EVALUATION OF SEVAFILACHEK IMMUNOASSAYS AND RAPID ICT-FILARIASIS TEST FOR DETECTION OF BANCROFTIAN FILARIASIS

R. Alli, S. Kulkarni, M. V. R. Reddy and B. C. Harinath

Department of Biochemistry & JB Tropical Disease Research Centre, MGIMS, Sevagram (Wardha)-442102, Maharashtra, India

ABSTRACT

A comparative analysis was made on the utility of SEVAFILACHEK-stick based immunoassays and commercially available ICT-filariasis test to detect active infection in different groups of bancroftian filariasis. The SEVAFILACHEK immunoassays were found to be useful to detect filarial infection in microfilaraemia and in a significant number of clinical filarial cases with acute, chronic and occult clinical manifestations. In the clinical cases, microfilariae are not usually detected in peripheral circulation. Employing SEVAFILACHEK assays 6 and 5 of the 7 samples of patients with chronic filarial disease, and 6 and 5 of 6 microfilaraemic cases gave positivity for filarial IgG antibodies and antigen respectively. Four of the 6 occult filarial samples were positive for antibodies and antigen. Filarial antigen was detected by ICT-filariasis test in blood samples of all the 6 microfilaraemic cases, 1 chronic filarial and 2 occult filarial samples. The main advantage of ICT assay is its rapid format and convenience for field use.

KEY WORDS: Bancroftian filariasis, antigen, antibody, immunoassays, rapid diagnostics.

INTRODUCTION

With about 120 million people infected globally (1), lymphatic filariasis caused mainly by *Wuchereria bancrofti* and *Brugia malayi* is still a major public health problem in many parts of tropics and subtropics. Those who reside in an endemic area for filariasis exhibit variety of clinical presentations (2, 3). While some individuals merely carry parasite (with microfilariae detectable in peripheral circulation during night hours) without having any overt disease, a few individuals suffer from acute, chronic and occult manifestations of the disease. A significant number of people considered to be 'endemic normals' are free from clinical disease and do not show microfilariae (mf) in night blood. The individual diagnosis of filariasis based on clinical examination or demonstration of mf in night

blood smear is neither useful for all stages of infection nor convenient for field use. Hence one of the essential requirements for effective control of filariasis based on the detection and treatment with antifilarial drug like diethylcarbamazine citrate, (DEC) is a rapid and reliable diagnostic test. Such a test will also help to effectively follow-up the clinical cases during therapy for clinical relief and cure.

In earlier communications from this laboratory we reported on the development of 'SEVAFILACHEK' assays that included two stick based immunoassays for the detection of active infection of bancroftian filariasis (4,5). One of these two assays is meant to detect filarial IgG antibodies in blood samples by indirect ELISA against *B. malayi* mf excretory-secretory (ES) antigen (4). The other assay detects circulating filarial antigen by inhibition ELISA using IgG fraction of clinical filarial serum immunoglobulins and *B. malayi* mf ES antigen conjugated to enzyme penicillinase as probe (5). The ICT filariasis test is a rapid-format test kit developed by ICT Diagnostics (Amrad, ICT, Australia) based on detection of

Author for correspondence:

Dr. M.V.R. Reddy
Additional Professor, at above address
Tel : 091-7152 84341 to 84355 Extn : 303
Telefax : (07152) 84038
E-mail : jbtddrc@nagpur.dot.net.in

circulating filarial antigen (6) using anti *Dirofilaria immitis* monoclonal antibody AD 12.1. In the present study we compared the 'SEVAFILACHEK' immunoassays with the commercially available ICT filariasis test kit to detect the active filarial infection in different groups of individuals across the wide spectrum of the disease.

MATERIALS AND METHODS

A total of 25 blood samples of individuals residing in Sevagram and surrounding villages, which is a non-costal endemic region for bancroftian filariasis were screened (table 1). The mf were checked in thick smears of 20 µl night blood samples collected during 20-22 hrs. The clinical filarial samples were from those who attended the general, surgical or paediatric out patient divisions of this institute with acute, chronic (with oedema or hydroceles for 2 to 5 years, several acute episodes of lymphangitis or epididymo-orchitis) or occult filarial manifestations (tropical pulmonary eosinophilia, bronchial asthma or polyarthritis).

ICT filariasis kit was a kind gift from Medical Diagnostics, Godrej Soaps Ltd. Chennai and the kit was used as per instructions from manufacturer. 'SEVAFILACHEK' immunoassays were used as described earlier (4,5).

