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## Field research in humanitarian medical programmes

### Médecins Sans Frontières interventions against kala-azar in the Sudan, 1989–2003

K. Ritmeijer<sup>1</sup> and R. N. Davidson<sup>1,2</sup> <sup>1</sup>Médecins Sans Frontières-Holland, Amsterdam, The Netherlands; <sup>2</sup>Department of Infectious and Tropical Medicine, Northwick Park Hospital, Harrow, UK

#### Abstract

Since 1989, Médecins Sans Frontières (MSF) has provided medical humanitarian assistance during outbreaks of visceral leishmaniasis (VL; kala-azar) in Sudan. First, in western Upper Nile in southern Sudan, where a VL epidemic occurred after the resumption of the civil war in Sudan in 1983, with an estimated 100 000 deaths. Later, MSF started interventions in eastern Upper Nile and Gedaref State. In these 2 endemic regions VL incidence has risen markedly since 2001, which could be the start of a new epidemic cycle. Outbreaks of VL in Sudan remain unpredictable, and access to affected populations in war-torn southern Sudan is often hampered by insecurity. Therefore, MSF takes a flexible approach, establishing treatment centres where patients can be accessed. From 1989 to 2002, MSF treated > 51 000 VL cases in Sudan. Despite very basic field conditions, high cure rates of 95% are being achieved. Lack of diagnostics is a major obstacle to treatment, especially during epidemic situations. Therefore, development of simple and rapid technologies is required, allowing reliable diagnosis under field conditions. For treatment of VL there is a limited choice of effective, affordable drugs. There are strong indications of an emerging resistance to antimonials in Malakal. Introduction of combination therapies is urgently needed to prevent the further emergence and spread of resistance to antimonials, which are still the mainstay of VL treatment in eastern Africa. Experience with combination therapy with sodium stibogluconate (SSG) and paromomycin is promising, and combinations of SSG with liposomal amphotericin B and miltefosine are currently being explored.

**Keywords:** visceral leishmaniasis, *Leishmania donovani*, humanitarian aid, antimonials, Sudan

#### Introduction

Since its resumption in 1983, the civil war in Sudan has resulted in estimates of over 2 million dead, 4 million internally displaced, and over 400 000 refugees (MSF, 2002). The war has seriously affected health systems and resources throughout the country, but especially in the south where many of the existing health structures have been destroyed. There has also been wide-scale displacement of qualified health staff, and drugs and medical materials are insufficient in most areas. Médecins Sans Frontières (MSF) has been active in Sudan since 1985, where it has been providing medical humanitarian assistance to populations affected by war and/or epidemics, in various parts of the country, both in government- and rebel-controlled areas.

Visceral leishmaniasis (VL; kala-azar) is a major health problem in the endemic areas in eastern and central Sudan where several outbreaks due to *Leishmania donovani* have been reported since the early 1900s. In this endemic belt (Fig. 1), *Phlebotomus orientalis* is the vector, associated with *Acacia seyal* and *Balanites aegyptica* vegetation, and black cotton soils. Despite the importance of the disease, very little is known about the ecology of the vector and the transmission dynamics of the disease (Thomson *et al.*, 1999)

#### Visceral leishmaniasis in southern Sudan

Until the late 1980s, VL had been thought to be endemic only in areas ranging from Malakal and the Sobat River in the south (Upper Nile Province) to Kassala in eastern Sudan (Zijlstra & El-Hassan, 2001).

However, in 1988 increasing numbers of VL cases were found by MSF teams in Khartoum, among internally displaced persons coming from the western Upper Nile region in southern Sudan (Perea *et al.*, 1989; De Beer *et al.*, 1990). This region, south of the Bahr-el-Ghazal River and west of the White Nile, had previously not been endemic for VL. Now people reported a 'killing disease', which was decimating entire families, and for which no treatment was available in their conflict-torn region. As a result of this, thousands of people travelled the 900 km north to Khartoum to seek medical care.

