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Full Length Research Paper

Outbreak of dengue fever in Ouagadougou, Burkina Faso, 2013

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From October 18 to November 8, 2013, two medical centers in the central region of Burkina Faso, received 43 patients with atypical symptoms evoking Dengue Fever (DF). A total of 43 available sera were screened for IgM and IgG antibodies against dengue virus (DENV), using rapid tests of SD BIOLINE Dengue IgG/IgM (SD Standard Diagnostics, Korea) at the Bacteriology and Virology Laboratory of the Teaching Hospital, Centre Hospitalier Universitaire Yalgado Ouédraogo (CHU-YO) in Ouagadougou. The molecular detection of DENV was performed at the Influenza National Reference Laboratory of Burkina Faso, using CDC DENV-1-4 Real-Time RT-PCR (rRT-PCR) reagents, primers and protocols. Seven of 43 sera (16.3%) were positive for IgM/IgG, while 21/43 (48.8%) sera were positive by rRT-PCR. DENV 3 subtype was identified in all positive specimens. The rapid test had a good specificity (95.4%), but a very low sensitivity (28.5%) in comparison to rRT-PCR testing. Except a missing age for one patient and one 17-years old patient, the other 19/21 (90.4%) dengue confirmed cases were from adult patients over 20 years. Our results identified DENV 3 subtype as the etiologic agent of occurred outbreak in Ouagadougou in 2013 and indicated the essential contribution of rRT-PCR for DF detection.

Key words: Dengue fever, outbreak, serologic diagnosis, molecular diagnosis, Burkina Faso.

INTRODUCTION

Dengue fever (DF) is a mosquito-borne viral disease caused by four serologically distinct, but closely related subtypes of dengue virus (DENVs 1, 2, 3 and 4) belonging to the *Flaviviridae* family (Yauch and Shresta, 2014; Zhao et al., 2014). DENVs are transmitted from human to human by anthropophilic *Aedes* mosquitoes, mainly *Aedes aegypti* and *Aedes albopictus* (Murray et al., 2013). DF is now a worldwide concern and it is found in tropical and sub-tropical areas, mostly in urban and semi-urban areas and 3.6 to 4.0 billion people are at risk of DF globally (WHO, 2009; Murray et al., 2013). DF has increased more than 30-fold over the last five decades (WHO, 2009) and the disease is endemic in 128 countries (Guha-Sapir, 2005). According to several reports (Fagbami et al., 1997; Murray et al., 2013), 390 million DENV infections are estimated to occur per year; over three times more than previous estimates suggested by the World Health Organization (World Health Organization, 2009). There is no specific treatment

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ABBREVIATIONS: Dengue fever, DF; Dengue virus, DENV.

against DF. Despite progress with the development and clinical evaluation of vaccines against DENV infection, no such vaccine is on the market yet (Gonzalez et al., 1985; Suzarte et al., 2014). The occurrence of the novel subtype, DENV 5 in 2013 (Mamani et al., 2014) increased the difficulty of setting up of effective vaccines against the disease. Thus, the crucial control of the populations of DENV vector mosquitoes, especially *Aedes aegypti* and *Aedes albopictus*, to limit their spreading to new regions is not yet efficient (WHO/SEARO, 2011; Murray et al., 2013). Over the last decade, the geographical distribution of DF has included new countries and more rural areas, making it the most rapidly expanding arboviral disease in the world (WHO, 2009; Were, 2012). In some countries of sub-Saharan Africa, the circulation of the four subtypes of dengue virus (DENVs 1, 2, 3 and 4) has been reported (Yauch et al., 2014).

In Burkina Faso, data concerning the burden of DENVs have been poorly explored. Only serological data in both urban and rural areas among pregnant women and blood donors (Collenberg et al*.,* 2006) and scarce cases of DENV 2 from foreigner travelers for 1980-1986 periods were reported (Roberts et al., 1993; Ridde et al*.,* 2014). The lacking data on emerging DENV infection, in the country, somehow in misdiagnosing the etiology of the fever is commonly attributed to malaria without any laboratory confirmation. It is understandable that in malaria endemic country, artemisinin-based combination therapy is the first line treatment of suspected fever. However in 2013, about 50 patients with clinical symptoms attributable to malaria did not respond to (ACT). We therefore suspect that this outbreak may be caused by viral infection including DENV, poorly reported in Ouagadougou. In 2013, atypical cases of fever and other symptoms related to DF were observed in patients in central region. This study aimed to establish the etiology of all suspected cases of DF in the central region of the country in 2013.

