Introduction

Dengue is caused by the four serotypes of dengue virus (DENV-1-4), a flavivirus transmitted primarily by *Aedes aegypti* and *Ae. albopictus* mosquitoes. Dengue is recognized as a major health problem globally, causing an estimated 100 million cases in over 100 countries in Asia, Africa and the Americas [1–3]. Even though dengue disease burden is highest in Asia, the Americas have experienced a dramatic increase in the number of reported cases over the last thirty years [4,5].

Fever is a common symptom among children seeking medical care. Differentiating dengue from other febrile illnesses is key to providing timely and appropriate care [6]. In response to outbreaks of dengue fever in the 1950s and 1960s in many countries of the Asia-Pacific region, the World Health Organization (WHO) developed guidelines for dengue diagnosis, treatment and control which were approved in 1975 [7,8]. These guidelines defined a probable dengue case as an acute febrile illness accompanied by two or more of the following criteria: headache, retro-orbital pain, myalgia, arthralgia, rash, hemorrhagic manifestations and leukopenia. The first four criteria are symptoms and thus, by definition, are reported by the patient and cannot be measured directly by the physician. Moreover, these symptoms cannot be easily verbalized by younger children, particularly those under 4 years [9].

In 2009, the WHO published new guidelines for dengue diagnosis, treatment, prevention and control [6], which includes a revised dengue case definition. In these guidelines, a probable dengue case was defined as fever and two of the following criteria: nausea/vomiting, rash, aches and pains, positive tourniquet test, leukopenia, and any warning sign (abdominal pain or tenderness, persistent vomiting, clinical fluid accumulation, mucosal bleeding, lethargy/restlessness, liver enlargement >2 cm, or increase in hematocrit concurrent with rapid decrease in platelet count) [6]. In this revised case definition, only one symptom – aches and pains – is included, the remaining criteria are signs that are evaluated by a healthcare professional.

Several studies have assessed the performance of the traditional dengue case definition in Thailand, Singapore, Peru, and Colombia [9–11]. Overall, the definition had high sensitivity
Ethics statement

Here, we compared the diagnostic utility of the traditional and revised WHO dengue case definitions in two different settings. First, we analyzed data from suspected dengue cases identified in a prospective pediatric cohort study based at primary health care center in Managua, Nicaragua, from 2004 to 2011 [13,14]. Second, we examined participants enrolled in a prospective study of dengue based at the National Pediatric Reference Hospital in Managua, Nicaragua, from 2005 to 2012 [15–19]. In each study, we evaluated the sensitivity, specificity and predictive values of the individual signs and symptoms of and of the traditional and revised dengue case definitions. In addition, we carried out receiver operating characteristic (ROC) analyses and tested the performance of both case definitions in younger versus older children.

Materials and Methods

Ethics statement

The protocols for the Pediatric Dengue Cohort Study and the hospital-based study were reviewed and approved by the Institutional Review Boards (IRB) of the University of California, Berkeley, and of the Nicaraguan Ministry of Health. Parents or legal guardians of all subjects in both studies provided written informed consent, and subjects 6 years of age and older provided assent. In the hospital study, children 6 years of age and older displaying signs of altered consciousness at the time of recruitment and thus unable to provide assent were excluded.

Cohort study

The Pediatric Dengue Cohort Study is an ongoing community-based prospective study established in 2004 in Managua, Nicaragua [13,14]. The study is based at the local municipal health center, the Health Center Socrates Flores Vivas (HCSFV). The area served by the HCSFV is District II of Managua, a low-to-middle-income area with a population of approximately 62,500.

In August–September 2004, children aged two to nine years old living in District II were enrolled, and new participants have been enrolled each year since then to maintain the cohort age structure. Children are withdrawn from the study when they reach 15 years of age. Participants are encouraged to present at the first sign of illness to the HCSFV, where study physicians provide medical care and screen for signs and symptoms of dengue. Suspected dengue cases, as defined by the traditional WHO dengue case definition [8] and febrile participants without other apparent origin (undifferentiated febrile illnesses) who present in the first six days of illness are screened by serological, molecular and virological methods for acute DENV infection [13,14]. A convalescent-phase blood sample (two weeks after onset of fever) is also collected for serological assays. Participants are followed during the acute phase of illness by study physicians at the HCSFV. Clinical data, including signs, symptoms, and treatment, are recorded at every visit. Data collected from August 2004 to December 2011 was used for this analysis. The presence or absence of each criterion for both case definitions was evaluated throughout the course of disease.

Hospital-based study

An ongoing hospital-based prospective study of dengue to study clinical, immunological and viral risk factors for severe dengue was established in 1998 in the Infectious Disease Ward of the Hospital Infantil Manuel de Jesús Rivera Hospital (HIMJR), the national pediatric reference hospital in Managua [15–20]. In-patients and out-patients between 6 months and 14 years of age are enrolled when they present at the HIMJR with documented or reported fever of less than 7 days and one or more of the following signs and symptoms: headache, arthralgia, myalgia, retro-orbital pain, positive tourniquet test, petechiae or others signs of bleeding. Enrollment occurs each year during the peak of the dengue season (August-January). Children with a defined focus other than dengue are excluded, as well as children weighing less than 8 kg.