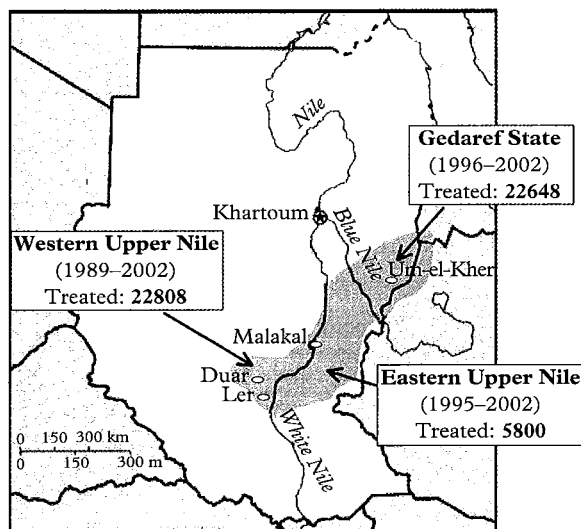


Fig. 1. Visceral leishmaniasis-endemic area (shaded) in Sudan, and MSF interventions by region, 1989–2002.

Address for correspondence: Koert Ritmeijer, Médecins Sans Frontières-Holland, Max Euweplein 40, P.O. Box 10014, 1001 EA Amsterdam, The Netherlands; phone +31 20 520 87 67, fax +31 20 620 51 70, e-mail koert\_ritmeijer@amsterdam.msf.org

MSF established an emergency hospital in Khartoum, where 700 VL cases were treated in 1989–90 (Zijlstra *et al.*, 1991). The increasing incidence in Khartoum and the testimonies of people from the area prompted an MSF investigation in western Upper Nile, which confirmed the outbreak of VL. It soon became clear that the epidemic had had a devastating impact on the Nuer and Dinka people who inhabit this region. Retrospective mortality surveys suggested that from the start of the epidemic in 1984 to 1994 around 100 000 deaths among a total population of 280 000 might be attributed to VL. In the most affected areas, up to 70% of the population had died from the disease (Seaman *et al.*, 1996). A combination of factors had created conditions suitable for *P. orientalis*, as well as for the introduction of the parasite into the community. Population movements in response to war and food shortages have been a major factor in transmission, and poor nutritional status probably increased the susceptibility to clinical disease after infection. The fact that people in western Upper Nile had no immunity to VL accounts for the high incidence in all age groups, and hence the high mortality in the absence of health care.

MSF started treatment of VL in Ler Hospital in 1989, but it was more than 1 year before security conditions allowed access to be established in Duar, 80 km north of Ler, in the epicentre of the epidemic. In the first year after arriving in Duar, almost 10 000 VL patients were treated in this location. Because of the complete absence of infrastructure, problematic logistical supply lines by air, and frequent evacuations due to insecurity, patients were treated under trees. Up to 2002, almost 23 000 cases have been treated in western Upper Nile in 10 different locations (Fig. 2). The numbers of cases treated during this period do not reflect the true incidence of the disease but rather access to treatment. In the early years of the intervention only a small proportion of the VL cases in western Upper Nile managed to reach treatment facilities. After the big epidemic, VL has become endemic in western Upper Nile, although sporadic localized outbreaks have been occurring.

An outbreak of VL occurred in eastern Upper Nile region in 1994, when food shortages had forced many people to forage for wild foods in the *Acacia/Balanites* forests, where they became infected. Many hundreds of people probably died before the outbreak was discovered, and before the security situation allowed effective access to affected populations by medical humanitarian aid (Seaman *et al.*, 1996). Since 1995 MSF has operated several VL treatment centres in rebel-controlled

areas in eastern Upper Nile, although access was often hampered due to insecurity and evacuations, or physical restrictions (unusable airstrips during the rainy season). In December 2001 MSF was granted access to the government-controlled town of Malakal and outlying villages along the Sobat River.

A new outbreak in eastern Upper Nile started in September 2002, with the number of cases more than doubling in comparison with previous years (Fig. 2).

#### Visceral leishmaniasis in northern Sudan

In 1996, MSF was requested by the Leishmania Research Group in Khartoum and the Ministry of Health in Gedaref State to assist in an emerging VL epidemic in Gedaref State, northern Sudan, close to the Ethiopian border. Visceral leishmaniasis had been reported in this endemic region since the early 1900s. Apart from localized studies (Zijlstra & El-Hassan, 2001), epidemiological data on VL in Gedaref State before 1996 are incomplete, and based mainly on cases reported from the 2 hospitals in Gedaref and Hawata. Poor reporting systems, limited treatment capacity in the hospitals, and (financially) limited accessibility of diagnostic and treatment services, have probably resulted in serious underreporting of VL in Gedaref State. Between 1980 and 1995 the number of reported cases never exceeded 1400 per year, and was < 100 per year between 1986 and 1991. Resumption of the civil war in 1983 may have negatively impacted on treatment capacities and health information systems in Sudan.

