BANCROFTIAN FILARIASIS IN EGYPT: VISUALIZATION OF ADULT WORMS AND SUBCLINICAL LYMPHATIC PATHOLOGY BY SCROTAL ULTRASOUND

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Abstract. The purpose of this study was to explore the value of scrotal ultrasound as a means of evaluating Bancroftian filariasis. Color Doppler ultrasound examinations were performed to look for subclinical hydroceles and motile adult filarial worms (dancing worms) in dilated lymphatics. Sixty-one male subjects from a filariasis-endemic area in Egypt were studied including 19 clinically normal microfilaria (MF) carriers (seven with dancing worms and eight with subclinical hydroceles), 13 MF-negative subjects with positive filarial antigen test results (three with dancing worms and seven with subclinical hydroceles), 22 exposed subjects with no MF and negative antigen test results (no dancing worms, four subclinical hydroceles), and seven subjects with clinical filariasis (no dancing worms, seven hydroceles). Thus, all men tested with clinical filariasis and most clinically normal subjects with either microfilaria or filarial antigenemia had abnormal ultrasound examination results. Ultrasound findings often changed after therapy with diethylcarbamazine, with disappearance of dancing worms and development of new scrotal calcifications or hydroceles. This study confirms the value of scrotal ultrasound as a means of noninvasively visualizing adult filarial worms and assessing subclinical lymphatic damage in Bancroftian filariasis.

Bancroftian filariasis is a deforming disease caused by the filarial nematode *Wuchereria bancrofti*. This parasite infects about 100 million people in many countries in the tropics and subtropics. Approximately 10–30% of those infected develop overt clinical disease (mainly hydroceles and/or lymphedema that can progress to elephantiasis). However, recent studies have shown that asymptomatic infections are often associated with subclinical disease including lymphatic damage demonstrable by lymphscintigraphy and nephritis manifested by proteinuria and hematuria. Amalar and others have recently reported visualization of adult *W. bancrofti* in scrota of infected men by ultrasound. They reported that adult filarial worms are often found in dilated lymphatics in the scrotum and showed that the location of worm nests in lymphatic vessels is highly stable over time. These investigators coined the term filarial dance sign to describe the ultrasound appearance of adult filariae. This technique has also been used with some success in India. The ability to observe adult worms in vivo provides an opportunity to directly observe effects of chemotherapy on the worms and the host. In addition, this technique provides a potential window for observing early hydroceles and for improving understanding of the pathogenesis of this disease.

Bancroftian filariasis is focally endemic in Egypt and prevalence rates are increasing in some areas. These infections are often clinically silent; lymphedema is uncommon, and hydroceles are the most common clinical manifestation of filariasis in Egypt. The present study was conducted with several objectives in mind. First, we sought to confirm the value of scrotal ultrasound as a means of observing living adult filarial worms in a different endemic area, namely Egypt. A second objective of the study was to document the extent of subclinical disease in asymptomatic men with microfilaria or with isolated filarial antigenemia compared with uninfected men residing in endemic and nonendemic areas in Egypt. The third objective of the study was to document changes in scrotal ultrasound findings following diethylcarbamazine (DEC) therapy.

SUBJECTS AND METHODS

Human subjects. Informed consent was obtained from all study subjects (and from parents of minors) for participation in this study. The study was approved by institutional review boards at Ain Shams University and at Barnes-Jewish Hospital in St. Louis.

Male subjects from filariasis-endemic villages in Qalubyia Governorate (approximately 35 km northeast of Cairo, Egypt) were selected to represent the following groups. Patients with clinical filariasis had clinically evident lymphedema or hydrocele. A hydrocele is an effusion of fluid in the scrotum between the two layers of the tunica vaginalis.

Subjects with fluid accumulations detectable by palpation were considered to have clinically detectable hydroceles. Microfilaria (MF) carriers had MF counts that ranged from 16 to 1,350/ml (median = 426/ml) and were free of symptoms or signs of clinical filariasis. Antigen-positive endemic normal subjects were amicrofilaric and clinically normal residents of filariasis-endemic villages with positive filarial antigen test results. Antigen-negative endemic normal subjects resided in endemic villages and had normal clinical examination results and negative test results for microfilaria and filarial antigenemia. Nonendemic normal subjects were Egyptian residents of Cairo with no evidence of hydrocele or lymphedema on physical examination and no history of residence in areas known to be endemic for filariasis. These subjects were referred for scrotal ultrasound because of infertility. The age of clinically normal subjects ranged from 10 to 60 years (median = 28.5 years), and these clinical groups did not differ significantly with respect to age. The clinical filariasis subjects were significantly older (median = 56 years, range = 31–62 years). Microfilariaemia was assessed by 5 μl membrane filtration of 1 ml venous blood collected between 9:00 PM and 1:00 AM. Filarial antigenemia was detected in plasma by monoclonal antibody-based ELISA as previously described.