Upon enrollment, a medical history is taken, and a complete physical exam is performed. Acute blood samples are collected daily for complete blood count and serological, virological, and molecular testing for DENV infection. A convalescent-phase blood sample (two weeks after onset of fever) is also collected. Both in-patient and out-patient subjects are followed clinically throughout the acute phase of illness. Clinical data, including signs, symptoms, and treatment, are recorded daily through ambulatory follow-up visits for out-patients, and at least every 12 hours for in-patients during hospitalization. All data is collected on standardized forms. Participants are followed for 3 to 8 days. Participants requiring more intensive therapies are transferred to the intensive care unit. Data collected from August 2005 to January 2012 was used for analysis. The presence or absence of each criterion for both case definitions was evaluated over the course of disease evolution.

Laboratory-confirmed dengue cases

For both studies, a participant was considered positive for DENV infection when laboratory tests met one or more of the following criteria: 1) dengue viral RNA was detected by RT-PCR [21,22]; 2) DENV was isolated [21]; 3) seroconversion of DENV-specific IgM was detected by MAC-ELISA in paired acute and convalescent samples [21,23]; and 4) DENV-specific antibody tier by Inhibition ELISA [20,24,25] demonstrated a 4-fold or greater increase between acute and convalescent sera. Primary DENV
infections were considered those in which the convalescent antibody titer was \(<2,560\), and secondary infections were considered those in which the convalescent antibody titer was \(\geq 2,560\) as determined by Inhibition ELISA. A case was considered indeterminate if RT-PCR yielded negative results, no DENV was isolated and a convalescent sample could not be obtained. Indeterminate cases were excluded from this analysis.

Dengue signs, symptoms, and case definitions

Signs and symptoms were defined and measured as described in Table 1. As children under 4 years old cannot easily express aches, reporting of headache, retro-orbital pain, myalgia, arthralgia, and aches and pains was only considered for children aged 4 years old or more. Dengue cases were defined according to WHO criteria. The traditional dengue case definition was the presence of fever (or history of fever) plus two or more of the following: headache, retro-orbital pain, myalgia, arthralgia, rash, hemorrhagic manifestations and leukopenia (white blood cell count \(<5,000\text{ cells/mm}^3\)) [7,8]. The revised dengue case definition was the presence of fever (or history of fever) plus two or more of the following: nausea/vomiting, rash, aches and pains, leukopenia, positive tourniquet test (petechiae \(\geq 20\text{ per inch}^2\)) and any warning sign (abdominal pain or tenderness, persistent vomiting, clinical fluid accumulation, mucosal bleeding, lethargy/restlessness, liver enlargement \(>2\,\text{cm}\), or increase in hematocrit concurrent with rapid decrease in platelet count) [6] (Table 1).

Table 1. Traditional and revised WHO dengue case and criteria definitions.

| Terms                                  | Definition                                                                 |
|----------------------------------------|---------------------------------------------------------------------------|
| Suspected dengue case (Traditional definition)  | Fever for less than 7 days plus two or more of the following criteria: headache, retro-orbital pain, myalgia, arthralgia, rash, hemorrhagic manifestations, leukopenia |
| Suspected dengue case (Revised definition)     | Fever for less than 7 days plus two or more of the following criteria: nausea/vomiting, rash, aches and pains, positive tourniquet test, leukopenia, any warning sign |
| Fever                                   | Temperature \(\geq 37.8\,\text{C}\) as recorded by study personnel or history of fever |
| Headache, retro-orbital pain, myalgia and arthralgia | Symptoms reported by participants aged \(\geq 4\) years old |
| Rash                                    | Change of color, appearance or texture of the skin observed by a study physician |
| Hemorrhagic manifestations             | Spontaneous petechiae, purpura, ecchymosis, hematoma, hemoptysis, epistaxis, gingival bleeding, melena, hematemesis, hematuria, subconjunctival hemorrhage, menorrhagia, or vaginal bleeding as observed by a study physician or reported by the patient, or positive tourniquet test |
| Leukopenia                              | White blood count \(<5,000\text{ cells/mm}^3\) |
| Nausea/vomiting                         | Nausea or at least one emesis reported by the patient or observed by a study physician |
| Aches and pains                         | Any of the following: headache, retro-orbital pain, myalgia or arthralgia |
| Positive tourniquet test                | \(\geq 20\text{ petechiae/inch}^2\) |
| Warning signs                           | Abdominal pain or tenderness, persistent vomiting, clinical fluid accumulation, mucosal bleeding, lethargy/restlessness, liver enlargement, increase in hematocrit concurrent with rapid decrease in platelet count |
| Abdominal pain or tenderness            | Pain in the abdominal region reported spontaneously by the patient or when palpated by a study physician |
| Persistent vomiting                     | Three or more emesis in a period of one hour, or five or more in a period of six hours |
| Clinical fluid accumulation             | Peri-orbital, facial or lower limb edema as reported by the study physician, or pleural effusion, ascites or gall-bladder wall thickening (\(\geq 3\) mm) as observed via X-ray radiography or ultrasonography |
| Mucosal bleeding                        | Any of the following: hemoptysis, epistaxis, gingival bleeding, melena, hematemesis, hematuria, menorrhagia, vaginal bleeding, or subconjunctival hemorrhage as observed by a study physician or reported by the patient |
| Lethargy/restlessness                   | Glasgow coma scale score \(<15\) for children aged 5 years or more or Blantyre coma scale \(<5\) for children under 5, as evaluated by a study physician |
| Liver enlargement                       | Liver enlarged \(\geq 2\,\text{cm}\) below the edge of the ribs as palpated by a study physician |
| Increase in hematocrit concurrent with rapid decrease in platelet count | Decrease in platelet count \(>10,000\text{ platelets/mm}^3\) in 24 hours, or increase in hematocrit accompanied by a platelet count \(<100,000\text{ platelets/mm}^3\) |