After establishment of an MSF VL treatment centre in Um-el-Kher, along the Rahad River in southern Gedaref State, the number of VL cases reporting to this centre increased dramatically, and a second treatment centre was established in the region in 1998. Between 1997 and 1999, more than 4000 cases were treated per year in the MSF centres (Fig. 2). Owing to the free-of-charge provision of all services, including food and accommodation for patients and carers, the MSF treatment centres greatly improved access to treatment.

Reported incidence data since 1996 show 2 subsequent epidemics in the region. The first epidemic occurred along the Rahad River basin, where VL incidence peaked in 1997 with 15 cases per 1000 per year. Reported incidence in the worst-affected local council area of Bandegeo (population ~27 000) was as high as 36/1000/year, and in several villages in 1997 the incidence of VL was > 50/1000/year. Later the epidemic spread to the Atbara River basin, reaching its

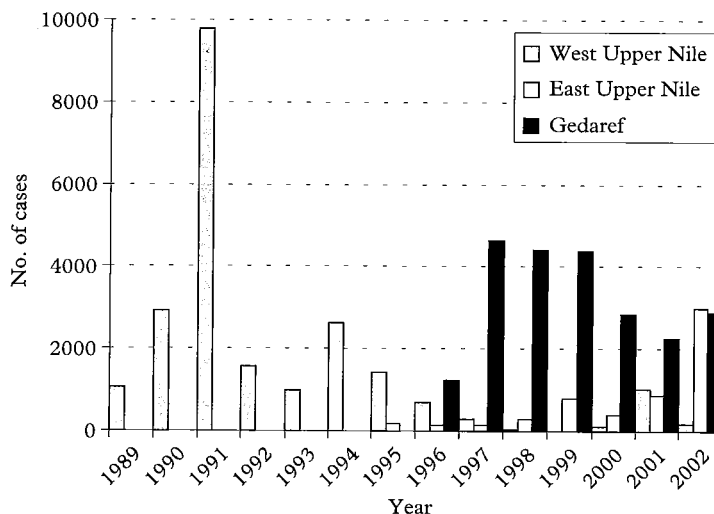


Fig. 2. Visceral leishmaniasis cases treated by MSF by year and by region in Sudan, 1989–2002.

peak in 1999 with an average VL incidence of 11/1000/year (Fig. 3).

In 2002, VL incidence in Gedaref State rose again, most markedly along the Rahad River, where the number of VL cases doubled from 2001 to 2002. This may be the start of a new epidemic cycle.

### Diagnosis

Diagnostic protocols for VL in the MSF clinics are based on clinical screening and laboratory confirmation. Because the clinical case definition of suspected VL (fever lasting > 2 weeks [with exclusion of malaria] in combination with either splenomegaly or lymphadenopathy) in Sudan has a prior-probability of ~50%, laboratory confirmation of the disease is required before starting treatment. Although demonstration of *Leishmania* amastigotes in stained lymph gland or spleen smears is the ultimate confirmation of the disease, parasitological screening of suspect cases in high prevalence situations and large numbers of patients is practically not feasible. Therefore, a serological direct agglutination test (DAT) is being used (Harith *et al.*, 1988). This test has shown an adequate sensitivity of ~95%, and a specificity of 85–95% when used in a 'borderline concept', i.e. patients with 'borderline' DAT titres (> 1:400 but < 1:6400) need to be parasitologically confirmed. This borderline concept is used in MSF projects, as a compromise to reduce the number of aspirations and to limit the aspirations to the group of patients with inconclusive DAT results.

These diagnostic protocols based on microscopy and DAT require laboratory services with well-trained laboratory technicians in the field, and are therefore not appropriate in all field conditions. In situations where field laboratories cannot be established (for reasons of security or capacity), transportation of blood filterpapers for DAT testing from peripheral units to centralized laboratories often results in unacceptable treatment delay. Simple, rapid, and cheap serological dipstick tests are urgently required. A recombinant antigen test (rK39) is available, and has shown good performance in Brazil and India (Badaro *et al.*, 1996; Sundar *et al.*, 1998). However, the test does not perform adequately in Sudanese VL (Zijlstra *et al.*, 2001; Veeken *et al.*, 2003). Therefore, further development of a rapid strip test for Sudanese VL is a priority.

