Identification of Malaria Retinopathy Improves the Specificity of the Clinical Diagnosis of Cerebral Malaria: Findings from a Prospective Cohort Study

Gretchen L. Birbeck,* Nicholas Beare, Susan Lewallen, Simon J. Glover,
Malcolm E. Molyneux, Peter W. Kaplan, and Terrie E. Taylor

International Neurologic and Psychiatric Epidemiology Program, Michigan State University, East Lansing, Michigan; St. Paul's Eye Unit, Royal Liverpool University Hospital, Liverpool, United Kingdom; Kilimanjaro Centre for Community Ophthalmology, Tumaini University/KCMC, Moshi, Tanzania; Anatomy Department, University of Malawi College of Medicine, Blantyre, Malawi; Malawi-Liverpool-Wellcome Trust Clinical Research Programme, College of Medicine, Malawi and The Liverpool School of Tropical Medicine and Hygiene, University of Liverpool, United Kingdom; Department of Neurology, Johns Hopkins Health Systems, Baltimore, Maryland; College of Osteopathic Medicine, Michigan State University, East Lansing, Michigan; Blantyre Malaria Project, University of Malawi College of Medicine, Blantyre, Malawi

Abstract. The diagnosis of cerebral malaria (CM) is difficult to confirm in endemic regions with limited neurodiagnostics. Accurate diagnoses are critical for trials and outcomes studies. Findings from an autopsy-based study suggest that identifying malaria retinopathy in children satisfying the standard clinical case definition of CM improves our ability to accurately diagnose CM *in vivo*. In a *post hoc* analysis of a prospective exposure-control study to evaluate CM as a risk factor for epilepsy, we stratified children meeting the standard case definition by their retinopathy status (presence versus absence) and compared these groups for pre-existing risk factors for epilepsy. We also compared them to the concurrently enrolled, non-comatose controls. Children meeting the standard case definition of CM who lacked malaria retinopathy had a higher prevalence of pre-existing developmental problems and family history of epilepsy. This subset of patients may represent children with a pre-existing propensity to adverse neurologic symptoms and outcomes.

INTRODUCTION

Distinguishing children with cerebral malaria (CM) from those with non-malarial comas or an incidental parasitemia is difficult, especially in malaria-endemic areas with high rates of asymptomatic parasitemia (e.g., no imaging facilities, limited laboratory support, and no recourse to electroencephalography). An autopsy study of children meeting the standard clinical case definition for CM (a Blantyre Coma Score of ≤ 2 in a child with asexual *Plasmodium falciparum* parasitemia and no other evident etiology for coma^{1–3}) who were admitted to Queen Elizabeth Central Hospital (QECH) in Blantyre, Malawi found that 23% (7/31) did not have sequestration of parasites in central nervous system (CNS) vasculature, the hallmark of CM pathology, but had other causes of death identified including pneumonia, Reye's syndrome, and head trauma.⁴ The clinical expertise and diagnostic facilities available at QECH, a tertiary care hospital, undoubtedly exceed those generally available in most African district hospitals and clinics. Rates of asymptomatic parasitemia in sub-Saharan Africa (SSA) may be as high as 80% in heavily affected regions.^{5–7} Therefore, the overdiagnosis of CM, or misclassification of coma cases as being caused by malaria when in fact another underlying cause exists, likely results in substantial limitations for clinical trials of antimalarial medications and other malaria-specific interventions as well as “under-diagnosis” (or delayed recognition) of non-malarial causes of coma in parasitemic children.

Studies aimed at quantifying the neuropsychiatric sequelae of CM may also be adversely impacted by the problem of overdiagnosis or misclassification. In a retrospective cohort study conducted in Kilifi, Kenya, researchers found that children with CM (using the standard clinical case definition) had an odds ratio of 4.4 (95% CI = 1.4–13.7) for the later development of epilepsy.⁸ Seizures are a common occurrence in malaria, typically identified in > 60% of pediatric CM cases,^{2,9}

so the Kilifi study also examined the risk of later epilepsy development among non-comatose children who had malaria complicated by seizures. Paradoxically, this non-comatose “malaria and seizure” group had an odds ratio for epilepsy of 6.1 (95% CI = 2.0–18.3), somewhat higher than that found in the CM population, many of whom had experienced seizures during their acute illness. These findings could be caused by the misattribution of coma to CM in the setting of incidental parasitemia among children with a pre-disposition to prolonged or recurrent seizures when challenged with an acute illness. Alternatively, malaria-induced seizures, even in the absence of coma, could be the epileptogenic etiology in this group of children who develop epilepsy after a malaria infection.