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Statistical analysis

For both studies, signs and symptoms presented at any time over the course of the disease were included in the analysis. A Chi-squared test was used to associate categorical variables with dengue laboratory results. To determine sensitivity, specificity and predictive values of each criterion for dengue diagnosis, the laboratory result was considered as the gold standard. A criterion with a sensitivity and specificity of 80% or more was considered of high diagnostic value [26]. ROC (Receiver operator characteristic) analysis was carried by giving one point per criterion present (maximum of 7 points/criteria for the traditional case definition and maximum of 6 points/criteria for the revised case definition). For each point, the sensitivity, specificity and accuracy for diagnosing dengue was calculated. The value of the area under the curve (AUC) was also computed. A test with an AUC value
between 0.51 and 0.70 was considered of poor diagnostic value; from 0.71 to 0.90 as useful for some purposes; and >0.90 as of high diagnostic value [27]. All data analyses were performed using Intercooled Stata 9.0 (StataCorp LP, College Station, Texas), with a 95% confidence level.

Results

Study population

Cases were identified in the Pediatric Dengue Cohort Study and a hospital-based study, both based in Managua, Nicaragua. In the cohort study, 3,617 cases were identified from August 2004 to December 2011 (Table 2). A total of 3,407 cases were included in the analysis. Two hundred and ten cases did not have a confirmed positive or negative laboratory result and were excluded from the analysis (see Methods). The median age of cases was 7.0 years (interquartile range [IQR]: 5.0–9.0 years). Cases were equally distributed among females (48.9%) and males (51.1%) (Z-test p = 0.145). The median day of illness at presentation was 2 (IQR: 1–2). A total of 476 (14.0%) cases were laboratory-confirmed DENV infections. Among laboratory-confirmed cases, 50.4% experienced a secondary DENV infection. Infecting DENV serotypes were detected by RT-PCR and confirmed by virus isolation. Most infections were caused by DENV-3 (52.3%) followed by DENV-2 (27.7%) and DENV-1 (12.0%). In 35 (7.4%) of the confirmed dengue cases, the infecting serotype was not identified. In the hospital study, 1,210 participants were enrolled from August 2005 to January 2012 (Table 2). Fifty (4.1%) participants had indeterminate results in the diagnostic assays and were excluded from the analysis. Information from the remaining 1,160 participants was used for analysis. The median participant age was 7.9 years (IQR: 4.9–11.1 years). Participants were equally distributed among females (47.8%) and males (52.2%) (Z-test p = 0.142). The median day of illness at enrollment was 4 (IQR: 3–5). A total of 723 (62.3%) cases were confirmed DENV infections. Of these, 53.1% were secondary infections. The infecting DENV serotype was mainly DENV-3 (58.9%) followed by DENV-2 (22.8%) and to a lesser extent DENV-1 (7.5%) (Table 2). The infecting DENV serotype was not identified in 77 (10.7%) of the confirmed dengue cases.

Diagnostic value of traditional and revised dengue case definition signs and symptoms

Using clinical data collected in both the cohort and the hospital-based studies, we compared the diagnostic utility of the revised versus the traditional WHO case definitions. First, we analyzed the association of each sign and symptom of both dengue case definitions (Table 1) with the positivity of dengue diagnosis as defined by laboratory testing, and calculated their sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV).

