USEFULNESS OF NEWLY ACQUIRED COMMERCIAL DENGUE NSI ANTIGEN TEST KIT FOR EARLY DETECTION OF DENGUE IN A GENERAL PRACTICE CLINIC IN BATU GAJAH, PERAK

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ABSTRACT

The non-structural protein 1 (NS1) of the dengue viral genome has been found useful for diagnosis of acute dengue infections, being detected from one day and up to 18 days post onset of symptoms (1). Rapid Dengue NS1 antigen test was made available in commercial test kits for private clinics in Perak in 2014. This study aimed to determine the use usefulness of dengue NS1 test kits in suspected dengue cases in a general practice clinic in Perak and the period in which positive test results were obtained after the onset of symptoms. Clinic records of all suspected dengue cases seen in the ten months from October 2014 to July 2015 were traced. Patients' demographic characteristics, presenting symptoms and the use of One Step Dengue NS1 Antigen test (Avo Diagnostics) were analyzed using SPSS version 17. Seventy one suspected dengue fever cases were seen in the study period, the highest number in July 2015 (25%). Majority were Chinese (80%), female (63%), and aged 40 years and above (51%). Most patients presented with fever (99%), body-ache (51%), gastrointestinal symptoms (51%) and headache (44%). Dengue NS1 antigen test was done in 94% of the suspected cases with 61% testing positive. The majority of the cases presenting one day to five days after onset of symptoms tested positive i.e. 59% (1 day), 90% (2 days), 54% (3-4 days) and 89% (5 days).

Using dengue NS1 antigen test, positive results can be obtained as early as one day after the onset of symptoms. This test is a useful tool to aid primary care physicians detect dengue fever early.

Key words: Dengue NSI Antigen test, early detection, dengue fever, general practice, Primary care physicians

INTRODUCTION

Dengue fever caused by dengue virus is a mosquito borne flavivirus, endemic in more than 100 countries including Malaysia (1). According to the study by Ang KT et al. 83.9% had sought medical consultation at primary care clinics prior to admission to hospital with 68.7% been seen on two or more occasions in primary care (2). The mean time of seeking treatment at primary care was 1.4 days of fever compared to 4.9 days to admission (2). Therefore primary care physicians play an important role in early recognition and management of dengue fever.

Patients often present to primary care within the first few days after onset of symptoms such as fever, headache, and body ache and are very anxious to know if they have dengue. But with relatively undifferentiated symptoms, these symptoms can be due to other causes. In primary dengue infection, IgM antibody is detected 5 days after onset of symptoms and IgG antibodies after 1 week. Secondary infection IgG antibody rises rapidly but the IgM antibody response is more variable (3). Therefore using serology tests alone, the primary care physician is unable to confirm dengue infection in first few days after onset of symptoms.

Non-structural protein 1 (NS1) of the dengue viral genome has been shown to be useful as a tool for diagnosis of acute dengue infections (3). Dengue NS1 antigen has been detected in the serum of infected patients as early as one day and up to 18 days post onset of symptoms. A positive result confirms diagnosis (4).

This test was first available commercially as Rapid dengue NS1 test kits for private clinics in Perak in 2014. The objectives of this study was to determine the use of Dengue NS1 test kits in suspected dengue cases in a general practice clinic in Batu Gajah, Perak after the clinic acquired the test kits and to determine the period in which positive test results were obtained after the onset of symptoms

METHODS:

Clinic records of all suspected dengue cases seen between October 2014 and July 2015 were traced. Just prior to the study period mentioned, the general practice clinic, situated in Batu Gajah, Kinta District, Perak acquired the Dengue NS1 Antigen test kits. The study period was chosen to start from the time the clinic acquired the test kits and was stopped ten months later in July 2015 to meet the dateline for abstract submission for oral presentation of its results at the Second International Conference on Tropical Medicine and Infectious Diseases held from 29/9/2015-1/10/15 in Ipoh, Perak. The patients' demographic characteristics, presenting symptoms and the use of one step Dengue NS1 Antigen test (Avo Diagnostics) (5) were compiled and analysed using SPSS version 17. The NS1 Antigen test used immunochromatographic assay method, the monoclonal antibody specific to Dengue virus NS1 antigen for accurate determination of Dengue virus infection. The test kit specified a sensitivity of 97.9% and specificity of 99 %(5). If blood sample was infected with dengue virus, a visible line appeared in test region on the membrane. The absence of coloured band in test region was a negative test result.