MATERIALS AND METHODS

Outbreak history

From October 18 to November 8, the teaching hospital "Centre Hospitalier Universitaire Yalgado Ouedraogo" (CHU-YO) and the medical centre "Clinique Médicale des Opportunités" (CMO) in Ouagadougou, received 43 patients with atypical symptoms: presence of fever and two or more of the following symptoms including retro-orbital or ocular pain, headache, rash, myalgia, arthralgia, or hemorrhagic manifestations. Without laboratory testing, physicians and nursing staff strongly suspected malaria, and in certain cases they started treatment against malaria. These treatments against malaria often encountered failures.

In Burkina Faso, malaria is endemic, above all, in rainy season (from May to September) and sporadic cases in dry season (from October to April). However, in 2013, the large majority of treated cases encountered failures despite the negative results to malaria tests (Giemsa-stained thick blood smear and rapid diagnostic test, RDT). This situation motivated physicians and nursing staff to investigate other diseases with similar symptomatology as malaria and dengue fever.

Study sites

The study was carried out in two medical structures in Ouagadougou, the capital city of Burkina Faso, located in the central region (Figure 1): the teaching hospital, CHU-YO and a private medical centre, "Clinique Médicale des Opportunités".

Blood samples collection

A blood sample (4 to 8 mL) was collected by venipuncture into a Vacutainer tube (Becton Dickinson, Plymouth, UK) for each clinically dengue suspected case. The samples collected in the private medical centre, CMO were transported in cool-system containing ice-boxes to the Bacteriology and Virology Laboratory of the CHU-YO, within six hours maximum. All the blood samples collected were centrifuged and serum from each patient was stored in two vials at - 80°C until their analysis for the detection of DENVs.

Serologic diagnosis

Sera available from a total of 43 patients were screened for IgM and IgG antibodies to DENV using rapid tests SD BIOLINE Dengue IgG/IgM (SD Standard Diagnostics, Korea) at the Virology Laboratory of the CHU-YO. The SD BIOLINE Dengue rapid test was performed according to the manufacturer recommendations.

Test principle

SD BIOLINE Dengue IgG/IgM test is a solid phase *in vitro* immunochromatographic assay designated to simultaneously detect and differentiate IgG and IgM antibodies against dengue viruses in human serum, plasma or whole blood. This test uses a mixture or recombinant dengue envelope proteins to detect all four dengue serotypes without further discrimination. SD BIOLINE Dengue IgG/IgM test device has pre-coated lines, "G" (dengue IgG test line), "M" (dengue IgM test line) and "C" (control line) on the surface of the membrane. All three lines in the result window are not visible before applying any samples. The control line is used for procedural control. Control line should always appear if the test procedure is performed properly and the test reagents of control line are working. As such, purple "G" and "M" lines will be visible in the result

