None
7	Systemic	RE 20/20 LE 20/200	Initiation of the disease (see VA at onset)	DRPE development	n.a. 10	n.a. 9	RE 20/20 LE 20/200	RE 20/20 LE 20/100	None Subretinal fibrosis

RD retinal detachment, RE right eye, LE left eye, FC finger counting, VA visual acuity, DRPE diffuse retinal pigment epitheliopathy, n.a. not applicable, SRF subretinal fluid

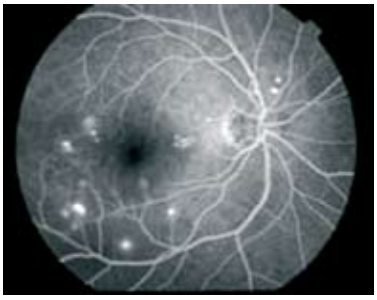
Table 3. Differential diagnosis of severe diffuse retinal pigment epitheliopathy (also termed chronic multiple central serous chorioretinopathy) associated with bullous retinal detachment and Vogt-Koyanagi-Harada disease and rhegmatogenous retinal detachment

	DRPE with RD	VKH	RRD
Racial predisposition	No racial predilection, might be more frequent in Asian	Asians, Black, Hispanics	None
Sex predilection	Predominantly males	No	No
Systemic history and associations	History of steroid usage	Malaise, nausea Neurologic signs (headaches, pleocytosis of cerebrospinal fluids) Dermatologic signs (alopecia, poliosis, vitiligo) Auditory signs (dysacusis, tinnitus)	absent
Ocular history and associations	Previous CSR possible	Usually none	Sometimes myopia, trauma, intraocular surgery
Typical ocular complaints	Metamorphopsia and decrease of visual acuity	Photophobia and slow decrease of visual acuity	Photopsia, floaters, visual field defect and/or decrease of visual acuity
Ocular findings	Multiple serous RD Shifting subretinal fluid No intraocular inflammation No retinal breaks	Serous RD Intraocular inflammation Optic disc hyperemia Sunset-glow fundus Peripheral retinal lesions	Retinal holes, tobacco dust No shifting fluid
FA angiography	Multiple serous RD with smoke stacks and ink-blot leaks, frequently PED	Many irregular leakage points (pin-point), Multiple serous RD Disc leakage, seldom PED	No RPE leakage, no PED
Role of steroid medication	Contraproductive, may aggravate the disease	Effective, may shorten the duration of RD	Ineffective

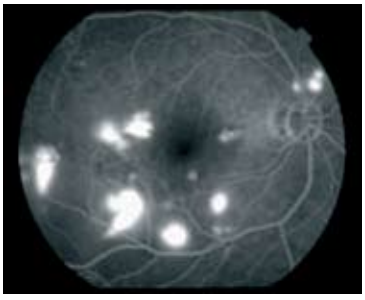
DRPE diffuse retinal pigment epitheliopathy, RD retinal detachment, VKH Vogt-Koyanagi-Harada disease, RRD rhegmatogenous retinal detachment, CSR central serous chorioretinopathy, PED pigment epithelial detachment, RPE retinal pigment epithelium



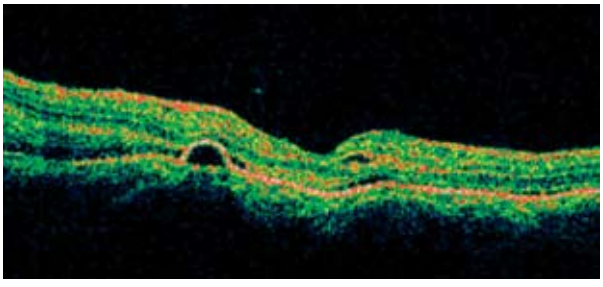
(a)



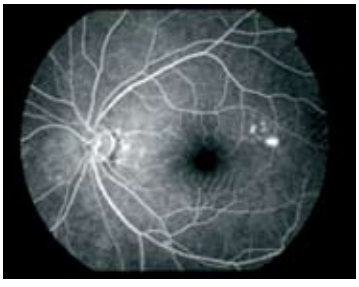
(b)



(c)

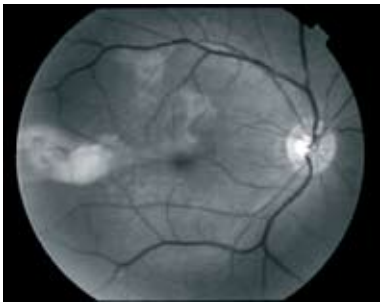


(d)

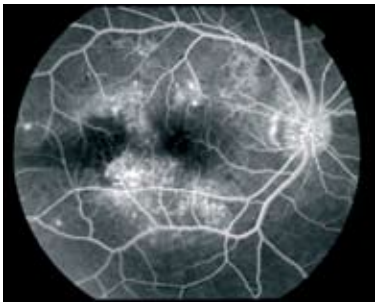


(e)

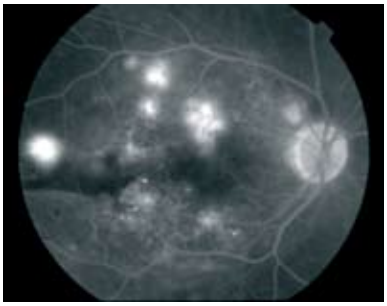
Figure 1. *Case 4:* (A) Fundus photograph of the right eye at the time of initial presentation with diffuse retinal pigment epitheliopathy (DRPE) and retinal detachment, illustrating multifocal exudative lesions in the posterior pole and bullous retinal detachment in the inferior quadrants. (B, C) Fluorescein angiogram, early (B) and late (C) frames of the same eye showing multiple leaking pinpoints, with increased leakage over time. Note also a smoke stack and absence of optic nerve head leakage in (C). (D) Cross-sectional scan through the fovea by optical coherence tomography (right eye), illustrating serous as well as pigment epithelial detachments. (E) Fluorescein angiogram of the left eye showing retinal pigment epithelium aberrations despite the absence of fundoscopic abnormalities and subjective complaints.