In January 2015, the clinic also acquired the Combo test (for NS1, IgG, IgM). This was used on some suspected dengue cases during the study period, the results were also noted and compiled in this study. As the Combo test was more expensive than the NS1 Antigen test, it was used only for those presenting 5-6 days or more after onset of symptoms or when a secondary dengue infection was suspected and if the patient could afford to pay for it.

RESULTS

Seventy one suspected dengue cases were

seen in the study period. Details of the demographic characteristics of these patients are listed in Table I. Majority were Chinese (80.3%), female (63%) and aged 40 years and above (50.8%).

Analysis of the suspected dengue cases by month, showed that the majority of the cases presented in the months of July 2015 (25%) and in November to December 2014 (35%), coinciding with the peak period of dengue cases throughout the country.

Ninety nine percent of the patients presented with fever, followed by musculoskeletal and gastrointestinal symptoms (refer Table 1 for details).

Out of the 71 suspected dengue cases, 67 patients took the NS1 Antigen test (94%). Four patients did not take the test, including one 13 year old who refused to take the test and the family members of three other patients (a child, teenager and elderly patient) requested referral to the nearby government hospital.

Table II shows the duration of onset of symptoms at time of presentation to the clinic and NSI Antigen test results. Among patients who presented on the first day after onset of symptoms 57% tested positive for NS1 Antigen. This rose to 90% for cases who presented at 2 days, 57% and 89% at 3-4 and 5 days after onset of symptoms (refer Table II for details). In two cases, the duration of onset of symptoms were not recorded.

Table III shows the results of the 11 suspected dengue cases where the Combo test was used. Again NS1 Antigen was positive in 3 out 4 cases where onset of symptoms were equal or less than 3 days.

DISCUSSION

In the review article on "Managing Dengue in Primary Care. A Practical Approach" published in Malaysian Family Physician journal, 2014, Lum et al emphasised the need to monitor patients through the febrile phase where the symptoms were undifferentiated to the next phase, the critical phase, where the fever dissipated but danger signs and symptoms appeared (6). However at the time of publication of the review article, nothing more in terms of investigation, except for a full blood count, could be offered in the first 72 hours in the febrile phase. A full blood count and differential count was recommended in centres where this test could be done to help to differentiate a viral from bacterial infection and to provide a baseline reading to monitor changes in haematocrit and platelet count during the critical phase.

In 2011, an article, published on the "Use of Dengue NS1 antigen for early diagnosis of dengue virus infection" by Fauziah Md Kassim et al from the Institute of Medical Research, Kuala Lumpur, Malaysia, recommended the use of Dengue NSI antigen to compliment the current antibody tests to increase the diagnostic efficiency for early diagnosis of dengue infection(7). However such test kits were not available commercially until 2014 for Dengue NS1 Antigen and 2015 for Combo test locally.

In this study, the majority of the patients presented with fever (99%), body-ache (51%), gastrointestinal symptoms (51%) & headache (44%). These were all nonspecific symptoms that could be attributed to other causes. However the Dengue NS1 test done on 94% of the suspected cases, showed 61% which tested positive during the study period. The majority who tested positive presented one day to five days after onset of symptoms. Positive results for dengue was obtained as early as one day after onset of symptoms in suspected cases, enabling the consulting primary care physician to detect dengue infection early and initiate management.

The Combo test had the advantage of testing for both NS1 antigen and IgG and IGM antibodies. It helped to differentiate between primary and secondary dengue infections or a previous infection. Since the Combo test was more expensive than the NS1 Antigen test, for private patients who could not afford it, the NS1 antigen test was still be very useful for primary care

physicians to detect dengue and initiate management early.