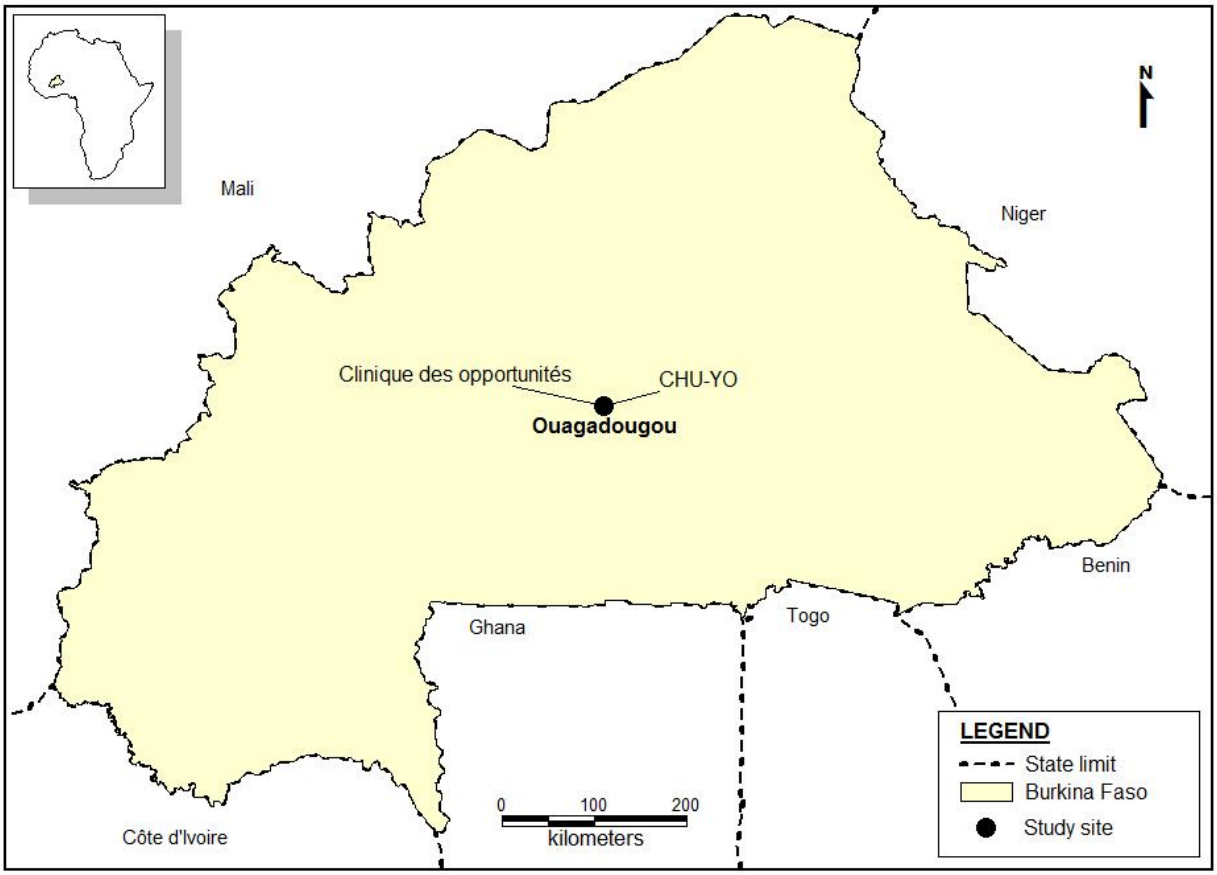

Figure 1. Medical structures (sites) which received the suspected cases of dengue in Ouagadougou, 2013: Clinique des Opportunités, Centre Hospitalier Universitaire Yalgado Ouedraogo (CHU-YO).

window if there are enough IgG and/or IgM antibodies against DENV in the sample.

Test procedure

Using micropipette, 10 µL of serum was added into the sample well marked "S". Then 3 or 4 drops of assay diluent were put into the diluent round shaped well. The test results were read in 15-20 min.

Test results interpretation

Five different cases were distinguished: (i) when two pink lines "C" and "M" appeared in the result window, it was positive for IgM even if "M" line is weak; in this case it was primary dengue infection; (ii) when two pink lines "C" and "G" appeared in the result window, it was positive for IgG even if "G" line is weak; in this case it was secondary or past dengue infection; (iii) when three pink lines "C", "M" and "G" appeared in the result window, it was positive for IgM and IgG; in this case it was late primary or early secondary dengue infection; (iv) when only one pink line "C" appeared in the result window at right, it was a negative case of dengue; (v) when no control "C" line was observed in the result window, it is invalid testing; it is recommended to re-test this specimen.

Molecular diagnosis

The molecular diagnosis was performed at the Influenza National Reference Laboratory, Institute of Research in Health Sciences, Bobo-Dioulasso, Burkina Faso.

RNA extraction

RNA was extracted from 140 µL serum using the QIAamp Viral RNA Mini Kit (Qiagen, Venlo, The Netherlands) according to the manufacturer's instructions. We stored extracted RNA at - 20°C if PCR was to be done within 24 h; otherwise, we kept the remaining RNA at -80°C.