Recent advances in CM diagnosis based on identification of a malaria retinopathy may provide an opportunity to improve on the standard clinical case definition of CM.^{10–12} Malaria retinopathy is characterized by retinal whitening, vessel changes, and/or hemorrhages (frequently with white centers) in patients meeting the standard clinical case definition of CM (Figures 1–4). Papilledema may accompany any of these features when they are severe, but when present without the other findings, papilledema is not specific. Children with malaria retinopathy differ significantly in a number of clinical and laboratory parameters compared with children who meet the standard clinical case definition of CM but do not have the retinopathy.¹² At autopsy, eyes containing retinal vessels with malaria retinopathy changes seen on clinical examination have retinal blood vessels containing many dehemoglobinized parasitized red blood cells on histopathological examination,¹¹ and angiography has confirmed that retinal whitening is associated with impaired perfusion.¹³ A validation study using autopsy as the gold standard comparison found that the presence of malaria retinopathy has 100% specificity and 95% sensitivity for the identification of CM in fatal disease.¹⁴

In an ongoing, prospective, exposure-control study aimed at evaluating CM as a risk factor for epilepsy, we recruited CM survivors from the Pediatric Research Ward at QECH in Blantyre, Malawi who, on admission, had met the standard

*Address correspondence to Gretchen L. Birbeck, #324 West Fee Hall, East Lansing, MI 48824. E-mail: Gretchen.Birbeck@ht.msu.edu

FIGURE 1. CM patient with retinal whitening in the central macula and subtle orange discoloration of the blood vessels. This figure appears in color at www.ajtmh.org.

clinical case definition of CM. As part of an ongoing autopsy study of clinicopathological correlates of fatal malaria, all children also underwent an ophthalmological assessment looking for evidence of malaria retinopathy. We undertook a *post hoc* analysis of children with CM (per the standard clinical case definition) to determine whether or not there was a difference in pre-existing risk factors for epilepsy between those with malaria retinopathy and those without it. Our purpose was to assess what proportion of children presenting to the hospital with apparent CM have evidence of a pre-existing susceptibility to seizures in order to take this into account in our ongoing study of subsequent epilepsy risk. Our hypothesis was that children with such a pre-disposition would be over-represented in the retinopathy-negative group.

MATERIALS AND METHODS

Study design. This was a *post hoc* analysis of admission characteristics among children participating in a prospective, exposure-control study of CM survivors. Analytic groups included CM with malaria retinopathy, CM without malaria retinopathy, and a non-comatose, age-matched, concurrently admitted control group of children from the general pediatrics ward. Admitting diagnoses among the control group included uncomplicated malaria, pneumonia, and gastroenteritis.

Setting and participants. On the QECH Pediatric Research Ward, children who met the standard clinical case definition of CM were evaluated for enrollment in a prospective exposure-

FIGURE 2. CM patient with several features of CM retinopathy including macular and peripheral retinal whitening, hemorrhages, and papilledema. This figure appears in color at www.ajtmh.org.

control study of CM outcomes aimed at determining whether or not CM was a risk factor for later epilepsy. Recruitment occurred between March and December of 2005 and then from January to June annually (peak malaria season) in 2006 and 2007. Each enrolled CM child was age-matched to a concurrently admitted child from the general pediatrics ward. Matching strategy was 1:1 in 2005–2006. When the importance of including CM retinopathy in our diagnostic criteria became evident in 2007, we changed our matching strategy to 2:1 to try and increase our power for the ongoing prospective study, because requiring this additional inclusion criterion would inevitably decrease our planned recruitment. Exclusion criteria for all groups included having any history of unprovoked seizure before the index illness and residing outside of the Blantyre area. The Epilepsy Screening Questionnaire was used to identify children with a history of previous unprovoked

FIGURE 3. Typical CM retinopathy retinal hemorrhage with central whitening. This figure appears in color at www.ajtmh.org.