### Treatment

The mainstay of treatment of VL is the pentavalent antimonial sodium stibogluconate (SSG) given at a

regimen of 20 mg/kg/d for 30 d. Until recently, MSF as well as the government services in Sudan were dependent on expensive branded SSG (Pentostam®, Glaxo Wellcome, London, UK), costing US\$120–200 per treatment. Several comparative clinical field trials, conducted by MSF in the late 1990s, proved that a generic version of SSG (Albert David Ltd, Calcutta, India) showed no difference in efficacy and tolerability, and was available at a much lower price of ~US\$20 per treatment (Veeken *et al.*, 2000; Moore *et al.*, 2001; Ritmeijer *et al.*, 2001). As a result of these studies, the generic version of SSG is now registered and used in Sudan, making treatment of VL more affordable.

### Treatment outcomes

From 1989 to 2002, MSF treated > 51 000 cases of primary VL, relapsed VL, and post-kala-azar dermal leishmaniasis (PKDL) in Sudan. Almost 43 000 primary VL patients were cured (cure rate 90.8%), and 3900 patients died during treatment (death rate 8.2%) (Table). Despite the relatively high cost of treatment, the cost-effectiveness of this humanitarian intervention is very favourable due to the high effectiveness of the treatment (the natural history of the disease is death, and treatment results in lifelong immunity) (Griekspoor *et al.*, 1999). Especially in southern Sudan, where even basically trained local staff are scarce, good treatment outcomes can be achieved only when diagnostic, treatment, and patient management procedures are governed by strict protocols and strictly supervised (MSF, 2003).

Improvement of care systems and protocols are still contributing to progress in quality of care provided, and over the past 6 years cure rates in primary VL are still increasing, and have now reached almost 95% (Fig. 4).

Analysis of risk factors for death indicate that both young age or old age are predictors of poor outcome. It is also not surprising that long duration of the illness, poor nutritional status, and anaemia are associated with higher death rates during treatment. The odds of death are clearly increased if the patient experiences at least one episode of vomiting or bleeding (usually epistaxis) during treatment. Currently, risk scoring systems are being developed and field-tested, in order to identify high-risk patients, who will be provided with more individualized care.

Although not yet documented in Africa, resistance to antimonials is common in India (Sundar *et al.*, 2000). When SSG is used alone, resistance will probably

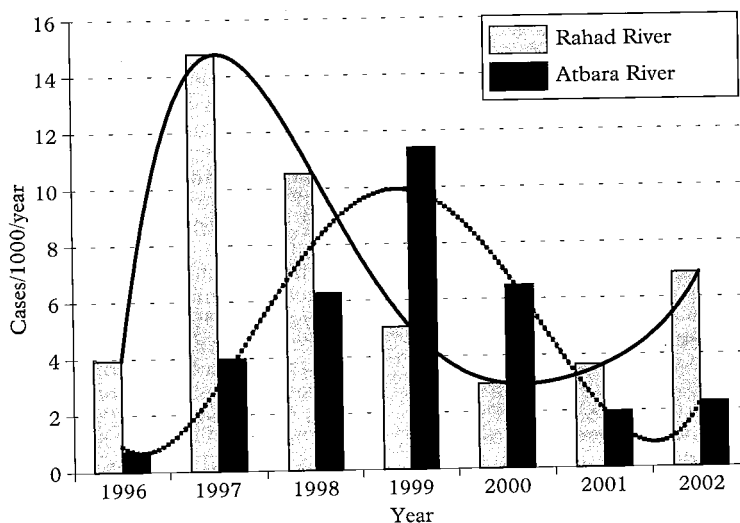


Fig. 3. Reported visceral leishmaniasis incidence in the Rahad and Atbara River basin areas, Gedaref State, Sudan, 1996–2002.

**Table. Visceral leishmaniasis cases treated and overall outcomes in MSF treatment centres in Sudan, 1989–2002**

	Northern Sudan	Southern Sudan	Total
Total admissions <sup>a</sup>	23 951	27 297	51 248
Primary VL admissions	21 951	25 406	47 357
Primary VL cured	20 354	22 620	42 974
Primary VL deaths	1 536	2 364	3 900
Overall cure rate (%) <sup>b</sup>	92.0	89.8	90.8
Overall death rate (%) <sup>b</sup>	6.9	9.4	8.2
Overall defaulter rate (%) <sup>b</sup>	1.2	0.8	1.0

<sup>a</sup>Includes visceral leishmaniasis (VL) relapses and post-kala-azar dermal leishmaniasis.