Ultrasound. Ultrasound examination was performed between 1:00 PM and 5:00 PM by an experienced sonologist with a Toshiba 270 SSD Color Doppler ultrasound machine.
TABLE 1
Scrotal ultrasound findings in untreated subjects by clinical group

<table>
<thead>
<tr>
<th>Clinical group</th>
<th>Number</th>
<th>Normal</th>
<th>Motile worms</th>
<th>Hydrocele</th>
<th>Calcification</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>No. %</td>
<td>No. %</td>
<td>No. %</td>
<td>No. %</td>
</tr>
<tr>
<td>Nonendemic</td>
<td>20</td>
<td>18</td>
<td>90</td>
<td>0</td>
<td>2</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>10</td>
<td>0</td>
</tr>
<tr>
<td>Endemic normal</td>
<td>22</td>
<td>17</td>
<td>77</td>
<td>0</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>18</td>
<td>1</td>
</tr>
<tr>
<td>Clinical cases</td>
<td>7</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>7</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2</td>
</tr>
<tr>
<td>Antigenemia</td>
<td>13</td>
<td>5</td>
<td>39</td>
<td>3</td>
<td>7</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>54</td>
<td>0</td>
</tr>
<tr>
<td>Microfilaremia</td>
<td>19</td>
<td>4</td>
<td>21</td>
<td>7</td>
<td>37</td>
</tr>
<tr>
<td></td>
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<td>2</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>11</td>
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</tr>
</tbody>
</table>

(Toshiba America, Inc., New York, NY) and a 7.5-mHz linear phased array color Doppler transducer. The ultrasound examination was performed blindly; the sonologist did not know the infection status of the subjects. Subjects were examined in the supine position. The scrotum, inguinal areas, and upper thighs were examined after application of sonographic gel. Color Doppler ultrasound imaging was used to distinguish filarial movement in lymphatic vessels from flow of blood in veins and arteries. The diagnosis of hydrocele by ultrasound was based on the presence of fluid surrounding the testis and epididymis.10

RESULTS
Pretreatment. Results in untreated subjects are summarized in Table 1, and representative ultrasound images are shown in Figures 1 and 2. Dancing worms appeared as undulating linear structures within dilated scrotal lymphatics. Multiple worms were occasionally observed in a single dilated lymphatic nest. Two of 10 cases with dancing worms had bilateral involvement. Filarial hydroceles tended to occur inferior and posterior to the testis and often had internal septa. More than half of the hydroceles were bilateral. Only two subjects with hydroceles also had dancing worms.

Seven subjects with clinical filariasis were examined. Of these, five had clinically evident hydroceles, one had lymphedema of the leg, and another had elephantiasis of one leg and a history of surgery for hydrocele. All of these subjects had hydroceles detected by ultrasound, including those with lymphedema and no clinical hydrocele. Only two of the clinical cases had microfilaremia and positive filarial antigen test results. No dancing worms were seen in subjects with clinical filariasis.

Fifteen of 19 subjects with asymptomatic microfilaremia had dancing worms1 or subclinical hydroceles.2 Eight of 13 antigen-positive endemic normal subjects had dancing worms,1 subclinical hydroceles,2 or both findings.2 Hydroceles were more common in subjects with microfilaremia or isolated filarial antigenemia compared with antigen-negative endemic normal subjects (P = 0.030, by chi-square test). Ultrasound abnormalities (hydroceles or dancing worms) were more common in asymptomatic infected subjects (with or without microfilaremia) more than 18 years of age than in younger subjects (16 of 20 versus 7 of 12, respectively; P = 0.05, by Fisher’s exact test). There was no significant relationship between MF counts or antigen levels and the likelihood of seeing dancing worms by ultrasound.

No dancing worms were observed in 22 antigen-negative endemic normal subjects, but four had subclinical hydroceles. No dancing worms were seen in nonendemic normal subjects, but two had subclinical hydroceles. Echo-dense lesions that probably represent scrotal calcifications were observed in five of 61 subjects from the filariasis-endemic area, and this abnormality was not observed in nonendemic subjects. Probable calcifications were located in scrotal lymphatic vessels or along the spermatic cord and not in blood vessels, the scrotal wall, or the testes.