RESULTS AND DISCUSSION

The results as summarized in Table 1. showed that all the three tests were useful to detect

microfilaraemic cases. SEVAFILACHEK assays showed all the six microfilaraemic samples as positive for filarial IgG antibodies and five samples as positive for filarial antigen. Filarial antigen was detected in all the six samples by using ICT kit. Earlier reports from our laboratory on SEVAFILACHEK immunoassays (4,7) showed the filarial IgG antibody assay giving a sensitivity of 80% for detection of microfilaraemic cases and a specificity of 90%. The detection of filarial antigen by inhibition ELISA had shown a sensitivity of 86% (for mf positive cases) and a specificity of 83%. However the results were quite different with the 7 samples of chronic filarial cases. While SEVAFILACHEK assays showed 6 of the 7 samples as positive for antibody and 5 samples as positive for antigen, the ICT-kit showed only one sample as positive. These results of ICT test kit are quite consistent with what was reported earlier in the extensive studies conducted on this test system. ICT test system was reported to detect filarial antigen in most of the microfilaraemic patients and most of the amicrofilaraemic patients with clinical filariasis showed negative antigen results (8).

Four of the 6 occult filarial samples were positive for filarial antigen and / or IgG antibodies using SEVAFILACHEK assays (table 2). As we had reported earlier both these 'SEVAFILACHEK' assays were found to be useful to detect active filarial infection in patients with variety of clinical presentations (9). A significantly high percentage of patients in paediatric age group were positive for filarial antibody and / or antigen and the assays were effectively used to

Table 1. Comparative evaluation of SEVAFILACHEK immunoassays and ICT-test for diagnosis of bancroftian filariasis

Group	No. of samples screened	Number positive using		
		ICT kit (Filarial Antigen)	SEVAFILACHEK EIAs	
			Indirect stick ELISA* (Filarial Antibody)	Inhibition stick ELISA* (Filarial Antigen)
Endemic Normals	6	2	1	1
Microfilaraemia	6	6	6	5
Chronic filariasis	7	1	6	5
Occult filariasis	6	2	4	4

* Sera showing filarial IgG antibody/ antigen titre of ≥ 300

Table 2. Analysis of blood sample of Acute, Chronic and Occult filariasis using ICT kit and SEVAFILACHEK EIAs.

S. No.	Clinical presentation (Age in years/ Sex*)	ICT test	Indirect stick ELISA	Inhibition stick ELISA
1	Epididymoorchitis (16/ M)	-	+	+
2	Oedema on Right leg (14/ F)	-	+	+
3	Pitting Oedema on Right hand (25/ M)	-	+	+
4	Hydrocele (23/ M)	-	+	+
5	Pitting Oedema on left lower limb (66/ M)	+	-	+
6	Pitting Oedema with pain on leg (22/ F)	-	+	-
7	Hydrocele (66/ M)	-	+	-
8	Tropical Pulmonary Eosinophilia (55/ F)	-	+	+
9	Tropical Pulmonary Eosinophilia (53/ F)	-	+	+
10	Tropical Pulmonary Eosinophilia (6/ M)	+	+	+
11	Breathlessness & Pain in chest (10/ F)	-	-	-
12	Bronchial Asthma (10/ M)	+	-	+
13	Polyarthritis (35/ F)	-	+	-

* M = male; F = Female

manage the clinical morbidity in such cases (10). Two of the occult filarial cases were positive for filarial antigen with ICT-kit. Both these were children, one having tropical pulmonary eosinophilia and the other suffering from bronchial asthma (table 2.).

Of the 6 samples from endemic normal individuals one was positive by SEVAFILACHEK assays whereas 2 were positive for filarial antigen by ICT-kit. Re-examination of their night blood samples by nucleopore membrane filtration technique (using 1 ml blood) did not show microfilariae. Weil *et al* (11) had shown that some sera of this group contain filarial antigen and demonstrated that antigenemia in these people is not an artifact but is an indicative of an active infection.

These results suggest that the ICT is a rapid-format antigen test and quite convenient to

detect microfilaraemia in field conditions. The SEVAFILACHEK immunoassays on the other hand are useful to detect active filarial infection as well as clinical disease cases associated with filariasis attending the hospital and thus help in the effective management of clinical morbidity and preventing recurrence in such cases in an endemic area.

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