<sup>b</sup>Overall outcome rates are calculated using total exit numbers as the denominator.

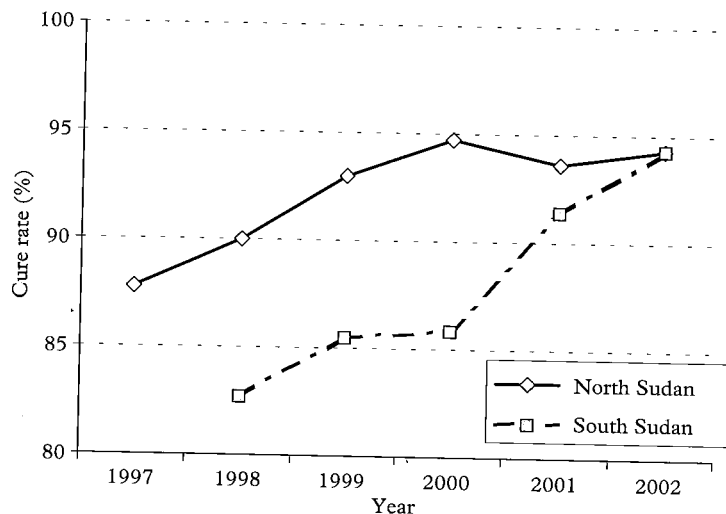


Fig. 4. Primary visceral leishmaniasis cure rates in MSF treatment centres in Sudan, 1997–2002.

emerge, even if it is used under strict adherence supervision. Therefore, resistance monitoring is necessary. An important indicator is the proportion of relapses among VL admissions. Although based on passive reporting, underreporting rates are not expected to differ over the years. Between 1997 and 2002 relapse admission rates have been stable at 3–4%, without an increasing trend. A second indicator for emerging resistance is the proportion of patients who still have a positive parasitological test-of-cure (TOC) by the end of 30 d SSG treatment. In Gedaref State positive TOC rates have been very stable at ~5% for the last 3 years, without an increasing trend. In Malakal, however, TOC positivity rate is high, and averaged 12.5% during 2002. Antimony-unresponsive primary VL is also more common in Malakal: 1.3% compared with < 0.1% in Um-el-Kher, Gedaref State. These strong indications of emerging resistance to SSG in Malakal are worrying, and possibly due to the availability of SSG in the private markets in and around Malakal before the MSF intervention, resulting in partial and subtherapeutic treatments. Because of the importance of Malakal as an intersection of travel and trading routes between northern and southern Sudan, resistance could spread rapidly to other endemic areas. MSF sees the provision of free treatment as the best way to eliminate the unregulated sale of SSG in markets.

#### Short-course combination therapy

There are several reasons why short-course combination therapies should be pursued. First, to prevent development of resistance, and protect the efficacy of current drugs. Second, to reduce the risk of disease outbreaks in immunocompromised VL patients in overcrowded treatment centres. Third, to improve

treatment outcomes with superior and less toxic drug combinations. Finally, to reduce hospitalization costs, both for providers and patients. Different short-course combinations are being considered, or are currently under evaluation by MSF.

**Paromomycin and SSG.** Paromomycin or aminosidine is an aminoglycoside antibiotic, which has been used successfully in treatment of relapses in southern Sudan over the last decade. In combination with SSG the duration of treatment can be reduced from 30 d with SSG alone to 17 d with a combination of SSG and paromomycin (Seaman *et al.*, 1993). The drug is, however, not yet licensed for treatment of VL. Under pressure from epidemic circumstances in eastern Upper Nile in September 2002, where the MSF treatment capacity was becoming unable to deal with the high case load of more than 400 VL admissions per month, and the subsequent risk of disease outbreaks in the treatment centres became critical, it was decided to start SSG/paromomycin combination therapy as first-line treatment. Reduction of the treatment course would significantly reduce the number of patients in the treatment centres. Preliminary comparison of outcomes among 577 patients receiving combination therapy to 1558 similar patients treated with SSG alone in previous years (1999–2001) indicates that the combination therapy is superior. Cure rates in the SSG/paromomycin group and the SSG group were 95.9% and 91.1%, respectively. The odds of death were 0.51 (95% CI 0.34–0.84) in the SSG/paromomycin group compared with the SSG monotherapy group.