Table 2. General characteristics of the study population.

| Characteristics                        | Cohort study (# %) | Hospital study (# %) |
|----------------------------------------|--------------------|----------------------|
| Cases evaluated                        | 3,617              | 1,210                |
| Cases included in the analysis         | 3,407              | 1,160                |
| Laboratory-confirmed dengue            | 476 (14.0)         | 723 (62.3)           |
| Age in years – median (interquartile range, IQR) | 7.0 (5.0–9.0) | 7.9 (4.9–11.1)              |
| <1 year                                | 1,661 (48.9)       | 555 (47.8)           |
| 1–3 years                              | 481 (14.1)         | 198 (17.1)           |
| 4–9 years                              | 2,100 (61.7)       | 521 (44.9)           |
| 10–14 years                            | 826 (24.2)         | 391 (33.7)           |
| Sex                                    |                    |                      |
| Female                                 | 1,661 (48.9)       | 555 (47.8)           |
| Day of illness at presentation (median, IQR) | 2 (1–2)          | 4 (3–5)              |
| Immune response*                       |                    |                      |
| Primary                                | 222 (46.6)         | 306 (42.3)           |
| Secondary                              | 240 (50.4)         | 384 (53.1)           |
| Indeterminate                          | 14 (3.0)           | 33 (4.6)             |
| Dengue virus serotype*                 |                    |                      |
| DENV-1                                 | 57 (12.0)          | 54 (7.5)             |
| DENV-2                                 | 132 (27.7)         | 165 (22.8)           |
| DENV-3                                 | 249 (52.3)         | 426 (58.9)           |
| DENV-4                                 | 1 (0.2)            | —                    |
| DENV-1 & DENV-2 co-infection           | 1 (0.2)            | —                    |
| DENV-1 & DENV-4 co-infection           | 1 (0.2)            | —                    |
| DENV-3 & DENV-4 co-infection           | —                  | 1 (0.1)              |
| Unknown                                | 35 (7.4)           | 77 (10.7)            |

*Among laboratory-confirmed dengue cases.

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both a high sensitivity and a high specificity. For instance, aches and pains and headache were very frequent in patients with dengue (sensitivity 85%), but also in patients without dengue (specificity 25%) (Table 3). On the contrary, nausea and vomiting, positive tourniquet test and rash, had high specificity but low sensitivity. The remaining signs and symptoms had sensitivities ranging from 40 to 70% and specificities ranging from 60 to 80%. Strikingly, all criteria had an NPV over 85%, meaning that the absence of these signs/symptoms could be used to rule out dengue. However, the PPV was consistently low (15–35%; except for rash, 55%).

In the hospital-based study, only six of the 11 criteria were associated with dengue, namely any warning sign, rash, leukopenia, hemorrhagic manifestations, positive tourniquet test and nausea/vomiting (chi-square test p < 0.05) (Table 4). However, none of these criteria had both high sensitivity and high specificity. Any warning sign, rash, leukopenia and hemorrhagic manifestations showed a high sensitivity (>80%) but a low specificity (15–55%) (Table 4). Positive tourniquet test was the only criterion with high specificity (80.3%) but its sensitivity was low (49.7%), while nausea/vomiting had low sensitivity (48.7%) and specificity (57.9%). The remaining criteria (aches and pains, headache, retro-orbital pain, myalgia and arthralgia) were not associated with laboratory diagnosis of dengue. None of the signs and symptoms yielded high predictive values, except for positive tourniquet test, which had a PPV of 80.7% (Table 4).

Receiver Operating Characteristics analysis

Next, we carried out a receiver operating characteristics (ROC) analysis of two diagnostic models based on the criteria included in the traditional and revised dengue case definitions. For each definition, we calculated the sensitivity, specificity and accuracy of the presence of only fever, fever plus one or more criteria, fever plus two or more criteria, etc. (Tables 5 and 6). By definition, the sensitivity, specificity and accuracy of the WHO dengue case definitions per se correspond to those calculated for fever plus two or more criteria.

As expected, the sequential addition of criteria decreased sensitivity and increased specificity (Tables 5 and 6). In the cohort

| Signs/symptoms                      | Sensitivity % (95% CI) | Specificity % (95% CI) | PPV % (95% CI) | NPV % (95% CI) |
|-------------------------------------|------------------------|------------------------|----------------|----------------|
| Aches and pains a,b                  | 91.8 (88.9–94.2)       | 18.4 (16.9–20.0)       | 16.7 (15.2–18.2) | 92.7 (90.1–94.8) |
| Headache a,b                         | 86.8 (83.3–89.9)       | 22.2 (20.6–23.9)       | 16.5 (15.0–18.1) | 90.5 (87.9–92.7) |
| Leukopenia b                         | 68.3 (63.9–72.4)       | 79.9 (78.4–81.3)       | 35.5 (32.4–38.7) | 93.9 (92.9–94.8) |
| Any warning sign b                   | 66.4 (61.9–70.6)       | 63.6 (61.9–65.4)       | 22.9 (20.7–25.2) | 92.1 (90.8–93.2) |
| Arthralgia a,b                       | 56.5 (51.7–61.1)       | 61.1 (59.1–63.0)       | 20.5 (18.2–22.9) | 88.8 (87.2–90.2) |
| Myalgia b                            | 52.4 (47.6–57.1)       | 66.4 (64.5–68.3)       | 21.7 (19.2–24.3) | 88.7 (87.2–90.1) |
| Hemorrhagic manifestations b         | 52.1 (47.5–56.7)       | 76.7 (75.2–78.3)       | 26.7 (23.8–29.6) | 90.8 (89.6–91.9) |
| Retro-orbital pain a,b               | 41.7 (37.1–46.5)       | 73.8 (72.0–75.5)       | 22.0 (19.2–25.0) | 87.7 (86.2–89.1) |
| Positive tourniquet test b,c         | 39.9 (35.5–44.5)       | 88.2 (87.0–89.4)       | 35.6 (31.6–39.9) | 90.0 (88.8–91.0) |
| Rash b                              | 29.6 (25.6–33.9)       | 96.1 (95.3–96.8)       | 55.3 (49.0–61.5) | 89.4 (88.2–90.4) |
| Nausea/vomiting b                    | 27.1 (23.2–31.3)       | 82.2 (80.7–83.5)       | 19.8 (16.8–23.1) | 87.4 (86.1–88.6) |