Pang et al in their study published in 2017, to assess changes in knowledge, attitude and practices on dengue diagnosis and management after the largest dengue epidemic in Singapore found a significant increase in the use of dengue diagnostic tests (combination of Dengue NSI plus IgM/IgG antibody detection), associated with a significant reduction in referral of dengue patients to hospital and a significant increase in frequency of clinic follow-ups (8). It was highlighted that the availability of the combination dengue diagnostic tests improved management. Prior to the Singapore dengue epidemic in 2013, dengue serology (IgM/IgG) test was used. In 2014, the combination dengue test was made available in Singapore. It was also found that both IgM/IgG serology and Dengue NS1 assay test were also more frequently used after the epidemic both in private and public clinics in Singapore(8).

This study was done during the period when Dengue NS1 antigen test kit was first introduced commercially in Malaysia and subsequently during the course of the study, the Combo test was made available, so most private general practice clinics in the community around where this study was done had not acquired the test kits. The public primary care clinics were just starting to acquire these Dengue NSI antigen and Combo test kits.

Dengue fever is endemic in Malaysia just like in Singapore. The majority of patients present first to general practice / primary care. Delay in detection and management can produce adverse outcomes. This study shows that using Dengue NS1 Antigen test in general practice allows for detection of dengue as early as 1-2 days of onset of symptoms. The test is easy to do in the clinic, yields immediate results and acceptable to the majority of patients, and can be done in the clinic itself at a reasonable cost and acceptable to the patients.

The limitation of this study is that it was done in one private general practice clinic with a small sample size. Also some data such as duration of onset of symptoms were not recorded in two cases. Therefore the authors suggest that a larger study like the one in Singapore to be conducted to look into the impact of the use of such test kits on the management of dengue fever in both private and public primary care clinics in Malaysia.

ACKNOWLEDGEMENT

The authors would like to thank the clinic staff who helped in tracing all patients' records.

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Table I. Demographic characteristics and presenting complaints of patients with suspected dengue fever (n=71)

Demographic characteristics / presenting complaints	Number (Percentage)
Gender	
i) Male	26 (36.6)
ii) Female	45 (63.4)
Ethnic group	
i) Malay	3 (4.2)
ii) Chinese	57 (80.3)
iii) Indian	10 (14.1)
iv) Others	1 (1.4)
Age group (years)	
i) 19 and below	22 (31.0)
ii) 20-39	13 (18.3)
iii) 40-59	18 (25.4)
iv) 60 and above	18 (25.4)
Presenting complaints	
i) fever	70 (98.6)
ii) musculoskeletal (bodyache, back/joint pains)	39 (54.9)
iii) gastrointestinal	36 (50.7)
iv) headache	31 (43.7)
v) rashes/flushing	22 (31.0)
vi) respiratory symptoms	21 (29.6)
vii) malaise	14 (19.7)

Table II. Onset of symptoms versus Dengue NS1 result

Onset of symptoms prior to presentation to clinic	**No of cases tested NS1 Antigen positive**	**No of cases tested NS1 Antigen negative**	**Refused NS1 / not done**	**Total no of cases**	**% tested positive to NS1 antigen**
< 1 day*	0	1	0/0	1	0%
1 day#	8	5	1/0	14	57%
2 days	9	1	0/0	10	90%
3 to 4 days	13	9	0/1	23	57%
5 days	8	1	0/0	9	89%
6 days	0	3	0/1	4	0%
7 days	3	2	0/1	6	50%
10 days	1	1	0/0	2	50%
Total	42	24	4	69	61%
Duration of onset not recorded	*1*	*1*	*0/0*	*2*	
Total	43	25	4	71	61%

Note: * <1 day means patient presented on the same day of onset of fever.
1 day means patient presented to clinic after one day of onset of fever

Table III. Suspected dengue cases using Combo test

Case number	Onset of symptoms prior to presentation	No of cases	NS1	IgG	IgM
1. 2. 3. 4.	<=3 days	4	Positive Positive Positive Negative	Positive Negative Negative Positive	Negative Negative Negative Negative
5. 6.	4-5 days	2	Negative Negative	Negative Positive	Negative Positive
7. 8. 9. 10. 11.	6 to 10 days	5	Negative Negative Negative Positive Negative	Positive Positive Positive Positive Positive	Positive Positive Positive Negative Positive