**Liposomal amphotericin B and SSG.** Amphotericin B has a strong antileishmanial efficacy but, because of its toxicity, it is suitable for use only under hospital conditions. However, liposomal amphotericin B

(AmBisome®, Gilead, Foster City, CA, USA) is virtually free of toxicity, and can therefore be given in high doses to severely ill VL patients (Seaman *et al.*, 1995). Although liposomal amphotericin B is used in southern Sudan in patients who would not survive a treatment course with SSG, or in multiple relapses, the high cost of the drug (US\$840 for the treatment of an average patient, even at a preferential drug price for MSF) prohibits the more general use of this very effective and safe drug for VL. A combination of liposomal amphotericin B followed by SSG is currently under evaluation among patients whose admission assessment shows them to be at high risk of death. Our aim is to allow appropriate patients to benefit from liposomal amphotericin B whilst keeping costs under control.

*Miltefosine and SSG.* Miltefosine, an alkyl phospholipid, has been newly developed as an oral antileishmanial drug. Studies in India have shown that this oral drug is very effective for treating VL in both adults and children, even in patients with multiple previous treatment failures (Sundar *et al.*, 2002). Although licensed by WHO for treatment of VL, miltefosine has not been used yet in eastern African VL. Before investigating possible short-course combination regimens with SSG, MSF is planning to evaluate the efficacy and tolerability of miltefosine in East African VL in comparison with SSG, in a field evaluation trial in Ethiopia in 2003.

### Conclusions

Outbreaks of VL within the endemic area of Sudan remain very unpredictable—they are probably driven by environmental changes, such as floods, abnormal rainfall or famines—and during outbreaks the incidence of VL can be very high. Equally, access to affected populations in war-torn southern Sudan has been unpredictable, with changing patterns of warfare and insecurity in the region. In order to effectively respond to outbreaks, MSF takes a flexible approach, establishing treatment centres where the patients can be accessed.

Because treatment for VL can be provided only on the basis of laboratory confirmation, lack of diagnostics is a major obstacle to treatment, especially during epidemic situations. This requires the urgent development of simple and rapid technologies (e.g. serological strip tests), allowing reliable diagnosis under field conditions.

For treatment of VL there is a limited choice of effective, affordable drugs. Although research and development of new drugs for treatment of VL are required, results are not expected within the next 5–10 years. Introduction of combination therapies is urgently needed to prevent the further emergence and spread of resistance to antimonials, which are still the mainstay of VL treatment in eastern Africa.