Post-treatment. Ultrasound examinations were repeated in seven MF carriers and four antigen-positive endemic nor-
mal subjects 1–2 years after DEC therapy (72 mg/kg over 12 days). Filarial dance signs disappeared in three of five cases that had been positive before treatment, and worm movement was decreased in the other two subjects. New, subclinical hydroceles developed in five of eight subjects who did not have hydroceles before treatment. New scrotal calcifications were observed in four of 11 men after treatment.

**DISCUSSION**

This study has confirmed the value of scrotal ultrasound as a means of visualizing living adult filarial worms in males with Bancroftian filariasis. Prior studies have not used color Doppler imaging, but we believe that this technology enhances the examination by facilitating differentiation of scrotal lymphatics from veins and arteries that have characteristic appearances with unidirectional flow. Adult filariae were observed in males with isolated filarial antigenemia or microfilaraemia but not in uninfected subjects from endemic or nonendemic areas. Most subjects with isolated filarial antigenemia had either dancing worms or subclinical hydroceles. These results provide further evidence that subjects with isolated filarial antigenemia are truly infected with adult filarial worms and that amicrofilaric infections in men are often associated with subclinical pathology detectable by ultrasound.

The frequency of positive filarial dance signs observed in subjects with microfilaraemia (37%) was slightly lower in this study than that reported by Amaral and others in Brazil (7 of 14 subjects). Subsequent studies from Brazil have reported visualization of adult worms by ultrasound in up to 80% of microfilaria carriers. Decreased sensitivity of the method in our series may be related to lower intensities of infection in our study subjects, younger subjects, decreased tropism of adult worms to the scrotum in Egypt related to parasite strain or unknown host factors, over-reading of ultrasound examinations in Brazil, or to under-reading (i.e., false-negative examination results) in this study. We believe the latter two explanations are unlikely since experienced sonologists usually have little difficulty identifying worms in scrotal lymphatics. Obviously, not all adult filarial worms
in infected men are located in scrotal lymphatics. The proportion of total worms that reside in this location is unknown and likely to be variable, but the lack of correlation between either MF number or antigen level (two measures of infection intensity) and the likelihood of detecting filarial worms by ultrasound suggests that the proportion is not high.

Motile filarial worms were not observed by ultrasound in men with clinical filariasis (lymphedema or hydrocele). All of these men resided in areas endemic for filariasis. Since none of the men with clinical filariasis had microfilaremia or positive filarial antigen test results, it is likely that they were not infected at the time that ultrasound was performed, although they presumably had infections in the past.

Prior reports have not emphasized the ultrasound findings of subclinical hydroceles and scrotal calcifications that were prominent in this study. Subclinical hydroceles were fairly common in all groups, but significantly more common in subjects with active infections (i.e., with microfilaremia or positive antigen test results). Adult filariae were usually not visualized in subjects with clinical or subclinical hydroceles. Subclinical hydroceles and scrotal calcifications seen in endemic normal men may reflect damage from past filarial infections. Two subclinical hydroceles were observed in non-endemic normal men. Although the prevalence of subclinical hydrocele in normal Egyptian men is not known, it may be less than the 10% observed in this series of men who were referred for examination because of infertility.

Filarial dance signs decreased or disappeared after DEC therapy in most subjects. This finding is consistent with prior reports that DEC has partial macrofilaricidal activity against *W. bancrofti*. Males sometimes develop scrotal pain and/ or nodules after DEC treatment of filariasis, and this is believed to be caused by inflammatory reactions to dying adult worms. Our ultrasound study has shown that DEC therapy is often associated with increases in subclinical pathology (nonpalpable hydroceles and scrotal calcifications) that are also probably related to the death of adult worms. In one of our cases, new bilateral hydroceles and scrotal calcifications present one year after treatment were completely resolved when the subject was reexamined one year later (two years after treatment). Additional study is needed to determine the long-term clinical significance of abnormalities observed by ultrasound after DEC treatment.

Acknowledgments: The assistance of the filariasis field research team of the Center for Research and Training on Vectors of Diseases, Ain Shams University is gratefully acknowledged.

Financial support: This work was supported in part by NIH grants AI-22488 and AI-35855.

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REFERENCES