*Children under 4 years were excluded from the analysis. N for negative cases = 2,485; N for positive cases = 441.
*bSigns and symptoms associated with dengue (p < 0.05).
*c23 with missing data among negative cases (N = 2,908).

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| Signs/symptoms                      | Sensitivity % (95% CI) | Specificity % (95% CI) | PPV % (95% CI) | NPV % (95% CI) |
|-------------------------------------|------------------------|------------------------|----------------|----------------|
| Any warning sign b                  | 92.8 (90.7–94.6)       | 15.6 (12.3–19.3)       | 64.5 (61.5–67.4) | 56.7 (47.3–65.7) |
| Rash b                              | 91.7 (89.4–93.6)       | 36.2 (31.6–40.9)       | 70.4 (67.4–73.3) | 72.5 (66.0–78.3) |
| Leukopenia b                        | 83.1 (80.2–85.8)       | 49.4 (44.6–54.2)       | 73.1 (69.9–76.1) | 63.9 (58.5–69.0) |
| Aches and pains a                   | 82.0 (78.7–84.9)       | 20.8 (16.4–25.7)       | 67.0 (63.5–70.4) | 37.0 (29.8–44.7) |
| Hemorrhagic manifestations b        | 80.6 (77.6–83.5)       | 52.6 (47.8–57.4)       | 73.8 (70.6–76.8) | 62.2 (57.0–67.1) |
| Headache a                          | 74.5 (70.8–77.9)       | 26.6 (21.8–31.9)       | 66.6 (62.9–70.1) | 34.7 (28.7–41.2) |
| Positive tourniquet test b          | 49.7 (45.9–53.4)       | 80.3 (76.3–83.9)       | 80.7 (76.7–84.2) | 49.1 (45.4–52.8) |
| Nausea/vomiting b                   | 48.1 (44.4–51.8)       | 57.9 (53.1–62.6)       | 65.4 (61.2–69.5) | 40.3 (36.4–44.2) |
| Retro-orbital pain a                | 41.6 (37.6–45.6)       | 61.0 (55.3–66.5)       | 67.7 (62.6–72.4) | 34.8 (30.7–38.9) |
| Myalgia a                           | 36.1 (32.3–40.1)       | 69.2 (63.7–74.3)       | 69.6 (64.2–74.7) | 35.6 (31.7–39.5) |
| Arthralgia a                        | 29.5 (25.9–33.3)       | 72.7 (67.4–77.6)       | 67.9 (61.9–73.5) | 34.5 (30.8–38.3) |

*Children under 4 years were excluded from the analysis. N for negative cases = 308; N for positive cases = 604.
*Signs and symptoms associated with dengue (p < 0.05).

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study, the ROC analysis based on the traditional dengue case definition showed that the highest percentage of cases correctly classified (accuracy) was for fever plus 6 or more criteria (86.9%).

For this number of signs/symptoms, the specificity was high (98.5%), but the corresponding sensitivity very low (15.6%) (Table 5). For the revised dengue case model, the highest accuracy was observed for fever and 4 or more criteria (88.4%), with a high specificity (96.8%) but again a low sensitivity (36.6%). No data point in the ROC analysis showed both a high sensitivity (>80%) and high specificity (>80%). The area under the curve (AUC) of the ROC curve was slightly higher (chi-square test p < 0.001) for the revised model, 0.80 (95%CI 0.78–0.82), than for the traditional model, 0.75 (95%CI 0.73–0.78) (Fig. 1A).

In the hospital study, the greatest accuracy was measured for fever plus 3 or more criteria in the traditional model (71.8%) with a sensitivity of 85.9% and a specificity of 48.5% (Table 6). For the revised model, the greatest accuracy was observed for fever plus 4 or more criteria (71.6%) with a sensitivity of 78.7% and a specificity of 60.0%. As in the cohort study, no data point in the ROC analysis showed both a high sensitivity (>80%) and high specificity (>80%). The area under the curve (AUC) of the ROC curve was slightly higher (chi-square test p < 0.001) for the revised model, 0.75 (95%CI 0.71–0.77) (Fig. 1B).