Real-time RT-PCR for dengue virus detection

Dengue virus detection was performed using CDC

Table 1. Interpretation of CDC DENV-1-4 Real-Time RT-PCR assay.

DENV-1	DENV-2	DENV-3	DENV-4	RP Target	Result
+	-	-	-	±	Positive DENV-1 detection*
-	+	-	-	±	Positive DENV-2 detection*
-	-	+	-	±	Positive DENV-3 detection*
-	-	-	+	±	Positive DENV-4 detection*
-	-	-	-	+	Negative for DENV, result does not preclude infection
-	-	-	-	-	Inconclusive test, likely poor extraction or sample quality**

* If sample is positive for 2 serotypes, repeat test is done as indicated above. If sample is repetitively reactive for both serotypes, the result is indicative of a dual infection and should be confirmed by the CDC-Dengue Branch.
**When an inconclusive result is obtained, re-extract the specimen and test the newly extracted RNA (recommended) or re-test the extracted RNA.

DENV-1-4 Real-Time RT-PCR (rRT-PCR) reagents, primers and protocols, which can detect DENV serotypes 1, 2, 3 or 4 RNA from human serum. For that, we used a 20 µL volume of the Master mix containing 5 µL RNA, 5.55 µL nuclease free water, 12.5 µL 2XPCR Master Mix, 0.45 µL DENV probe, 0.5 µL SuperScript™ III, 0.5 µL forward primer and 0.5 µL reverse primer. The amplification was carried out (Initial activation step 30 min at 50°C, 2.0 min at 95°C and Cycling step with denaturation of 15 s at 95°C, annealing of 1 min at 60.0°C for 45 cycles) in a PCR machine 7500 FAST (Applied Biosystems 7500 FAST). We included negative control in these rRT-PCR assays and used CDC DENV-1-4 Real-Time assay users guide report for results interpretation. The identification of DENV subtypes was performed in singleplex mode, with four separate reactions, one for each serotype (Table 1).

Statistical methods

In this study, the prevalence of dengue fever within the sample applied to seropositivity (detection of IgM and IgG), to rRT-PCR-detection and their 95% confidence intervals (95% CIs) was calculated. We calculated the mean of cycle threshold-values (CT-values). Specificity, sensitivity, positive predictive value and negative predictive value of rapid test SD BIOLINE were calculated in comparison to rRT-PCR detection. Chi^2 of Pearson or Fisher's exact were calculated when comparing frequencies between IgM/IgG positivity and rRT-PCR results. A significant difference was considered when $p \leq 0.05$. Data analysis was performed using Stata Statistical Software release 10 (StataCorp. 2007, StataCorp LP, College Station, TX).

RESULTS

DENV infection among patients

The median age of enrolled patients was 37.6 years (minimum of 4 years, maximum of 63 years) and the males to females ratio was 1.5:1. The main characteristics of participating patients are summarized in Table 2.

Of the 43 tested individuals, three were positive for anti-DENV IgM (7%) and 7 tested positive for anti-DENV IgG (16.3%). The three positive sera for IgM were also positive for IgG. No acute phase sample (positive for IgM only) was found. With regard to the clinical signs presented by the patients, the four positive for only IgG were in advanced convalescence and cured on the viral plan.

The molecular diagnosis surprisingly showed positive results in 21 (48.8%) patients. Only sera from two patients were positive to rRT-PCR, IgM and IgG. One serum was positive for IgM/IgG but negative for rRT-PCR. All 21 positive for dengue virus cases belonged to DENV-3 subtype. The mean of the CT-value was 25.2±6.5 (Minimum CT-value was 13±6.5 and the maximum CT-value was 36.9±6.5). When we compared IgM/IgG positive sera to rRT-PCR positive sera, Fisher's X^2 value was 0.3102 (p=0.518). When comparing the frequency of positive sera with sera obtaining positive rRT-PCR for DENV 3, X^2 value was 3.9 (p=0.095).