### References

- Badaró, R., Benson, D., Eulálio, M. C., Freire, M., Cunha, S., Netto, E. M., Pedral-Sampaio, D., Madureira, C., Burns, J. M., Houghton, R. L., David, J. R. & Reed, S. G. (1996). rK39: a cloned antigen of *Leishmania chagasi* that predicts active visceral leishmaniasis (VL). *Journal of Infectious Diseases*, **173**, 758–761.
- De Beer, P., Harith, A. E., Van Grootheest, M. & Winkler, A. (1990). Outbreak of kala-azar in the Sudan. *Lancet*, **335**, 224.
- Griekspoor, A., Sondorp, E. & Vos, T. (1999). Cost-effectiveness analysis of humanitarian relief interventions: visceral leishmaniasis treatment in the Sudan. *Health Policy and Planning*, **14**, 70–76.
- Harith, A. E., Kolk, A. H., Leeuwenburg, J., Muigai, R., Huigen, E., Jelsma, T. & Kager, P. A. (1988). Improvement of a direct agglutination test for field studies of visceral leishmaniasis. *Journal of Clinical Microbiology*, **26**, 1321–1325.
- Moore, E., O'Flaherty, D., Heuvelmans, H., Seaman, J., Veeken, H., de Wit, S. & Davidson, R. N. (2001). A randomised comparison of generic sodium stibogluconate (sodium antimony gluconate, Albert David Ltd., Calcutta) and Pentostam (Glaxo Wellcome, London) for the treatment of visceral leishmaniasis in Kenya. *Bulletin of the World Health Organization*, **79**, 388–393.
- MSF (2002). *Violence, Health, and Access to Aid in Unity State/Western Upper Nile, Sudan*. Amsterdam: Médecins Sans Frontières.
- MSF (2003). *MSF Manual for the Diagnosis and Treatment of Visceral Leishmaniasis (Kala-Azar) Under Field Conditions*. Amsterdam: MSF-Holland.
- Perea, W. A., Moren, A., Ancelle, T. & Sondorp, E. (1989). Epidemic visceral leishmaniasis in southern Sudan. *Lancet*, **ii**, 1222–1223.
- Ritmeijer, K., Veeken, H., Melaku, Y., Leal, G., Amsalu, R., Seaman, J. & Davidson, R. N. (2001). Ethiopian visceral leishmaniasis: generic and proprietary sodium stibogluconate are equivalent; HIV co-infected patients have a poor outcome. *Transactions of the Royal Society of Tropical Medicine and Hygiene*, **95**, 668–672.
- Seaman, J., Price, D., Sondorp, E., Moody, A., Bryceson, A. D. M. & Davidson, R. N. (1993). Epidemic visceral leishmaniasis in Sudan: a randomised trial of aminosidine plus sodium stibogluconate versus sodium stibogluconate alone. *Journal of Infectious Diseases*, **168**, 715–720.
- Seaman, J., Boer, C., Wilkinson, R., de Jong, J., de Wilde, E., Sondorp, E. & Davidson, R. (1995). Liposomal amphotericin B (AmBisome) in the treatment of complicated kala-azar under field conditions. *Clinical Infectious Diseases*, **21**, 188–193.
- Seaman, J., Mercer, A. J. & Sondorp, E. (1996). The epidemic of visceral leishmaniasis in Western Upper Nile, southern Sudan: course and impact from 1984 to 1994. *International Journal of Epidemiology*, **25**, 862–871.
- Sundar, S., Reed, S. G., Singh, V. P., Kumar, P. C. & Murray, H. W. (1998). Rapid accurate field diagnosis of Indian visceral leishmaniasis. *Lancet*, **351**, 563–565.
- Sundar, S., More, D. K., Singh, M. K., Singh, V. P., Sharma, S., Makharia, A., Kumar, P. C. & Murray, H. W. (2000). Failure of pentavalent antimony in visceral leishmaniasis in India: report from the center of the Indian epidemic. *Clinical Infectious Diseases*, **31**, 1104–1107.
- Sundar, S., Jha, T. K., Thakur, C. P., Engel, J., Sindermann, H., Fischer, C., Junge, K., Bryceson, A. & Berman, J. (2002). Oral miltefosine for Indian visceral leishmaniasis. *New England Journal of Medicine*, **347**, 1739–1746.
- Thomson, M. C., Elnaïem, D. A., Ashford, R. W. & Conner, S. J. (1999). Towards a kala azar risk map for Sudan: mapping the potential distribution of *Phlebotomus orientalis* using digital data of environmental variables. *Tropical Medicine and International Health*, **4**, 105–113.
- Veeken, H., Ritmeijer, K., Seaman, J. & Davidson, R. (2000). A randomised comparison of branded sodium stibogluconate (Pentostam, Glaxo Wellcome, London) and generic sodium stibogluconate (Albert David Ltd., Calcutta) for the treatment of visceral leishmaniasis under field conditions in Sudan. *Tropical Medicine and International Health*, **5**, 312–317.
- Veeken, H., Ritmeijer, K., Seaman, J. & Davidson, R. N. (2003). Comparison of an rK39 dipstick rapid test with direct agglutination test and splenic aspiration for the diagnosis of kala-azar in Sudan. *Tropical Medicine and International Health*, **8**, 164–167.
- Zijlstra, E. E. & El-Hassan, A. M. (2001). Leishmaniasis in Sudan: 3. Visceral leishmaniasis. *Transactions of the Royal Society of Tropical Medicine and Hygiene*, **95**, supplement 1, S1/27–S1/58.
- Zijlstra, E. E., Siddig Ali, M., El-Hassan, A. M., El-Toum, I. A., Satti, M., Ghalib, H. W., Sondorp, E. & Winkler, A. (1991). Kala-azar in displaced people from southern Sudan: epidemiological, clinical and therapeutic findings. *Transactions of the Royal Society of Tropical Medicine and Hygiene*, **85**, 365–369.
- Zijlstra, E. E., Nur, Y., Desjeux, P., Khalil, E. A. G., El-Hassan, A. M. & Groen, J. (2001). Diagnosing visceral leishmaniasis with the recombinant K39 strip test; experience from Sudan. *Tropical Medicine and International Health*, **6**, 108–113.

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