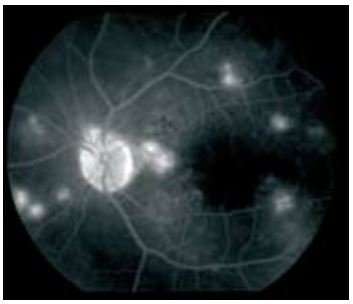
(a)



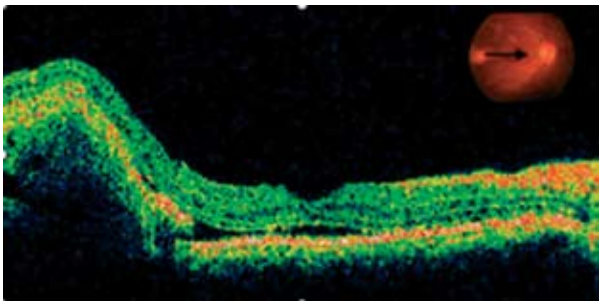
(b)



(c)

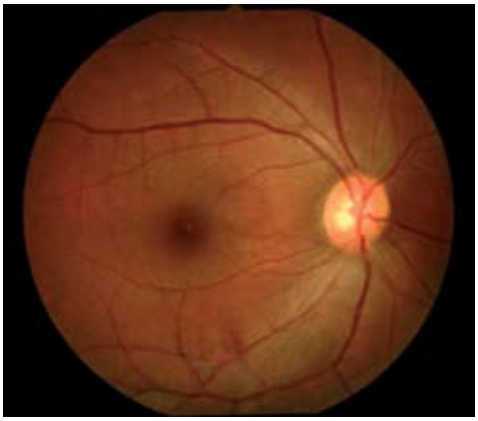


(d)

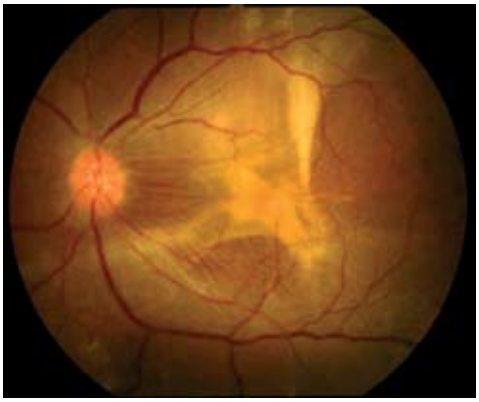


(e)

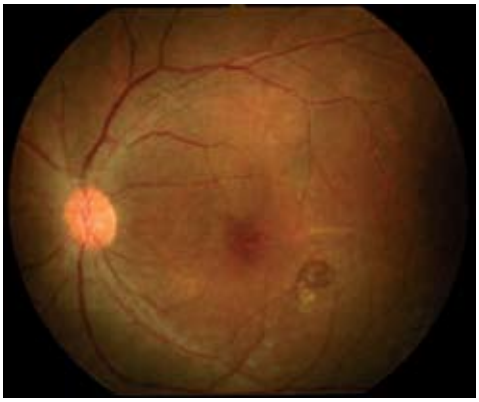
Figure 2. *Case 5:* (A) Red-free fundus photograph of the right eye taken after 1 month of administration of 30 mg of oral prednisolone given as a consequence of initial incorrect diagnosis of Harada disease. Note the subretinal exudates located temporally to the macular area. (B, C) Fluorescein angiogram early (B) and late (C) frames of the same eye showing multiple leakage pinpoints with increased leakage over time. (D) Fluorescein angiogram of the left eye demonstrating multiple areas of leakage at RPE level. (E) Cross-sectional optical coherence tomography scan (right eye) through the exudative lesion, illustrating detachment of thickened retina with cloudy subretinal space.



(a)

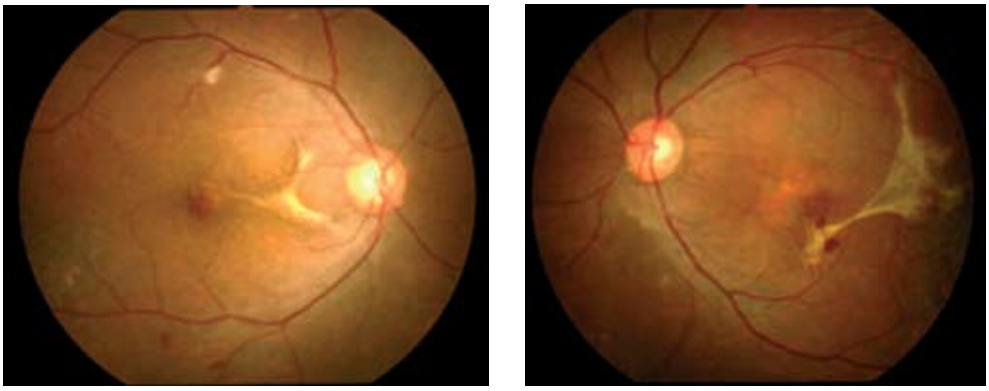


(b)



(c)

Figure 3. *Case 7:* Fundus photograph at the time of the initial presentation with diffuse retinal pigment epitheliopathy, which took place 5 months after the administration of oral prednisolone (50 mg/day) was initiated. (A) Right eye shows local extramacular serous detachments. (B) Left eye shows extensive white-yellow subretinal exudates and the beginning of the fibrosis with traction at the posterior pole. (C) Fundus photograph of the left eye 9 months after the lowering of the prednisone to 10 mg per day demonstrates subretinal fibrotic scar located in the extramacular area and also documents attenuation of the retinal arteries possibly due to longstanding retinal detachment.



(a) (b)

Figure 4. *Case 3:* Fundus photograph taken 1 year after disease onset, illustrating subretinal fibrosis, which caused metamorphopsia and decreased visual acuity in both eyes.

DISCUSSION

We report on 7 Asian patients with the severe form of DRPE associated with exudative bullous RD and complicated by subretinal fibrosis in 70% of eyes with RD. The majority of patients were primarily considered to have Harada disease leading to an initially incorrect treatment in 3 patients. The absence of inflammatory signs and nonleaky disc on FA were helpful in making the correct diagnosis as well as the worsening of the disease with steroids. DRPE (or chronic CSC) associated with bullous retinal detachment is a severe variant of DRPE, which is probably frequently misdiagnosed. This disorder may mimic more common ocular disorders such as rhegmatogenous retinal detachment or all sorts of exudative detachment, including Vogt-Koyanagi-Harada disease (VKH), uveal effusion, posterior scleritis, and other disorders. It is aggravated by the use of steroids.[3–5] Probably, many ophthalmologists would not even consider the possibility of DRPE initially in patients with extensive serous retinal detachment. In contrast, VKH disease was included in the differential diagnosis in all of our patients and three had received high steroid dosages, which aggravated their ocular symptoms; one patient was already scheduled for reattachment surgery because of suspected rhegmatogenous RD. Since most publications on this severe DRPE type come from Asia, it might be feasible that, similarly to VKH, this severe form of DRPE with retinal exudative detachment might be more prevalent in Asia.[10–14] Steroids form a frequently described cause of CSC regardless of their route of administration (systemic as well as various local applications were associated with CSC),[15–17] and can also prolong the duration of the disease for many months, which may result in definitive loss of visual acuity and a substantial amount of subretinal fibrosis. In contrast, cases that

developed without steroids were usually unilateral and tended to resolve faster with good visual outcome.[10] This phenomenon was also present in our patients. The role of steroids in the development of DRPE is not yet clear. Also patients with elevated endogenous levels of steroids may develop CSC and/or DRPE.[17] It was speculated that increased plasma cortisol levels may be responsible for pathologic permeability of the choriocapillaris. Cortisol also increases capillary fragility, resulting in fibrinous exudation in the subretinal space. The addition of exogenous steroids might aggravate the leakage as well as inhibit the eye's ability to repair the capillary damage.[3] Withdrawal of the steroids will usually be sufficient to treat the majority of steroid-associated cases[18,19]; this is also shown in our series. Unfortunately, some of these patients may be steroid-dependent and need ongoing administration (e.g., post-transplant patients); in these patients steroid-sparing medications should be explored and evaluated. In patients with threatened visual acuity, symptomatic treatment with laser photocoagulation[13] and/or PDT might be an option.[20–22]