FIGURE 4. Vessel whitening typical of CM retinopathy with involvement of the capillary bed as well as larger retinal vessels. Retinal whitening is also seen. This figure appears in color at www.ajtmh.org.

seizure. This nine-item questionnaire has been previously used with 79.3% sensitivity and 92.9% specificity in similar populations.¹⁵

Variables. At enrollment, data obtained for the CM and comparison populations included demographic data on age and gender, biomedical characteristics including birth weight, an Apgar proxy assessment, body mass index (BMI), and admission temperature, and information regarding any pre-existing risk factors for epilepsy including a history of developmental problems and a family history of epilepsy.

Birth weight was abstracted from the health passport, medical record, or parental report. Mothers were asked whether or not their child cried immediately after delivery, and the answer was treated as a dichotomous Apgar proxy. BMI was derived from height and weight measurements taken on the day of discharge. A historical Ten Questions screen was used to identify pre-index illness developmental problems. This instrument has been shown to be valid in children as young as 2 years^{16,17} and has been evaluated in similar environments with good reliability ($\kappa = 0.67$) and 85% sensitivity for detect-

ing moderate neurodevelopmental disabilities. The Epilepsy Screening Questionnaire was used to determine whether or not the child had a prior history of unprovoked seizure and to ascertain whether or not the study subjects had any first-degree relatives (parents or siblings) with a history of epilepsy. Further detailed questions were asked by a physician if any ambiguity remained after the Ten Questions and Epilepsy screens. All data were recorded on standardized questionnaires before entry into Microsoft Access and analysis using EPI INFO (<http://www.cdc.gov/epiinfo/about.htm>).

Statistical methods. An analysis of variance (ANOVA) was used to compare admission characteristics across the three analytic groups (CM with retinopathy, CM without retinopathy, and controls). Then, two additional analyses were conducted comparing 1) the malaria retinopathy negative CM children to controls and 2) the malaria retinopathy positive CM children to controls to obtain odds ratios (with 95% confidence intervals). Where any subgroup included < 5 cases, Fisher's Exact tests were used. If Bartlett's test for inequality indicated the need, the Kruskal-Wallis test was applied.

Role of the funding sources. The study sponsors played no role in the study design, collection, analysis, or interpretation of the data, in writing this manuscript, or in the decision to submit this paper for publication.

RESULTS

During the observation period, 167 children meeting the standard clinical case definition for CM were enrolled. Among these, 132 (79.0%) had malaria retinopathy, and these children were matched to controls (2:1 matching commenced in year 3 of enrollment, resulting in a total of 264 controls). Table 1 provides the comparisons across all three groups (CM with malaria retinopathy, CM without retinopathy, and controls). Compared with controls, the children who met the standard clinical case definition of CM but lacked malaria retinopathy were significantly more likely to have a pre-existing history of developmental delays (OR = 5.75; 95% CI = 2.06–16.02) and a positive family history of epilepsy (OR = 3.50; 95% CI = 1.51–10.62). The CM children with malaria retinopathy did not differ from the control group on these or any other characteristics assessed ($P > 0.05$).

TABLE 1
Comparison of children meeting the standard clinical case definition of CM with and without malaria retinopathy vs. controls