Table 5. Receiver operating characteristics (ROC) analysis of two diagnostics models based on the list of criteria included in each dengue case definition, cohort study.

| Number of criteria | Traditional dengue case definition | Revised dengue case definition |
|--------------------|-----------------------------------|-------------------------------|
|                    | Sensitivity | Specificity | Accuracy | Sensitivity | Specificity | Accuracy |
| Fever only         | 100.0       | 0.0         | 14.0     | 100.0       | 0.0         | 14.0     |
| Fever plus 1 or more criteria | 97.1       | 15.7       | 27.0     | 96.0       | 15.6       | 26.9     |
| Fever plus 2 or more criteria | 89.3     | 43.1       | 49.6     | 86.6       | 55.2       | 59.6     |
| Fever plus 3 or more criteria | 76.7       | 61.2       | 63.4     | 65.6       | 83.3       | 80.8     |
| Fever plus 4 or more criteria | 59.5       | 76.8       | 74.4     | 36.6       | 96.8       | 88.4     |
| Fever plus 5 or more criteria | 36.8       | 91.9       | 84.2     | 11.0       | 99.8       | 87.5     |
| Fever plus 6 or more criteria | 15.6       | 98.5       | 86.9     | 2.3        | 100.0      | 86.3     |
| Fever plus 7 criteria | 4.8        | 99.7       | 86.4     | —          | —          | —        |

*Dengue case definition. 

Table 6. Receiver operating characteristics (ROC) analysis of two diagnostics models based on the list of criteria included in each dengue case definition, hospital-based study.

| Number of criteria | Traditional dengue case definition | Revised dengue case definition |
|--------------------|-----------------------------------|-------------------------------|
|                    | Sensitivity | Specificity | Accuracy | Sensitivity | Specificity | Accuracy |
| Fever only         | 100.0       | 0.0         | 62.3     | 100.0       | 0.0         | 62.3     |
| Fever plus 1 or more criteria | 99.6       | 5.3         | 64.1     | 99.9       | 2.5         | 63.2     |
| Fever plus 2 or more criteria | 96.7       | 22.0       | 68.5     | 99.3       | 8.5         | 65.1     |
| Fever plus 3 or more criteria | 85.9       | 48.5       | 71.8     | 93.8       | 29.1       | 69.4     |
| Fever plus 4 or more criteria | 62.0       | 66.1       | 63.5     | 78.7       | 60.0       | 71.6     |
| Fever plus 5 or more criteria | 35.7       | 82.4       | 53.5     | 48.0       | 85.4       | 62.1     |
| Fever plus 6 or more criteria | 20.9       | 91.8       | 47.6     | 16.6       | 95.9       | 46.5     |
| Fever plus 7 criteria | 11.8       | 97.5       | 44.1     | —          | —          | —        |

*Dengue case definition. 

Effect of age in the diagnostic utility of the dengue case definitions

We then evaluated the diagnostic utility of the traditional and revised dengue case definitions. Sensitivity and specificity as well as positive and negative predictive values were calculated for each case definition. In the cohort study, the traditional case definition had a sensitivity of 89.3% and a specificity of 43.1% (Table 7). The NPV was high (96.1%) and the PPV low (20.3%). We then stratified this analysis by age, taking into account that symptoms such as arthralgia, headache, myalgia, and retro-orbital pain can only be reliably reported by children aged 4 years or more. In children under the age of 4, for whom these symptoms were not included, the sensitivity dropped to 37.1%, 2.5-fold lower than in children over 4 (91.4%) (Table 7). On the contrary, the specificity was higher in the younger age group compared to the older (95.3% versus 39.4%). Comparable observations were made for the revised case definition. Overall, the revised definition had similar sensitivity, PPV and NPV when compared to the traditional definition. Its specificity was higher to some extent (55.2% versus 43.1%, McNemar test p < 0.001). When stratifying by age, children under the age of 4 showed a strong reduction in sensitivity and an increase in specificity (Table 7). This effect was observed despite the fact that in the revised dengue case definition,
only one criterion is not accurately reported by children under 4 years old (aches and pains).

In the hospital study, the traditional definition showed a very high sensitivity (96.7%) and a low specificity (22.0%) (Table 8). Its positive and negative predictive values were 67.2% and 80.0%, respectively. In children under 4 years old, the sensitivity decreased slightly while the specificity improved. The revised case definition had an even higher sensitivity (99.3%, McNemar test \( p < 0.001 \)) and a lower specificity (8.5%, McNemar test \( p < 0.001 \)) than the traditional definition (Table 8). Similar to the traditional definition, the sensitivity in children under the age of 4 showed a marginal decrease and the specificity was increased when compared to older children.

