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CASE REPORTS

Applying the Milwaukee Protocol to treat canine rabies in Equatorial Guinea

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Abstract
In this first report of rabies in Equatorial Guinea, problems accompanying the application of the Milwaukee Protocol are described. With its apparent success, and despite a subsequent death from complications of malnutrition, we sound a note of optimism that canine as well as bat rabies may be treatable.

Introduction
Rabies is an almost universally fatal encephalitis caused by rabies virus and other lyssaviruses. In 2004, a teenager survived rabies after the use of intense anti-excitatory strategy that included prolonged general anesthesia, antiviral drugs with supportive intensive care, and no immune prophylaxis until the native immune response matured [1]. This has been termed the Milwaukee Protocol (MP).

Subsequently, it has been applied to 17 other rabies patients. During the first 12 attempts, biochemical and physiological complications of human rabies were identified [2,3], and consequently the MP was updated (available at www.mcw.edu/rabies). Subsequent to our case, 2 other rabies survivors have been reported in the general press [4]. A Colombian girl survived rabies without rabies vaccine. A Brazilian boy, a rabies vaccine failure, also survived. The Milwaukee survivor and these 2 unpublished cases all involved rabies viruses of bat origin. Most rabies is of canine origin, which is hypothesized to be more virulent than bat strains [5].

The objective of this case report is to describe the possible recovery of a child from canine rabies using the MP and to describe the immunological correlates of this child’s neurological recovery. Unfortunately, the child died of complications of malnutrition.

Case report
A 5-y-old resident of Equatorial Guinea had been bitten by a dog on the neck 5 weeks before hospitalization. His wound was cleaned, and he was given antibiotics and tetanus, but not rabies prophylaxis. On 16 December 2007 he developed difficulties in swallowing, and 2 d later he was hospitalized with hydrophobia, aerophobia, phonophobia, agitation, anxiety, combativeness, unsteady gait and drooling, resulting in a diagnosis of rabies. On admission (d 1), leukocyte count was $21.2 \times 10^9$/mm$^3$ and hemoglobin 11.7 g/dl. Total protein was 5.6 g/dl, albumin 2.8 g/dl, aspartate aminotransferase 130 U/l, lactic dehydrogenase 1477 U/l, and creatine kinase >2000 U/l. Brain computed tomography on d 3 and 10 was normal.
After the diagnosis and prognosis of rabies was clearly explained, the parents agreed to treatment following the published MP [1]. The patient was intubated and sedated with a fixed ratio of 20 mg ketamine/9 mg midazolam, delivering on average 3.12 mg/kg/h of ketamine and 1.4 mg/kg/h of midazolam. Oral amantadine (6 mg/kg/d) was started on d 3 and coenzyme Q10 on d 5. Oral ribavirin (15 mg/kg/6 h) was started on d 5. Hemoglobin was maintained above 10 g/dl with blood transfusions.

Throughout the first 4 d of treatment, the patient had eyelash and facial tremors. Having no EEG or bispectral index monitoring, we used these manifestations as clinical guidance to his cerebral electric activity, and suppressed it by boluses of phenobarbital, propofol or diazepam. Despite the continuous anesthesia, the patient appeared to be awake and restless, resisting the ventilator and continuous anesthesia, the patient appeared to be resting. An isolated convulsion occurred the next d.

The patient developed an ileus on d 12. Enteral feeding and oral medications were stopped; total parenteral nutrition was not available. Serum albumin declined to 1.7 mg/dl and the patient developed edema. On d 14, he developed signs of diabetes insipidus. Lacking antiuretic hormone replacement, we attempted to maintain renal function with large quantities of fluids and diuretics. Two d later, the patient’s bowl function improved and nasogastric feeding was resumed. He developed anasarca. Fresh frozen plasma and albumin were not available. Blood pressure instability was treated with hydrocortisone, dopamine, dobutamine and adrenalin. Sedation was stopped on d 17, in order to evaluate neurological function. By the evening of that d, he could feebly move his legs in response to verbal requests, but no arm movements could be elicited. He reacted to pain and light touch. On d 20 he became anuric and died on the twenty-second d of hospital stay.

**Laboratory findings**

Cerebrospinal fluid, serum, saliva and skin were sent at diagnosis to Israel for testing in a rabies reference laboratory. Saliva was positive for rabies by reverse transcriptase polymerase chain reaction, and the skin was positive for rabies by direct fluorescent antibody testing. The virus was isolated from mice injected with the skin biopsy. Molecular analysis of the entire nucleoprotein gene sequences (GenBank accession number FJ440104) showed 99% identity with a Gabon canine rabies type virus, later confirmed at the Center for Disease Control, USA.

Rabies vaccination and administration of immune globulin were avoided, as stipulated in the MP. Rabies neutralizing antibody was first detected in serum (1:39 titer) on d 14 of hospitalization (18 d of symptoms), and in the cerebrospinal fluid (CSF) 2 d later (titer 1:39). Antibody titers increased over the next 3 d to 1:660 (serum) and 1:116 (CSF). This early part of immune response was delayed by approximately 1 week compared to that reported for the recovered survivor [1].