The development of subretinal exudates is characteristic of this disorder [7,13,23] and is followed by gradual transformation from subretinal exudation to subretinal fibrosis. It can be present during initial presentation [13] or develop later in the course of the disease.[23,24] All of our patients exhibited these exudates. It was observed that retinal detachment persistent for longer than 4 months might be associated with a definitive reduction of visual acuity despite resolution of the serous detachment because of the atrophy of photoreceptors and retinal pigment epithelial cells.[25] Therefore, the timely diagnosis and treatment of DRPE with exudative RD are imperative. Combination of ophthalmoscopic, FA, and OCT findings (aided by ICG angiography) may be helpful (Table 3).

We recommend including DRPE in the differential diagnosis of all patients with exudative RD of undetermined cause. Patients with possible DRPE should be evaluated for potential steroid use and preferably undergo FA. In Asian patients, the distinction from VKH is important (Table 3). The presence of intraocular inflammation and typical extraocular signs are required for the diagnosis of VKH while in DRPE with exudative RD, the absence of inflammation and of systemic signs are typical as well as the frequent association with some kind of steroid administration. The distinction of these two disorders on angiography alone is quite difficult as the resemblance is great, and inflammatory signs in VKH are not always apparent. The major help in distinguishing FA includes the absence of inflammatory signs as well as the frequent presence of associated retinal pigment epithelial detachments in DRPE.

In summary, we report on a severe variant of DRPE associated with exudative RD, the correct diagnosis of which might prevent the complications from harmful medications as well as unnecessary surgery and visual loss.

REFERENCES

1. Cohen D, Gaudric A, Coscas G, Quentel G, Binaghi M. Diffuse retinal epitheliopathy and central serous chorioretinopathy. J Fr Ophtalmol. 1983;6:339–349.
2. Uyama M, Matsunaga H, MatsubaraT, Fukushima I, Takahashi K, NishimuraT. Indocyanine green angiography and pathophysiology of multifocal posterior pigment epitheliopathy. Retina. 1999;10: 12–21.
3. Gass JD, Little H. Bilateral bullous exudative retinal detachment complicating idiopathic central serous chorioretinopathy during systemic corticosteroid therapy. Ophthalmology. 1995;102:737–737.
4. Gass JD. Bullous retinal detachment: an unusual manifestation of idiopathic central serous choroidopathy. Am J Ophthalmol. 1973;75:810–812.
5. Mazzuca DE, BensonWE. Central serous retinopathy: variants. Surv Ophthalmol. 1986;31:170–174.
6. Quillen DA, Gass DM, Brod RD, et al. Central serous chorioretinopathy in women. Ophthalmology. 1996;103:72–79.
7. Gass JD. Central serous chorioretinopathy and white subretinal exudation during pregnancy. Arch Ophthalmol. 1991;109:677–681.
8. Fastenberg DM, Ober RR. Central serous choroidopathy in pregnancy. Arch Ophthalmol. 1983;101: 1055–1058.
9. Chumbley LC, Frank RN. Central serous retinopathy and pregnancy. Am J Ophthalmol. 1974;77: 158–160.
10. Sahu DK, Namperumalsamy P, Hilton GF, de Sousa NF. Bullous variant of idiopathic central serous chorioretinopathy. Br J Ophthalmol. 2000;84:485–492.
11. Otsuka S, Ohba N, Nakao K. A long-term follow-up study of severe variant of central serous chorioretinopathy. Retina. 2002;22:25–32.
12. Akiyama K, KawamuraM, Ogata T, Tanaka E. Retinal vascular loss in idiopathic central serous chorioretinopathy with bullous retinal detachment. Ophthalmology. 1987;94:1605–1609.
13. Sharma T, Badrinath SS, Gopal L, et al. Subretinal fibrosis and nonrhegmatogenous retinal detachment associated with multifocal central serous chorioretinopathy. Retina. 1998;18:23–29.
14. Fawzi AA, Holland GN, Kreiger AE, Heckenlively JR, Arroyo JG, Cunningham ET. Central serous chorioretinopathy after solid organ transplantation. Ophthalmology. 2006;113:805–813.
15. Wakamura M, Ishikawa S. Central serous chorioretinopathy complicating systemic corticosteroid treatment. Br J Ophthalmol. 1984;68:329–331.
16. Haimovici R, Graqoudas ES, Duker JS, Sjaarda RN, Eliott D. Central serous chorioretinopathy associated with inhaled or intranasal corticosteroids. Ophthalmology. 1997;104:1653–1660.
17. Bouzas EA, Karadimas P, Pournaras CJ. Central serous chorioretinopathy and glucocorticoids. Surv Ophthalmol. 2002;47:431–448.
18. Loo JL, Lee SY, Ang CL. Can long-term corticosteroids lead to blindness? A case series of central serous chorioretinopathy induced by corticosteroids. Ann Acad Med Singapore. 2006;35:496–499.

19. Sharma T, Shah N, Rao M, et al. Visual outcome after discontinuation of corticosteroids in atypical severe central serous chorioretinopathy. Ophthalmology. 2004;111:1708–1714.
20. Yannuzzi LA, Slakter JS, Gross NE, Psaide RF, et al. Indocyanine green angiography-guided photodynamic therapy for treatment of chronic central serous chorioretinopathy. Retina. 2003;23:288–298.
21. Piccolino FC, Eandi CM, Ventre L, et al. Photodynamic therapy for chronic central serous chorioretinopathy. Retina. 2003;23:752–763.
22. Lai TYY, Chan WM, Li H, Lai RYK, Liu DTL, Lam DSC. Safety enhanced photodynamic therapy with half dose verteporfin for chronic central serous chorioretinopathy: a short term pilot study. Br J Ophthalmol. 2006;90:869–874.
23. Schatz H, McDonald HR, Johnson RN, et al. Subretinal fibrosis in central serous chorioretinopathy. Ophthalmology. 1995;102:1077–1088.
24. Hooymans JM. Fibrotic scar formation in central serous chorioretinopathy developed during systemic treatment with corticosteroids. Graefes Arch Clin Exp Ophthalmol. 1998;236:876–879.
25. Wang MS, Sander B, Larsen M. Retinal atrophy in idiopathic central serous chorioretinopathy. Am J Ophthalmol. 2002;133: 787–793.