Characteristic	Controls (N = 264)	Retinopathy positive (N = 132)	Retinopathy negative (N = 35)	P value (ANOVA)	Retinopathy negative vs. control*	Retinopathy positive vs. control
Age (months)	42.8	42.3	42.9	0.98	$P = 0.99$	$P = 0.85$
Sex (% male)	56.1	49.2	40.0	0.13	OR = 0.52 (CI = 0.25–1.07)	OR = 1.32 (CI = 0.87–2.00)
Birthweight (kg)	3.2 (N = 173)	3.4 (N = 80)	3.3 (N = 30)	0.67	$P = 0.89$	$P = 0.88$
Admit temp (°C)	38.4	38.3	38.2	0.72	$P = 0.43$	$P = 0.68$
Body mass index	15.6	15.6	15.4	0.82	$P = 0.88$	$P = 0.99$
Apgar proxy†	8.0 (3.0%)	4.0 (3.1%)	2.0 (5.7%)	0.69	OR = 0.52 (CI = 0.11–2.53)	OR = 1.01 (CI = 0.30–3.41)
History of severe malaria‡	12.0 (4.5%)	6.0 (4.5%)	21.0 (2.9%)	0.90	OR = 0.62 (CI = 0.08–4.90)	OR = 1.00 (CI = 0.37–2.73)
History of developmental problem(s) (%)§	11.0 (4.2%)	11.0 (8.3%)	7.0 (20.0%)	0.0014	OR = 5.75 (CI = 2.06–16.02)	OR = 0.48 (CI = 0.20–1.13)
Family history of epilepsy (%)	12.0 (4.5%)	10.0 (7.6%)	5.0 (14.3%)	0.06	OR = 3.50 (CI = 1.51–10.62)	OR = 0.58 (CI = 0.24–1.38)

*t test or χ^2 test used.

†Mother was asked, "Did the child cry immediately after delivery?"

‡Malaria requiring prior hospital admission per the health passport.

§History of developmental problems as per the Ten Question Screen.

DISCUSSION

Children who meet the standard clinical case definition of CM but who lack malaria retinopathy are more likely to have pre-index illness risk factors for seizures. In this *post hoc* analysis, children with pre-existing neurodevelopmental abnormalities and/or a genetic pre-disposition to seizures are more likely to experience prolonged and/or complicated seizures when challenged with a toxic, metabolic, or infectious perturbation.^{18–20} Unfortunately, inclusion of these children in CM clinical trials or outcomes studies may produce misleading results, because these children already have pre-determined neurologic outcomes before the index CM illness of interest. Interventions aimed at decreasing long-term neurologic sequelae of CM that include children with a pre-disposition to adverse neurologic outcomes will, at the very least, be substantially underpowered to find a beneficial effect. Outcomes studies of such children may incorrectly attribute adverse outcomes to CM. Adding the presence of malaria retinopathy to the diagnostic criteria for pediatric CM will increase diagnostic specificity and may help researchers avoid potential confounding factors related to CNS injuries that preceded the acute CM infection. Future clinical trials and outcomes studies of CM should include the CM retinopathy criterion.

Received September 9, 2009. Accepted for publication November 16, 2009.

Acknowledgment: The authors thank the Blantyre Malaria Project Epilepsy Study (BMPEs) nurses for their important role in this work.

Financial support: Funding for this work was provided by National Institutes of Health Grant NIH/NINDS K23 NS046086-01, National Institutes of Health Grant NIH/NIAID R01 AI034969-10A1, and The Wellcome Trust.

Authors' addresses: Gretchen L. Birbeck, Michigan State University, International Neurologic and Psychiatric Epidemiology Program, East Lansing, MI, E-mail: Gretchen.Birbeck@ht.msu.edu. Nicholas Beare, St. Paul's Eye Unit, Royal Liverpool University Hospital, Liverpool, E-mail: nbeare@btinternet.com. Susan Lewallen, Kilimanjaro Centre for Community Ophthalmology, Tumaini University/KCMC, Moshi, Tanzania, E-mail: slewallen@kcco.net. Simon J. Glover, University of Malawi, Anatomy Department, Blantyre, Malawi, E-mail: simontheyeman@hotmail.com. Malcolm E. Molyneux, Malawi-Liverpool-Wellcome Trust Clinical Research Programme, College of Medicine, Blantyre, Malawi, E-mail: mmolyneux999@gmail.com. Peter W. Kaplan, Johns Hopkins Health System, Department of Neurology, Baltimore, MD, E-mail: pkaplan@jhmi.edu. Terrie E. Taylor, Michigan State University, College of Osteopathic Medicine and University of Malawi College of Medicine, Blantyre Malaria Project, East Lansing, MI, E-mail: taylort@msu.edu.

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