**Discussion**

We report the possible recovery from confirmed canine rabies in a child treated with the MP. This is first confirmation of rabies in Equatorial Guinea.

Rabies causes progressive neurological decline, culminating in coma that precedes death. Rabies usually causes death within 7 d of onset of symptoms, while the 90th percentile for survival from rabies among rabies cases reported from North America and Europe in 2000–2007 was 19 d [2]. Time from onset of neurological symptoms to coma for 19 rabies patients that had received intensive care in 2000–2007 is depicted in Figure 1. (Excluded were cases of transplantation associated rabies, that have shorter latency to coma, and patients treated with the MP, who have a longer latency.) Coma was invariably present within 12 d. It is unusual for a patient with furious rabies to follow commands, much less survive, after 18 d of rabies, as our patient did.

The demonstration of higher order neurological functions on the 11th and 17th d in our case could be interpreted in 2 ways. First, the patient’s course might be considered atypically slow but consistent with progression of rabies to hypotension and death. This explanation affirms the capability of the MP to attenuate the course of rabies, as reported previously [2]. Coma, the most common terminal sign of rabies, was notably absent, differentiating this patient’s clinical course from that of several other patients in whom coma was present, electrographically and clinically, for d prior to death [2,6]. Under
this reasoning, the patient was surviving rabies, and his death resulted from malnutrition and renal failure.

The second possibility is that this patient showed neurological recovery with the advent of rabies-neutralizing antibody. Recovery correlated better with first detection of neutralizing antibody in serum than in the CSF. The acquired diplegia on d 17 is reminiscent of the previous survivor, who was flaccid (on d 11), with recovery of reflexes in the legs (d 14) before the arms (d 16). Under this interpretation, neurological recovery in rabies is associated with low titers of neutralizing antibody in the Milwaukee survivor and this patient. In a murine model of rabies, non-pathogenic clearance of attenuated rabies virus also occurred at low levels of serum anti-rabies antibodies and of intrathecal antibody synthesis [7].

This patient’s immune response to rabies was delayed more than in most reports [8]. He was at risk for immunosuppression from malnutrition, ascariasis infection, anemia and low serum albumin. He progressed to overt malnutrition following his rabies associated ileus. He also received barbiturates, which are recognized immunosuppressors, as well as ribavirin, an antiviral drug with immunomodulatory properties [9,10]. The revised MP now recommends against use of ribavirin and barbiturates.

Among the 17 known attempts to replicate the MP (including our patient), this case and 2 others met its assumptions and complied closely in the use of drugs. The preceding 2 patients suffered complications attributable to spasm of the basilar arteries [2,6]. Our patient was the first rabies patient to receive a calcium channel blocker. The revised MP recommends considering nimodipine prophylaxis in patients with confirmed rabies, which prevents complications due to vasospasm following subarachnoid hemorrhage in adults [11]. Since nimodipine was unavailable, we administered nifedipine. The use of the calcium channel blocker might have resulted in improved outcome following the suspected vasospasm and the respiratory arrest. The dose of ketamine in this patient exceeded by 50% that used in other patients, so a beneficial dose response might be inferred. The ketamine dose far exceeded, in bioequivalence, that used in discordant rodent models assessing ketamine treatment of rabies [12,13]. Our patient did not receive tetrahydrobiopterin supplementation because it was not available, but such had been associated with normalization of dopamine and serotonin turnover in the CSF of 2 rabies patients [3].

Our patient did not survive. Thus, he cannot fully support the effectiveness of the MP for treating canine rabies, but we can sound a note of optimism that canine as well as bat rabies may be treatable. Definitive treatment of rabies requires an appropriate animal model followed by controlled clinical trials. In the interim, careful, motivated attempts to replicate the MP could produce further biochemical

Figure 1. Kaplan-Meier product-limit estimate of the time from onset of neurological symptoms to development of coma in 19 rabies patients supported by intensive care and published in 2000-2007 (see text for exclusions). Coma was defined as the authors’ use of the terms ‘coma’ or ‘major deterioration in neurological function’ after intubation, or from the onset of pupillary abnormalities, or cerebral edema by neuroimaging. Patients electively intubated, without precise description of coma onset, were censored at time of intubation. The neurological status of our patient is shown relative to this curve.
and physiological insights and, hopefully, recovery from rabies, even when attempted in developing countries.

**Declaration of interest:** The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

**References**


**Community acquired Staphylococcus aureus meningitis in adults**

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**Abstract**

We present 9 patients with community acquired Staphylococcus aureus meningitis. Foci of infection outside the central nervous system were present in 8 (89%) patients, mostly endocarditis and pneumonia. Cardiorespiratory complications occurred frequently and 6 patients died (67%). Identification and treatment of the primary focus of infection should be a priority in these patients.

**Introduction**

Staphylococcus aureus is an uncommon cause of community acquired meningitis, and has been associated with endocarditis [1–3]. Here we describe 9 cases with community acquired staphylococcal meningitis from a nationwide prospective cohort study. This observational study with anonymous patient data was carried out in accordance with Dutch...