CHAPTER 10

Summary and Conclusions

In this thesis we identify the causes of blindness in a tertiary center in Thailand and point out the importance of uveitis as a cause of visual impairment. We report on a spectrum of uveitis entities observed in the Thai population, perform the analysis of intraocular fluid samples by polymerase chain reaction and describe the clinical manifestations of diverse infectious uveitis entities. We further report on the dynamics of the human immunodeficiency virus (HIV) across the blood-retina barrier and describe the novel uveitic entity of HIV-induced uveitis. In addition we report on HLA B27- associated anterior uveitis and ocular sarcoidosis in the Thai population and describe the masquerade syndrome of chronic central serous chorioretinopathy associated with serous retinal detachment, which might be easily confused with Vogt-Koyanagi-Harada disease.

CHAPTER 1

Here we introduce the background information and summarize the literature on uveitis in Southeast Asia with special emphasis on Thailand. Further, we raise specific research questions and describe the plans for individual studies included in this thesis and identify its main purpose.

CHAPTER 2

In this chapter we assess the underlying causes of blindness and low vision in 2,951 consecutive new patients referred to our university hospital in northern Thailand. We investigate the anatomical location and specific causes of blindness and low vision in these patients and provide the information on location, etiology and preventability of blindness-inducing disorders.

We found that out of 2951 patients, 150 had bilateral blindness and/or low vision and 219 unilateral blindness and/or low vision, amounting to a total of 369 (12.5%) patients with visually impaired eye(s). The most frequent causes of visual loss consisted of age-related ocular diseases (35%; 128/369) followed by infections (18%; 66/369) and trauma (12%; 43/369/). Cytomegalovirus retinitis in HIV-infected patients was the cause of bilateral blindness in 19% (14/73) of all patients. Blindness and low vision were considered avoidable in 70% of cases.

We concluded that in 2005, age-related ocular disorders and infections were the most common causes of visual impairment encountered in a tertiary center in northern Thailand. We point out the leading role of CMV retinitis in HIV–infected patients as a cause of blindness in that period of time given that there was not (yet) freely available access to HAART.

CHAPTER 3

In this part, we assessed the anatomical types and causes of uveitis in Thailand by undertaking a prospective study of 200 new consecutive patients with uveitis and determined their specific diagnoses on the basis of clinical manifestations and the results of screening, which included erythrocyte sedimentation rate, red and white blood cell counts, serological tests for HIV, syphilis and toxoplasmosis, urine analysis, and radiological chest examination in all types of uveitis and additional HLA B27 determination in patients with anterior uveitis .

We established that HIV was positive in 31% (62/200) of all those with uveitis and that posterior uveitis was the most frequent anatomical location in the HIV-positive population; CMV was the most common cause of their uveitis (85%; 53/62). In the HIV-negative group, the most common anatomical type was anterior uveitis (34%; 47/138). Infectious uveitis was diagnosed in 22% (30/138) of HIV-negative patients, and in this study, based on clinical presentation and serology, toxoplasmosis was the most common infection (8.7%; 12/138). Association with systemic disease was noted in 26% (36/138) and established clinical entities were diagnosed in 33% (46/138). The undetermined origin of uveitis was observed in 19% (26/138). The most common non-infectious clinical entities were Vogt–Koyanagi–Harada disease (18.5%; 22/108) and HLA-B27-associated acute anterior uveitis (9%; 10/108).

We concluded that the spectrum of uveitis in northern Thailand encompassed 31% of HIV-positive patients. Out of all 200 patients with uveitis, 27% (53/200) was caused by cytomegalovirus retinitis in HIV-infected patients. Causes of HIV-negative uveitis were distinct from those found in the West, but similar to those observed in the Far East. However, specific prevalences of these uveitis entities were distinct from those found in India and Japan.

CHAPTER 4

To determine which infectious agents might play a role in the etiology of infectious uveitis in Thailand, we investigated the seroprevalence of various infections (known to cause uveitis in other parts of the world) in 101 consecutive Thai patients with HIV-negative uveitis and in 100 HIV-infected patients with retinitis and compared the results with 100 non-uveitis controls.

Positive antibodies against *T. gondii* in were found in 31% in HIV-negative patients, mostly in those with posterior uveitis and focal retinitis. The prevalence of positive toxoplasma serology was higher in those with posterior uveitis than in the other anatomical entities examined. Antibodies against *T. pallidum* and Leptospira were observed more frequently in HIV-positive patients with retinitis though the role of these pathogens in inciting uveitis could not be established. The higher seroprevalence for syphilis in HIV-positive patients implies that

the possibility of concurrent syphilis should be considered in all HIV-positive patients with intraocular inflammation.

We concluded that *T. gondii* might be a cause of posterior uveitis in HIV-negative patients. Further investigations into the concurrent presence of syphilis and syphilitic uveitis in Thai HIV-infected patients are needed.

CHAPTER 5

In this chapter we evaluate the results of PCR analysis for HSV, VZV, CMV and *T. gondii* in intraocular fluid samples of 100 HIV-negative patients and in 47 HIV-positive patients with uveitis of unknown origin assessed in our center between May 2006 and October 2009 and determine the usefulness of PCR for the diagnosis of uveitis in Thailand.

Positive PCR results in intraocular fluids were found in 45% (66/147) of all patients with uveitis. The positive PCR results were lower in the HIV-negative patients (33%; 33/100) than in HIV-positive patients (70%; 33/47). CMV was the most common infectious agent found in both HIV-negative and HIV-positive patients with uveitis (49%; 16/33 and 91%; 30/33, respectively). In HIV-negative patients, CMV was predominantly associated with anterior uveitis but in HIV-positive patients CMV-associated ocular disease manifested itself mostly as posterior and panuveitis. CMV-positive anterior uveitis in HIV-negative patients was generally unilateral and was associated with a high intraocular pressure and absence of posterior synechiae. CMV retinitis was also diagnosed in 3 HIV-negative immmunocompetent patients. Infections with *T.gondii* had a high prevalence of atypical features; specifically 3 patients with anterior uveitis and positive PCR results for T.gondii were found and their clinical manifestations described.

We concluded that PCR analysis of intraocular fluid samples was a highly valuable diagnostic assay in patients with uveitis, which revealed that CMV was the most common cause of infectious uveitis in Thailand, in both, HIV-negative and HIV-positive patients. Prevalence of specific infections and their clinical manifestations were different from those observed in the West.

CHAPTER 6

In this part of the thesis, we determined intraocular and plasma HIV-1 RNA loads in 40 HIV-positive patients with uveitis and studied the dynamics of HIV across the blood-retinal barrier. In addition we described clinical manifestations of patients with detectable intraocular HIV load, with the emphasis on patients with intraocular HIV loads exceeding the HIV loads of plasma.