During the short period of occurrence of dengue suspected cases (from 8th October to 18th November), 21/43 (48.8%) cases were confirmed to be positive to dengue fever by rRT-PCR. Twelve confirmed cases (six by 10/08/2013 and six others by 10/23/2013) of dengue fever were registered at the medical centre, CMO, in Ouagadougou. The nine other cases were registered in the teaching hospital CHU-YO in Ouagadougou from 10/25/2013 to 11/08/2013 (Table 3).

Predictable values calculation of the rapid test SD BIOLINE used for serologic diagnosis in this study

Sensitivity of SD BIOLINE in comparison to dengue rRT-PCR

To test whether a sera containing dengue antigen by

Table 2. Main characteristics of patients with symptoms of dengue fever in the outbreak in Burkina Faso, 2013.

Variable	Number (n)	Percent
Age (years)		
4-9	1	2.3
10-19	1	2.3
20-29	5	11.6
30-44	11	25.6
≥45	24	55.8
Missing age	1	2.3
Sex		
Male	26	60.5
Female	17	39.5
DENV IgM antibodies		
Yes	3	7.0
No	40	93.0
DENV IgG antibodies		
Yes	7	16.3
No	36	83.7
DENV rRT-PCR		
Yes	21	48.8
No	22	51.2
DENV subtypes		
DENV 3	21	100
Other subtypes	0	0

rRT-PCR often contains IgM/IgG, we calculated the sensitivity. Applying the formula, the Sensitivity (Se) of SD BIOLINE in comparison to dengue rRT-PCR, Se = a/(a+b); a = true positive for IgM/IgG; b = false negative for IgM/IgG; Se= 6/(6+15)= 6/21= 28.5% (Table 4).

Specificity of SD BIOLINE in comparison to dengue rRT-PCR

To test whether a sera containing dengue antigen by rRT-PCR often is negative for IgM/IgG, we calculated the specificity. Applying the formula, the Specificity (Sp) of SD BIOLINE in comparison to dengue RT-PCR, Sp = d/c+d; c = false negative for rRT-PCR; d = true negative for rRT-PCR; Sp = 21/(1+21) = 95.4%.

Positive predictive value of SD BIOLINE in comparison to dengue rRT-PCR

We defined the positive predictive value (PPV) as follows: if sera are positive for IgM or/and IgG, how often are they positive to dengue by rRT-PCR? Applying the formula of the positive predictive value, PPV = a/(a+c); PPV = 6/(6+1) = 85.7%.

Negative predictive value of SD BIOLINE in comparison to dengue rRT-PCR

We defined the negative predictive value (NPV) as follows: if sera are negative for IgM or/and IgG, how often are they negative to dengue by rRT-PCR? Applying the formula of the negative predictive value, NPV = d/(d+b); NPV = 21/(21+15) = 58.3%.

DISCUSSION

The epidemiology of dengue fever (DF) was still poorly documented in Burkina Faso, and the present study is the first aiming at confirming dengue suspected cases by real time RT-PCR and assessing rapid diagnostic tests (RDTs) used for dengue detection at Ouagadougou in 2013. Most African countries have adopted artemisinin-based combination therapy (ACT) as first-line treatment for uncomplicated malaria to improve efficacy and limit the selection of drug-resistant parasites (Yeka et al., 2008). In the same aim, although several ACTs exist, currently only two have been widely adopted into policy in Burkina Faso: artemether- lumefantrine (AL) and artesunate-amodiaquine (AS/AQ), each of which is the recommended therapy for uncomplicated malaria. Subsequently, failures to malaria treatment using ACTs in Burkina Faso are very scarce (Zongo et al., 2007). However, in 2013 the large majority of treated cases encountered failures despite the negative results to malaria tests (Giemsa-stained thick blood smear and RDT).