**Discussion**

Here, we compared the traditional WHO dengue case definition, which was implemented for over 40 years, and the revised definition approved in 2009. The case definitions were compared in two distinct pediatric studies in Managua, Nicaragua: a prospective cohort study based at a municipal health care center, and a clinical study based at the national pediatric reference hospital. In the cohort study, the traditional case definition had high sensitivity (89.3%) and low specificity (43.1%), while the revised definition had similar sensitivity (86.6%) and higher specificity (55.2%, \( p < 0.001 \)). In participants under 4 years, both case definitions showed a dramatically reduced sensitivity (37.1% and 45.7% for the traditional and revised definition, respectively) as well as an increased specificity (95.3% and 80.5%, respectively). In the hospital study, the traditional case was also highly sensitive (96.7%) and poorly specific (22.0%), whereas the revised case definition had slightly higher sensitivity (99.3%, \( p < 0.001 \)) but lower specificity (8.5%, \( p < 0.001 \)). In participants under 4 years, both case definitions displayed a reduction in sensitivity (87.4% for the traditional definition and 96.6% for the revised definition) and an increase in specificity (50.4% and 22.5%, respectively).

Several studies have assessed the traditional WHO dengue case definition [9–12]. These studies have been conducted in different countries (Thailand [9], Peru [10], Colombia [11], and Singapore [12]) and settings (hospitals [9] and health centers [10–12]) and with different inclusion criteria. Three studies relied on clinical data from a single visit, although they differed in the time of disease evolution at the time of inclusion (within 3 [12] or 4 days [10,11]). Similarly to our study, one study included clinical data from initial and follow-up patient evaluations throughout the course of the disease [9]. Despite these differences, all studies

**Table 7.** Diagnostic values of the traditional and revised WHO dengue case definitions by age, cohort study.

| WHO Dengue case definition | Sensitivity % (95% CI) | Specificity % (95% CI) | PPV % (95% CI) | NPV % (95% CI) |
|---------------------------|------------------------|------------------------|----------------|----------------|
| **Traditional case definition** | | | | |
| All patients              | 89.3 (86.2–91.9)       | 43.1 (41.3–44.9)       | 20.3 (18.6–22.1) | 96.1 (94.9–97.1) |
| Children < 4 years old    | 37.1 (21.5–55.1)       | 95.3 (92.9–97.1)       | 38.2 (22.2–56.4) | 95.1 (92.1–96.9) |
| Children ≥ 4 years old    | 91.4 (88.4–93.8)       | 39.4 (37.5–41.3)       | 21.1 (19.3–23.0) | 96.3 (94.9–97.3) |
| **Revised case definition** | | | | |
| All patients              | 86.6 (83.2–89.5)       | 55.2 (53.4–57.0)       | 23.9 (21.9–26.0) | 96.2 (95.2–97.1) |
| Children < 4 years old    | 45.7 (28.8–63.4)       | 80.5 (76.5–84.1)       | 15.7 (9.2–24.2)  | 94.9 (92.5–96.9) |
| Children ≥ 4 years old    | 91.2 (88.1–93.6)       | 45.9 (44.5–48.5)       | 23.3 (21.4–25.4) | 96.7 (95.5–97.7) |

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Figure 1. Receiver operating characteristic (ROC) curves of two diagnostic models based on the criteria included in each dengue case definition. (A) Cohort study. The area under the curve (AUC) of the ROC curve was slightly higher for the revised model (0.80) than for the traditional model (0.75) (chi-square test \( p < 0.001 \)). (B) Hospital-based study. The AUC was slightly higher for the revised model (0.75) than for the traditional model (0.71) (chi-square test \( p < 0.001 \)).