Intraocular HIV-1 RNA was detected in 33% (13/40) of HIV-positive patients with uveitis. Intraocular HIV-1 RNA loads were associated with high HIV-1 RNA plasma loads and not being on HAART therapy. In addition, detectable intraocular HIV-1 RNA levels were associated with inflammatory cells present in the eye and with the absence of retinal lesions. Furthermore, in 4 patients, the levels of intraocular HIV-1 RNA exceeded the concurrent HIV-1 RNA plasma loads. Three of these 4 patients had anterior uveitis and/or vitritis without retinal lesions and had no laboratory evidence of intraocular opportunistic infections.

We concluded that positive intraocular HIV loads are detectable in a considerable percentage of HIV-positive patients with uveitis, especially in those with high HIV plasma loads, and those not receiving antiretroviral therapy. Anterior uveitis, without associated retinal lesions in HIV-positive patients with high plasma HIV1-RNA loads, is probably caused by HIV itself.

CHAPTER 7

To identify whether HLA B27-associated AAU is a uveitis entity also prevalent in the Thai population, we studied the frequency of HLA-B27 in healthy Thai individuals without AAU (n=100) and in patients with acute anterior uveitis (n=121). In addition, we described the clinical features of HLA B27-positive AAU in this particular population.

The prevalence of HLA-B27 was 10% among the blood donors in contrast to 44% in the AAU group. The clinical characteristics of HLA-B27-associated AAU were similar to those published elsewhere in the world. At least 15% of the HLA B27-positive group had radiological signs of ankylosing spondylitis. The prevalence of HLA B 27-associated systemic diseases was markedly lower than reported in other parts of the world and might be explained by the presence of the specific HLA-B27 subtypes, prevalent infections in the population and by the underdiagnosis of our patients (not all of our patients underwent a detailed rheumatologic evaluation).

We concluded that HLA B27-associated AAU is present in the Thai population and has similar clinical characteristics to the cases reported in the West. The causes of the different pattern of associations between HLA B27-positive AAU and systemic disease noted in our study should be further studied.

CHAPTER 8

In this chapter, we investigate whether ocular sarcoidosis is current in Thailand by searching for the signs of pulmonary sarcoidosis in 209 new consecutive patients with uveitis, and subsequently describe ocular features of Thai patients with uveitis and pulmonary sarcoidosis.

We concluded that at least 4 out of 209 (2%) uveitis patients suffered from pulmonary sarcoidosis. From 209 chest x-ray (CXR) examinations, one patient (0.5 %) exhibited radiological signs typical of stage 1 sarcoidosis. Chest CT of 3 patients with posterior multifocal chorioretinitis (PMC) revealed abnormalities (bilateral hilar lymphadenopathy) suggesting the diagnosis of pulmonary sarcoidosis. Tissue biopsy was performed in 2 patients with PMC and confirmed the diagnosis of sarcoidosis in both. All three PMC patients were females older than 50 years; they had no pulmonary complaints and their CXRs were without abnormalities. All PMC patients developed cystoid macular edema with epiretinal membranes.

We deduced that ocular sarcoidosis is more common in Thailand than previously thought. We suggest that more patients with sarcoidosis would be identified if diagnosis was thought of and ancillary tests were systematically performed.

CHAPTER 9

In this section, we report on a masquerade syndrome mimicking the VKH disease in 7 patients suffering from chronic central serous chorioretinopathy (CSR or diffuse retinal pigment epitheliopathy, DRPE) associated with bullous retinal detachment and report on their clinical presentations, response to treatment and signs which differentiate patients with severe CSR from those with VKH.

Seven patients presented with serous retinal detachment and were initially suspected to have VKH disease. Four patients had unilateral complaints only, but on fluorescein angiography retinal pigment epithelium changes were visible in both eyes in all patients. In addition, large exudative bullous retinal detachments were encountered accompanied by multiple characteristic leakage points. However, none of the patients exhibited leakage of the optic disc on the late frames of angiography. In addition, on ophthalmologic examinations inflammatory cells were not encountered. Disease was induced by steroid medications in 3 patients and an additional 3 patients received steroid treatment after they were initially considered to have VKH disease. The administration of steroids caused worsening in all patients.

We concluded that chronic central serous chorioretinopathy associated with bullous retinal detachment is a severe variant of DRPE, which might be easily mistaken for VKH disease. The major differentiating signs consist of optic disc leakage and inflammatory reaction as well as the systemic signs, which are regularly present in VKH disease but absent in DRPE. The early diagnosis of DRPE might prevent the adverse complications from medications as well as unnecessary surgery and visual loss.

MAIN CONCLUSIONS

In this thesis, we report on the so far unknown causes of uveitis in Thailand and describe their clinical manifestations.

We found out that intraocular infections were the second most common cause of blindness and low vision in a tertiary center in northern Thailand. These infections predominantly consisted of CMV retinitis in HIV–infected patients. The spectrum of uveitis in northern Thailand included approximately 30% of HIV-infected patients with cytomegalovirus retinitis. Causes of HIV-negative uveitis seemed to be similar to those observed elsewhere in the Far East. However, PCR analysis of intraocular fluid revealed that CMV was the most frequent cause of infectious uveitis not only in HIV-positive patients but surprisingly also in HIV-negative patients. While CMV retinitis was most prevalent in HIV-positive patients, CMV-positive hypertensive anterior uveitis was typically present in HIV-negative patients. PCR analysis of intraocular fluids appeared to form a valuable diagnostic procedure as 45% of those examined exhibited positive results. We found out that positive intraocular HIV 1-RNA loads in HIV-positive patients with uveitis were predominantly present in the eyes of untreated patients with high plasma HIV loads. Further, we describe a novel infectious uveitis entity, namely HIV-induced anterior uveitis and report on its characteristics. Additionally we describe the clinical entities of HLA B27-associated AAU and ocular sarcoidosis in the Thai population and point out severe DRPE as an important masquerade of VKH disease.

Principally, this thesis increases our knowledge in the field of uveitis in Southeast Asia and gives an account of the main uveitis entities present in this population. Based on the results reported in this thesis, future research should address the diagnostic role of Goldmann Witmer coefficient in patients with uveitis, which might assist the diagnosis of chronic ocular infections. Further exploration of clinical manifestations of ocular toxoplasmosis is desirable as the correct diagnosis makes the focused treatment possible. The presence of ocular sarcoidosis in Thailand could be clarified by a systemic study of radiologic and CT chest examinations in patients with uveitis and their diagnostic value in patients with uveitis could further be determined. The systematic taking of tissue biopsy and QuantiFeron tests could be further applied to determine the exact role of tuberculosis and sarcoidosis in Thai patients with uveitis. The study of human ocular tissues (using iris or retinal biopsies) might help to identify the exact cell population where HIV can replicate. Further analyses of intraocular HIV could determine whether the virus situated within the eye is distinct from the virus population present in the blood. Last, but not least, it would be valuable to expand the PCR facilities analyses and make the PCR analysis of intraocular fluids accessible to patients and ophthalmologists from the areas outside our hospital.