DF suspected cases occurred in the last second quarter of the year, after the end of the rainy season in 2013. Except a missing age for one patient and one 17-years old patient, the other 19/21 (90.4%) dengue confirmed cases were from adult patients over 20 years. In Senegal, Faye et al. (2007) reported 93.7% of dengue fever in adults (35-41 years old) in 2003–2004. However, this is the first report of dengue virus subtype 3 (DENV-3) infections in Burkina Faso. Apart from DENV-3 virus outbreak which occurred in Cape Verde affecting more than 16,000 persons including 6 deaths, concomitant of imported cases from Senegal to Italy (Franco et al., 2010) and dengue outbreak in Côte d'Ivoire in 2008 (Akoua-Koffi et al., 2014), DENV-3 was rarely reported in the West Africa sub-region. The findings of this study pointed out Burkina Faso as a second dengue emerging country with DEN-3 virus after Côte d'Ivoire outbreak in 2008 (WHO, 2009). We can hypothesize that since 2008, when DENV-3 is circulating in the neighboring country, Côte d'Ivoire, the diffusion of this subtype DENV-3 virus has successfully crossed the border of Burkina Faso with the movement of the population between the two countries.

Table 3. Dengue fever positive patients' characteristics, serology and rRT-PCR results.

Patient	Age	Sex	Date of symptoms onset	IgM	IgG	DENV-3	CT-value
01	23	M	10/18/2013/CO	Negative	Negative	Positive	32.38
03	17	M	10 /18/2013/CO	Negative	Positive	Positive	26.93
04	33	F	10 /18/2013/CO	Negative	Negative	Positive	26.03
05	58	F	10 /18/2013/CO	Negative	Positive	Positive	29.50
06	26	M	10/18 /2013/CO	Positive	Positive	Positive	36.92
07	24	F	10 /18/2013/CO	Positive	Positive	Positive	17.38
11	53	M	10 /23/2013/CO	Negative	Negative	Positive	32.14
12	38	F	10 /23/2013/CO	Negative	Negative	Positive	22.36
13	41	M	10/23/2013/CO	Negative	Negative	Positive	25.13
17	34	F	10 /23/2013/CO	Negative	Negative	Positive	15.28
18	57	M	10 /23/2013/CO	Negative	Negative	Positive	20.99
20	36	F	10 /23/2013/CO	Negative	Negative	Positive	34.02
21	-	M	10 /25/2013/CHU-YO	Negative	Negative	Positive	28.27
23	53	M	10 /29/2013/CHU-YO	Negative	Positive	Positive	27.57
26	46	F	11/04/2013/CHU-YO	Negative	Positive	Positive	29.17
28	58	M	11/05/2013/CHU-YO	Positive	Positive	Negative	-----
31	26	F	11/05/2013/CHU-YO	Negative	Negative	Positive	13.00
36	46	F	11/08/2013/CHU-YO	Negative	Negative	Positive	23.10
39	30	F	11/06/2013/CHU-YO	Negative	Negative	Positive	22.32
43	43	M	11/07/2013/CHU-YO	Negative	Negative	Positive	28.74
45	21	F	11/06/2013/CHU-YO	Negative	Negative	Positive	16.03
49	35	M	11/08/2013/CHU-YO	Negative	Negative	Positive	23.17

Table 4. 2×2 results of dengue diagnosis by rapid test SD BIOLINE and rRT-PCR.

Dengue diagnosis	Positive rRT-PCR	Negative rRT-PCR
Positive for IgM/IgG	a=6	c=1
Negative for IgM/IgG	b=15	d=21

a = true positive for IgM/IgG; b = false negative for IgM/IgG; c = false negative for rRT-PCR; d = true negative for rRT-PCR.

Simultaneously in 2013, a dengue survey was conducted in other regions of Burkina Faso and it reported febrile cases of 2.7% (3/112) in the city of Zorgho, and 9.9% (15/151) in the city of Kaya, which were positive for IgM/IgG RTD, including two AgNS1 positive cases (SD BIOLINE Dengue Duo), but the subtypes of dengue virus were not identified (Ridde et al., 2014).

The lack of records of occurrence of dengue fever in Burkina Faso may be due to the confusion of dengue fever with clinical symptoms of malaria in absence of laboratory diagnosis (Costa Ade et al., 2010; Demanou et al., 2014). The literature on the low incidence of Dengue Hemorragic Fever (DHF) in Africa is still debatable that an African ancestry appeared to be relatively protective for severe forms of dengue but not for classic dengue fever (Halstead et al., 2001; Blanton et al., 2008; Demanou et al., 2014). Dengue fever was thought to be inexistent in Burkina Faso and was neglected until 2013 outbreak. In our sample, 7/43 (16.3%) specimens were only positive for specific IgM/IgG against DENV, leading to a good specificity of 100% but a very low sensitivity of 28.5%. In Table 3, as regards the patient N°28, a male of 58 years of age, from CHU-YO, had positive serology for IgM/IgG and negative result for rRT-PCR to DF.