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The 6 criteria included in the revised case definition, only one, study (37.1% versus 91.4% in children aged 4 years and more). Of under 4 years old. The reduction was considerable in the cohort sensitivity of the traditional case definition when used in children phenomenon was most pronounced in the youngest children [30]. cases did not meet the traditional WHO case definition, and this stratify its results by age [9]. In a previous report about our cohort One study included children aged one to 13 years old but did not at significant risk for dengue [29]. Most studies evaluating the these symptoms cannot be accurately reported by younger than the evaluation of signs by a health practitioner. Moreover, Reporting of these symptoms by the patient is more subjective, namely headache, retro-orbital pain, myalgia and arthralgia. Of these, 4 are symptoms reported by the patient, firstly the inclusion criteria in the studies are different. In the cohort study, suspected dengue [8] and all febrile cases without an apparent cause were studied. In the hospital, febrile cases accompanied by at least one of the following signs or symptoms were included: headache, arthralgia, myalgia, retro-orbital pain, positive tourniquet test, petechiae or others signs of bleeding. Second, the criteria included in the case definitions are measured more frequently in the hospital than at the health center where the cohort study is based. Third, the equipment available to health practitioners in both settings are different. For instance, the use of X-ray radiography or ultrasonography is limited to the hospital. Fourth, more severe cases are seen in the hospital than at the health center where the cohort study includes more mild cases. Finally, participants present earlier during the course of illness to the health center than to the hospital. In summary, we show in two different settings that the revised dengue case definition performs similarly when compared to the traditional case definition. Both case definitions had high sensitivity (over 85%) but low specificity (55% or less). Owing to their diagnostic performance, both case definitions should be primarily used for screening purposes. However, in a primary care setting, neither case definition performed well as a screening test in younger children, as they showed low sensitivity and high specificity. Thus, when using either of the case definitions, particular attention should be paid to younger children. These results further emphasize the need for confirmatory dengue diagnostic methods. At present, laboratory-based testing remains aches and pains, is a symptom, which in theory makes this definition more objective and thus more applicable to younger children. However, our analysis showed that the sensitivity of the revised case definition is also dramatically affected by age. In the cohort study, the sensitivity for children aged less than 4 years was twice as low as the sensitivity for older children (45.7% versus 91.2%). Finally, no significant difference in the sensitivity of both case definitions in the younger age group was observed (37.1% and 45.7% for the traditional and revised definition, respectively). Taken together, these results show that despite containing less criteria that cannot be expressed by younger children, the revised case definition does not perform better than the traditional definition in children aged less than 4 years.

Although this analysis was not designed to compare the cohort and hospital studies, the differences in the diagnostic utility of the dengue case definitions are striking. The sensitivity of both case definitions is ~10 percentage points higher in the hospital study. Their specificity is higher in the cohort study (two- and five-fold higher for the traditional and revised definition, respectively). Several reasons might explain these differences. First and foremost, the inclusion criteria in the studies are different. In the cohort study, suspected dengue [8] and all febrile cases without an apparent cause were studied. In the hospital, febrile cases accompanied by at least one of the following signs or symptoms were included: headache, arthralgia, myalgia, retro-orbital pain, positive tourniquet test, petechiae or others signs of bleeding. Second, the criteria included in the case definitions are measured more frequently in the hospital than at the health center where the cohort study is based. Third, the equipment available to health practitioners in both settings are different. For instance, the use of X-ray radiography or ultrasonography is limited to the hospital. Fourth, more severe cases are seen in the hospital, whereas the cohort study includes more mild cases. Finally, participants present earlier during the course of illness to the health center than to the hospital.

In summary, we show in two different settings that the revised dengue case definition performs similarly when compared to the traditional case definition. Both case definitions had high sensitivity (over 85%) but low specificity (55% or less). Owing to their diagnostic performance, both case definitions should be primarily used for screening purposes. However, in a primary care setting, neither case definition performed well as a screening test in younger children, as they showed low sensitivity and high specificity. Thus, when using either of the case definitions, particular attention should be paid to younger children. These results further emphasize the need for confirmatory dengue diagnostic methods. At present, laboratory-based testing remains

### Table 8. Diagnostic values of the traditional and revised WHO dengue case definitions by age, hospital-based study.

| WHO Dengue case definition | Sensitivity % (95% CI) | Specificity % (95% CI) | PPV % (95% CI) | NPV % (95% CI) |
|----------------------------|------------------------|------------------------|----------------|----------------|
| **Traditional case definition** |                         |                        |                |                |
| All patients               | 96.7 (95.1–97.9)        | 22.0 (18.2–26.1)       | 67.2 (64.3–70.1) | 80.0 (71.7–86.7) |
| Children <4 years old      | 87.4 (80.1–92.8)        | 50.4 (41.5–59.3)       | 61.9 (54.1–69.3) | 81.3 (71.9–89.1) |
| Children ≥4 years old      | 98.5 (97.2–99.3)        | 12.7 (9.2–16.9)        | 68.9 (65.7–71.9) | 81.3 (67.4–91.1) |
| **Revised case definition** |                         |                        |                |                |
| All patients               | 99.3 (99.2–100.0)       | 8.5 (6.0–11.5)         | 64.2 (61.3–67.0) | 88.1 (74.4–96.0) |
| Children <4 years old      | 96.6 (91.6–99.1)        | 22.5 (15.6–30.7)       | 53.5 (46.6–60.3) | 87.9 (71.8–96.6) |
| Children ≥4 years old      | 99.8 (99.1–100.0)       | 3.9 (2.0–4.7)          | 67.1 (63.9–70.1) | 92.3 (64.0–99.8) |

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essential both for dengue diagnosis and surveillance. The development of point-of-care tests based on the early detection of DENV antigens (such as the NS1 rapid test [12,31]) along with the discovery of new biomarkers for dengue could prove invaluable to enhancing dengue diagnosis, clinical management, and surveillance.

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Author Contributions

Conceived and designed the experiments: GG LG AB EH. Performed the experiments: GG MAP DE WA GK. Analyzed the data: GG LG AB EH. Contributed reagents/materials/analysis tools: AB EH. Wrote the paper: GG LG AB EH.

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