CHAPTER 11

Samenvatting

SAMENVATTING IN HET NEDERLANDS

Dit proefschrift heeft als doel de etiologie, diagnostiek en klinische manifestaties van uveitis in Thailand in kaart te brengen. In het eerste deel onderzoeken wij de oorzaken van blindheid en slechtziendheid op de afdeling Oogheelkunde van een universitair ziekenhuis in Chiang Mai, Thailand, en benadrukken de belangrijke rol die uveitis speelt als oorzaak van blindheid en slechtziendheid in deze populatie. Vervolgens onderzoeken wij het spectrum van uveïtisentiteiten in de Thaise populatie en onderzoeken het intraoculaire vocht van uveitis patiënten met de PCR methode om de infectieuze oorzaken vast te stellen en beschrijven de bijzondere kenmerken van diverse infectieuze uveitis entiteiten. Wij hebben vastgesteld dat HIV zelf uveitis kan veroorzaken en beschrijven de typische klinische en laboratorium kenmerken van de HIV geassocieerde uveitis. Verder analyseren wij de klinische kenmerken van HLA-B27 geassocieerde uveitis en oculaire sarcoïdose in Thailand. Tot slot beschrijven wij een bijzondere vorm van centrale sereuze chorioretinopathie als maskerade syndroom van de ziekte van Vogt Koyanagi Harada.

HOOFDSTUK 1

Hierin vatten wij de beschikbare literatuur over uveitis in Zuidoost Azië samen inclusief alle gepubliceerde artikelen over uveitis in Thailand. Wij beschrijven de belangrijkste doelen van dit proefschrift, benoemen de onderzoeksvragen en beschrijven de plannen voor de individuele studies.

HOOFDSTUK 2

In dit hoofdstuk onderzoeken wij de oorzaken van blindheid en slechtziendheid in 2951 opeenvolgende nieuwe patiënten in de periode van februari tot met juni 2005 van de afdeling Oogheelkunde van het universiteitsziekenhuis in Chiang Mai, Thailand. Wij geven een overzicht van de specifieke oorzaken en de bijbehorende anatomische locaties en vermelden tevens of de blindheid en slechtziendheid voorkomen had kunnen worden. In totaal waren er 369 (12.5%) patiënten met een blind of slechtziend oog (150 bilateraal en 219 unilateraal). De meest voorkomende oorzaak van visusverlies waren leeftijd geassocieerde aandoeningen (35%; 128/369) gevolgd door infecties (18%; 66/369) en trauma (12%; 43/369).Bilaterale blindheid was aanwezig bij 73 patiënten en 19% (14/73) van dubbelzijdige blindheid werd veroorzaakt door dubbelzijdige Cytomegalovirus retinitis bij HIV-geïnfecteerde patiënten.

HOOFDSTUK 3

In dit hoofdstuk hebben wij een prospectieve studie beschreven van 200 nieuwe consecutieve patiënten met uveitis. Patiënten werden geclassificeerd op basis van de anatomische locatie van uveitis en van de etiologie of associatie met een systeemziekte. De diagnoses in deze studie werden enerzijds op klinische kenmerken gebaseerd en anderzijds op de resultaten van de screening waaronder bezinkingsnelheid van erytrocyten, bloedbeeld, serologische testen voor HIV, syfilis en toxoplasmose, urine onderzoek en röntgen foto van de thorax. Bovendien werd bij alle patiënten met uveitis anterior HLA B27 bepaald. Onze resultaten laten positieve HIV serologie zien bij 31% (62/200) van alle uveitis patiënten. Binnen deze populatie was uveitis posterior de meest frequente anatomische entiteit en CMV vormde de meest frequente oorzaak (85%; 53/62). HIV negatieve patiënten hadden frequent uveitis anterior (34%; 47/138). Een infectieuze oorzaak werd vastgesteld in 22% (30/138) en mogelijke oculaire toxoplasmose (focale retinitis met positieve toxoplasma serologie) vormde de meest frequente intraoculaire infectie (8.7%;12/138). Geassocieerde systeemziekte werd gevonden in 26% (36/138) en klinisch gedefinieerde syndromen werden gediagnosticeerd in 33% (46/138). De ziekte van Vogt›Koyanagi›Harada (18.5%; 22/108) en HLA-B27 geassocieerde uveitis anterior (9%; 10/108) waren de voornaamste oorzaken binnen de niet-infectieuze uveitis. In 19% (26/138) bleef de oorzaak onopgehelderd. Onze studie liet zien dat een derde van alle uveitis patiënten in noord Thailand HIV positief was. Van alle 200 patiënten met uveitis, werd 27% (53/200) veroorzaakt door CMV. De oorzaken van uveitis en associaties met systeemziekten binnen de HIV negatieve groep waren (voor zover bekend) vergelijkbaar met andere landen in zuidoost Azië, echter verschilden van de oorzaken van uveitis in Europa en VS.

HOOFDSTUK 4

Om de vraag te beantwoorden welke infectieuze micro-organismen een rol kunnen spelen bij het ontstaan van infectieuze uveitis in Thailand, hebben we een seroprevalentie studie verricht bij 101 HIV negatieve patiënten met uveitis en 100 HIV positieve patiënten met retinitis. De serologische resultaten werden vergeleken met de uitslagen van 100 controles zonder uveitis. Wij hebben naar de antistoffen gezocht tegen bekende met uveïtis geassocieerde microorganismen. Positieve antistoffen tegen ***T.gondii*** werden gevonden bij 31 HIV negatieve patiënten. Deze uitslag was verrassend, omdat eerdere serologische studies een lagere seroprevalentie vermelden en oculaire toxoplasmose betrekkelijk zeldzaam is. De prevalentie van positieve toxoplasma serologie binnen de uveitis posterior groep (met name bij patiënten met focale retinitis) was hoger dan bij de andere anatomische uveitis entiteiten. Wij concludeerden dat ook in Thailand rekening gehouden moet worden met ***T.gondii*** als oorzaak

van uveitis posterior. Antistoffen tegen *T.pallidum* en *Leptospira* werden vaker gevonden bij de HIV positieve patiënten met retinitis, maar hun oorzakelijke rol bij het ontstaan van retinitis kon niet aangetoond worden. De hogere seroprevalentie van syfilis bij HIV positieve patiënten suggereert dat co-infectie van HIV met syfilis vaak voorkomt.