The RTD for specific IgM/IgG detection for dengue fever diagnostic is an undeniable contribution compared to viral isolation because it saves time during an epidemic situation; however IgM/IgG generally appears after 7 days of the date of onset of the disease and can cause a false negative result (Akoua-Koffi et al., 2014). In the opposite, viral RNA presence in the first 3 or 5 days of the disease makes the molecular RT-PCR technique

more sensitive than the RTD at this period for dengue detection, as shown in Table 2. Indeed, the molecular test confirmed 21 DF cases in our sample, while RTD, SD BIOLINE detected only 7 positive cases; this means that rRT-PCR method detected 15 cases of dengue which were negative for RTD specific IgM/IgG. Molecular tools have become essential for dengue diagnosis especially as it allowed us to discriminate virus into different existing subtypes. All positive cases for dengue in our sample belonged to DENV-3 subtype.

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Conclusion

Our results confirmed the circulation of DENV-3 subtype in Burkina Faso. These findings demonstrated the importance to add dengue fever on the list of diseases on epidemiological surveillance as yellow fever or influenza in Burkina Faso. The contribution of molecular tests revealed indispensable tools for the dengue fever cases detection and the DENV subtypes discrimination.

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REFERENCES

Akoua-Koffi CG, Akran V, Faye O, Grandadam M, Ekaza E, Kouassi KS, Coulibaly D, Sall A, Dosso M (2014). Yellow Fever and Dengue Fever Serotype 3 Viruses Cocirculation in Côte d'Ivoire in 2008. African Journal of Pathology and Microbiology Vol. 3 (2014), doi:10.4303/ajpm/235834.

Blanton RE, Silva LK, Morato VG, Parrado AR, Dias JP, Melo PR, Reis EA, Goddard KA, Nunes MR, Rodrigues SG, Vasconcelos PF, Castro JM, Reis MG, Barreto ML, Teixeira MG (2008). Genetic ancestry and income are associated with dengue hemorrhagic fever in a highly admixed population. Eur J Hum Genet. Jun;16(6):762-5.

Collenberg E, Ouedraogo T, Ganamé J, Fickenscher H, Kynast-Wolf G, Becher H, Kouyaté B, Kräusslich HG, Sangaré L, Tebit DM (2006). Seroprevalence of six different viruses among pregnant women and blood donors in rural and urban Burkina Faso: a comparative analysis. J Medical Virology 78 (5): 683-692.

Costa Ade P, Bressan Cda S, Pedro RS, Valls-de-Souza R, Silva SD, Souza PR, Guaraldo L, Ferreira-da-Cruz Mde F, Daniel-Ribeiro CT, Brasil P (2010). Delayed diagnosis of malaria in a dengue endemic area in the Brazilian extra-Amazon: recent experience of a malaria surveillance unit in state of Rio de Janeiro. Rev Soc Bras Med Trop 43(5):571–4.

Demanou M, Pouillot R, Grandadam M, Boisier P, Kamgang B, Herve JP, Rogier C Rousset D, Paupy C (2014). Evidence of dengue virus transmission and factors associated with the presence of anti-dengue virus antibodies in humans in three major towns in cameroon. PLoS Negl Trop Dis., (7):e2950.

Fagbami AH, Mataika JU, Shrestha M, Gubler DJ (1995). Dengue type 1 epidemic with haemorrhagic manifestations in Fiji, 1989-90. Bulletin of the World Health Organization., 73(3):291-7.

Faye O, Diallo M, Dia I, Ba Y, Faye O, Mondo M, Sylla R, Faye PC, Sall AA (2007). Approche intégrée de la surveillance de la fièvre jaune : étude Pilote au Sénégal en 2003–2004, Bull Soc Pathol Exot, 100, 187–192.

Franco L, Di Caro A, Carletti F, Vapalahti O, Renaudat C, Zeller H, Tenorio A (2010). Recent expansion of dengue virus serotype 3 in West Africa, Euro Surveill, 15, art 19490.

Gonzalez J, Du Saussay C, Gautun J, McCormick J, Mouchet J (1985). La dengue au BurkinaFaso (ex Haute-Volta)) : épidémies saisonnières en milieu urbain à Ouagadougou. Bull Soc Pathol Exot Filiales, 78: 7–14

Guha-Sapir D, Schimmer B (2005). Dengue fever: new paradigms for a changing epidemiology. Emerging themes in epidemiology., 2(1):1.

Halstead SB, Streit TG, Lafontant JG, Putvatana R, Russell K, Sun W, Kanesa-Thasan N, Hayes CG, Watts DM (2001). Haiti: absence of dengue hemorrhagic fever despite hyperendemic dengue virus transmission. Am. J. Trop. Med. Hyg., 65(3):180-3.

Mamani E (2014). New serotype 5 of dengue virus: need to strengthen the molecular surveillance in Peru. Revista peruana de medicina experimental y salud publica.31(1):169-80.

Murray NEA, Quam MB, Wilder-Smith A (2013). Epidemiology of dengue: past, present and future prospects. Clinical Epidemiology, 5: 299–309.

Ridde V, Carabali M, Ly A, Druetz T, Kouanda S, Bonnet E, Haddad S (2014). The Need for More Research and Public Health Interventions on Dengue Fever in Burkina Faso. PLoS Negl. Trop. Dis, 8(6): e2859. doi:10.1371/journal. pntd.0002859.

Robert V, Lhuillier M, Meunier D, Sarthou JL, Monteny N, Digoutte JP, Cornet M Germain M, Cordellier R (1993). (Yellow fever virus, dengue 2 and other arboviruses isolated from mosquitos, in Burkina Faso, from 1983 to 1986. Entomological and epidemiological considerations). Bulletin de la Société de pathologie exotique, 86(2):90-100.

Suzarte E, Marcos E, Gil L, Valdes I, Lazo L, Ramos Y,

Pérez Y, Falcón V, Romero Y, Guzmán MG, González S, Kourí J, Guillén G, Hermida L (2014). Generation and characterization of potential dengue vaccine candidates based on domain III of the envelope protein and the capsid protein of the four serotypes of dengue virus. Arch Virol., 159(7):1629-40.

Were F (2012). The dengue situation in Africa. Paediatr. Int. Child Health. 32 Suppl., 1:18-21.

WHO, (2009). Dengue: guidelines for diagnosis, treatment, prevention and control; 147p.

WHO/SEARO (2011). Comprehensive guidelines for prevention and control of dengue and dengue haemorrhagic fever;196 p.

World Health Organization (2009). Dengue in Africa: emergence of DENV-3, Côte d'Ivoire, 2008. Wkly Epidemiol Rec 84: 85–88.

Yauch LE, Shresta S (2014). Dengue virus vaccine development. Advances in virus research., 88:315-72.

Yeka A, Dorsey G, Kamya MR, Talisuna A, Lugemwa M, Rwakimari JB, Staedke SG, Rosenthal PJ, Wabwire-Mangen F, Bukirwa H (2008). Artemether-lumefantrine versus dihydroartemisinin-piperaquine for treating uncomplicated malaria: a randomized trial to guide policy in Uganda. PLoS One, 3(6):e2390.

Zhao S, Fung-Leung WP, Bittner A, Ngo K, Liu X (2014). Comparison of RNA-Seq and microarray in transcriptome profiling of activated T cells. PloS one, 9(1):e78644.

Zongo I, Dorsey G, Rouamba N, Tinto H, Dokomajilar C, Guiguemde RT, Rosenthal PJ, Ouedraogo JB (2007). Artemether- lumefantrine versus amodiaquine plus sulfadoxine-pyrimethamine for uncomplicated falciparum malaria in Burkina Faso: a randomised non-inferiority trial. Lancet., 10;369(9560):491-8.