HOOFDSTUK 5

In dit hoofdstuk wilden wij de diagnostische waarde van PCR in het oogvocht van patiënten met uveïtis onderzoeken. Wij hebben daarom een prospectieve studie verricht van 100 HIV negatieve en 47 HIV positieve patiënten met uveitis van onbekende oorzaak. Deze patiënten hadden de afdeling Oogheelkunde van het universiteitsziekenhuis in Chiang Mai bezocht tussen mei 2006 en oktober 2009. Positieve PCR uitslagen in het oogvocht werden gevonden in 45% (66/147) van alle patiënten, vaker bij de HIV positieve dan bij de HIV negatieve patiënten (70%; 33/47 versus 33%; 33/100). CMV was de meest voorkomende infectieuze oorzaak van uveitis, zowel in de HIV positieve als in de HIV negatieve populatie. Bij HIV positieve patiënten veroorzaakte CMV vooral uveitis posterior, echter in de HIV negatieve groep was CMV vooral geassocieerd met uveitis anterior. CMV geassocieerde uveitis anterior was vaak unilateraal en werd vooral gekenmerkt door hoge oogdruk tijdens actieve periodes en door het ontbreken van synechiae posteriores. De diagnose CMV retinitis kon tevens gesteld worden bij 3 HIV negatieve imuuncompetente patiënten. Infecties met *T.gondii* lieten vaak atypische kenmerken zien en verder vonden wij een positieve PCR voor *T.gondii* bij 3 patiënten met uveitis anterior. Onze conclusie is dat PCR analyse van intraoculair vocht een hoge diagnostische waarde heeft en tevens hebben wij vastgesteld dat de klinische manifestaties van intraoculaire infecties in Thailand kunnen verschillen van die in westerse landen.

HOOFDSTUK 6

In dit deel van het proefschrift hebben wij de intraoculaire en plasma HIV-1 RNA loads van 40 HIV positieve patiënten met uveitis onderzocht en beschrijven wij de klinische kenmerken van uveitis met aantoonbare intraoculaire HIV (met name bij de patiënten waarvan de HIV-1 RNA load in het oog hoger was dan in hun plasma). HIV-1 RNA in het oog was aantoonbaar bij 33% (13/40) van de HIV positieve patiënten met uveitis. Intraoculaire HIV was geassocieerd met hogere plasma loads en het niet hebben van HAART. Bij 4 patiënten waren de HIV loads in het oog hoger dan in plasma. Drie van deze patiënten hadden uveitis anterior en vitritis zonder geassocieerde retinale laesies. Geen van deze patiënten had aanwijzingen voor een andere (opportunistische) infectie. Allen reageerden zeer vlot op de HAART behandeling. Wij concluderen dat intraoculaire HIV loads detecteerbaar zijn bij een aanzienlijke percentage

HIV positieve patiënten met uveitis, met name bij (nog) onbehandelde patiënten met hoge HIV plasma loads. Onze resultaten laten zien dat uveitis anterior zonder geassocieerde retinale laesies kan veroorzaakt worden door HIV zelf.

HOOFDSTUK 7

In dit hoofdstuk onderzoeken we het voorkomen van de HLA B27-geassocieerde AAU in Thailand en beschrijven de klinische karakteristieken. Eerst hebben wij de frequentie van HLA B27 bepaald bij 100 gezonde Thaise mensen en bij 121 patiënten met uveitis anterior. De frequentie van HLA B27 was 10% bij de bloeddonoren en 44% bij de patiënten met AAU ($p< 0.001$). De oculaire klinische kenmerken kwamen overeen met die in de westerse landen. Tenminste 15% van de HLA B27 geassocieerde uveitis had radiologische kenmerken van ankyloserende spondylits vergeleken met ongeveer 40%-50% uit de (westerse) literatuur. Een mogelijke oorzaak van de discrepantie kan zijn, dat niet al onze patiënten door een reumatoloog geëvalueerd werden. We concluderen dat HLA B27 geassocieerde uveitis anterior voorkomt in Thailand en dezelfde oogheelkundige karakteristieken heeft als elders in de wereld. De oorzaken van verschillen in het voorkomen van geassocieerde systeemziekten zouden verder onderzocht moeten worden.

HOOFDSTUK 8

Sarcoidose is een uiterst zeldzame diagnose in Thailand en oculaire sarcoidose werd tot dusver nooit beschreven. In dit onderdeel verrichtten wij een retrospectieve studie naar het bestaan van de oculaire sarcoidose in Thailand. Hiertoe bestudeerden wij 209 röntgen thorax foto's verricht bij 209 nieuwe opeenvolgende patiënten met uveitis van de afdeling Oogheelkunde van het universiteitsziekenhuis in Chiang Mai. Tevens bestudeerden wij 7 CT's van de longen van de patiënten met vitritis (n=4) en perifere multifoale chorioditis (PMC, n=3). Vervolgens beschreven we oculaire manifestaties van Thaise patiënten met uveitis en pulmonale sarcoidose. Wij hebben vastgesteld dat tenminste 4/209 (2%) van de uveitis patiënten tekenen van pulmonale sarcoidose toonden. Van de 209 geanalyseerde thoraxfoto's waren slechts bij 1 patiënt (0.5 %) afwijkingen kenmerkend voor sarcoidose stadium 1 gevonden. Bilaterale hilaire lymphadenopathie op de CT's van de thorax was vastgesteld bij alle 3 patiënten met PMC en was afwezig bij de patiënten met vitritis. Bij 2 patiënten met PMC en bilaterale lymphadenopathie op de CT werd een longbiopsie verricht met positieve uitkomst in beide gevallen. Alle 3 patiënten met pulmonale lymphadenopathie en PMC waren vrouwen ouder dan 50 jaar zonder geen pulmonale klachten met een geheel normale röntgen foto. Wij tonen aan dat oculaire sarcoidose in Thailand voorkomt en vaker dan eerder was aangenomen.

Wij verwachten dat meer patiënten geïdentificeerd zullen worden als de diagnose sarcoidose overwogen wordt en de diagnostische mogelijkheden benut.

HOOFDSTUK 9

In dit hoofdstuk beschrijven wij een bijzonder maskerade syndroom bij 7 patiënten die aanvankelijk verdacht werden van de ziekte van Vogt Koyanagi Harada (VKH). Zij werden echter bij nader onderzoek gediagnosticeerd met diffuse retinale pigment epitheliopathie (DRPE) met multipele sereuze retinale loslatingen. Wij beschrijven de klinische presentatie en reactie op de therapie bij deze patiënten en bespreken de kenmerken die DRPE kunnen differentiëren van de ziekte van VKH. Hoewel de retinale beelden van beide aandoeningen kunnen overlappen, hebben patiënten met VKH tevens een geassocieerde vitritis en neuritis, zoals op de late opnames van fluoresceïne angiografie vastgesteld kan worden. Tevens hebben VKH patiënten extra-oculaire symptomen die bij DRPE patiënten ontbreken. DRPE patiënten hebben meestal tevoren corticosteroiden gebruikt en reageren met verergering van de oogafwijkingen indien deze medicatie gebruikt wordt. Wij concluderen dat de ernstige varianten van DRPE geassocieerd met bulleuze sereuze retinale loslating verward kunnen worden met VKH en dat de vroege en juiste diagnose van DRPE ernstige bijwerkingen van de onnodige medicatie, operaties en zelfs blijvend visusverlies kan voorkomen.

CONCLUSIES

Dit proefschrift identificeert de belangrijkste oorzaken van uveitis in de Thaise populatie en beschrijft bovendien tot dusver onbekende oorzaken van uveitis in Thailand en hun typische klinische kenmerken. Onze resultaten geven aan dat de behandeling en het toekomstige onderzoek op het gebied van uveitis sterk zouden kunnen profiteren van de analyse van intraoculair vocht door middel van GWC. Hiermee is het mogelijk om patiënten met chronische intraoculaire infecties, die negatief zijn in het PCR onderzoek, alsnog te diagnosticeren en gericht te behandelen. Verder onderzoek naar de klinische kenmerken van oculaire toxoplasmose is wenselijk omdat onze voorlopige resultaten suggereren dat de klinische kenmerken van oculaire toxoplasmose in Thailand verschillen van het klassieke beeld van focale necrotiserende retinitis in westerse landen. De prevalentie van oculaire sarcoidose in Thailand zou nader opgehelderd kunnen worden door een prospectieve studie van (een cohort van) uveitis patiënten met onder andere een CT-scan van de thorax, een gevoeligere methode voor de diagnose sarcoidose dan de meer gangbare en goedkopere röntgen foto. Een vergelijkend

onderzoek van röntgenfoto en CT kan hun respectievelijke diagnostische waarde binnen de uveitispopulatie nader verduidelijken. Het systematisch uitvoeren van weefselbiopsieën en het uitvoeren van de QuantiFeron test bij uveïtispatiënten zal meer duidelijkheid kunnen verschaffen betreffende de prevalentie van sarcoidose en tuberculose binnen de uveïtispopulatie. De vraag in welke cellen HIV zich binnen het oog kan repliceren kan beantwoord worden door het bestuderen van humaan weefsel (bijv. iris- of retinabiopsieën). Nader onderzoek van de karakteristieken van intraoculaire HIV en vergelijking met het in bloed gevonden virus kunnen hun eventuele verschillen duidelijk maken. Tenslotte, het zou wenselijk en waardevol zijn om de PCR faciliteiten binnen oogheelkundig Thailand uit te breiden, zodat analyse van intraoculaire vloeistoffen ook mogelijk zal worden voor patiënten en oogartsen buiten ons ziekenhuis.

ACKNOWLEDGEMENTS

First, I would like to acknowledge all foundations and organizations which supported our research projects, specifically Dr F.P. Fischer-Stichting, Amersfoort, Dr. P. Binkhorst Foundation for Ophthalmologic research, Nijmegen; Stichting Blindenhulp, s-Gravenzande; Stichting Nederlands Oogheelkundig Onderzoek, Rotterdam, Rotterdamse Vereniging Blindenbelangen, Rotterdam; Landelijke stichting voor Blinden en Slechtzienden, Utrecht; Katholieke Stichting voor Blinden en Slechtzienden, Grave, and Stichting Nelly Reef Fund, Ugchelen, the Netherlands.

I feel very much obliged to my Promotor "Professor Dr. Aniki Rothova" who has opened the opportunities and possibilities for me to achieve my PhD degree at University Medical Center Utrecht. She gave me the inspiration and encouragement. I have learned tremendously from her, not only how to work but also how to think wisely. Without her, I could not have come this far. Thank you so much for being my guide, great teacher and guru.

Professor Dr. Aniki Rothova, is however not only my promotor and supervisor. She has also brought many new useful technologies to Thailand which had not been available before. With the introduction of this technology, we were able to solve several of the "mysterious diseases" in Chiang Mai. The use of the novel diagnostic techniques is very helpful to the doctors and patients. Dr. Aniki also opened the "Gate to Asia" for medical students of University Medical Center Utrecht. With this new connection, several medical students come to visit our department for "Co-schappen oogheelkunde" every year.

I would like to thank Prof I.M. Hoepelman, Prof Dr S.M. Imhof, Prof Dr A. Kijlstra, Prof Dr J. S. Stilma and Dr J.H. de Boer for their work being the members of my PhD committee.

I am extremely grateful to Jolanda de Groot-Mijnes PhD and Annemarie Weersink PhD from the Department of Virology, University Medical Center Utrecht and Natedoa Kongyai, Assistant Professor Wasna Sirirungsi PhD, Assistant Professor Pranee Leechanachai PhD from Department of Medical Technology, Faculty of Associated Medical Sciences, Chiang Mai University. Thank you very much for teaching and bringing a lot of advanced technology to Chiang Mai. This is a "Breakthrough" phenomenon for Uveitis research in Thailand. It is my honor to work with you and I feel very lucky to have all of you around me.

I would like to thank Dr. Mary Van Schooneveld, Dr. Joke H. de Boer, Dr. Ninette H. ten Dam-van Loon for your help, kindness and advice. You have been very friendly to me.

My deepest gratitude to Professor Somsaguan Ausayakhun, Head of Department of Ophthalmology, Chiang Mai University, who introduced me to Dr. Aniki. Without you and your article, I would never found this "Treasure". I appreciate your support and encouragement very much. Thank you for being my best boss.

I am grateful to all of my colleagues especially Dr. Paradee Kunavisarut; Associate Professor Dr. Sopa Wattananikorn; Associate Professor Dr. Nitmitr Ittipankul; Associate Professor Dr. Prapatsorn Patikulsila; Associate Professor Dr. Direk Patikulsila; Associate Professor Dr. Napaporn Tananuvat and Dr. Janejitr Choovuthayakorn and all residents from Department of Ophthalmology, Chiang Mai University for your support and participation in the studies. Your help has hugely contributed to the completion of my research work.

I am deeply indebted to all patients and their family members participating in the studies. You have not been solely a patient, but also a "teacher" to me. You helped me help other patients.

Last but not least, I dedicate all the effort and success of this thesis to my beloved father, mother, brothers and sisters who always loved and supported me since the day I was born. They are always with me no matter what happens. Thank you very much.

CURRICULUM VITAE

The author was born on November 23, 1970 in Bangkok, Thailand. She received her medical degree in 1992 at Chulalongkorn University, Bangkok and her specialization in Ophthalmology in 1995 at Chiang Mai University, Chiang Mai. She pursued her subspecialty in Glaucoma at University of British Columbia, Vancouver, Canada in 1997 and in Uveitis at the same University in 1998. At present she is an instructor at the Department of Ophthalmology, Faculty of Medicine, Chiang Mai University, Thailand.

In 2005-2010, she performed research at Uveitis clinic, Department of Ophthalmology, University Medical Center Utrecht, The Netherlands and Uveitis clinic, Department of Ophthalmology, Chiang Mai University, Thailand and also with the Department of Medical Technology, Faculty of Associated Medical Sciences, Chiang Mai University, Thailand. Her work was supervised by Professor Aniki Rothova and resulted in this PhD thesis.

Kessara Pathanapitoon
Uveitis in Thailand
Emphasis on clinical patterns and novel developments
in diagnostics using intraocular fluid analysis

ISBN: 978-90-393-53707

Cover Design and Lay-out: Within Design Co., Ltd.

Printed by: Within Design Co., Ltd.