Temporal relationship between occurrences of hand, foot and mouth disease, respiratory virus detection and febrile seizures in children in tropical Singapore: a time-series analysis

Y. Chen[1], C. Y. Chong[2], A. R. Cook[1], N. T. W. Sim[3], P. Horby[4] and H. H. La[1]

[1]Saw Swee Hock School of Public Health, National University of Singapore and National University Health System, Singapore, Singapore; [2]Infectious Diseases, Department of Paediatrics, KK Women's & Children's Hospital, Singapore, Singapore; [3]Department of Pathology and Laboratory Medicine, KK Women's & Children's Hospital, Singapore, Singapore and [4]Centre for Tropical Medicine and Global Health, University of Oxford, Oxford, UK

Abstract

Febrile seizure (FS) in children is a common complication of infections with respiratory viruses and hand, foot and mouth disease (HFMD). We conducted a retrospective ecological time-series analysis to determine the temporal relationship between hospital attendances for FS and HFMD or respiratory virus infections. Epilepsy attendance was used as a control. Data from 2004 to 2012 FS and epilepsy hospital attendance, HFMD notifications to the Ministry of Health and from laboratory-confirmed viral respiratory infections among KK Women's and Children's Hospital inpatients were used. A multivariate linear regression analysis was conducted to evaluate the relationship between FS and the virus time series. Relative risks of FS by age were calculated using Bayesian statistical methods. Paediatric accident and emergency (A&E) attendances for FS were found to be associated with influenza A (extra 0.47 FS per influenza A case), B (extra 0.32 per influenza B case) and parainfluenza 3 (extra 0.35 per parainfluenza type 3 case). However, other viruses were not significantly associated with FS. None of the viruses were associated with epileptic seizure attendance. Influenza A, B and parainfluenza 3 viruses contributed to the burden of FS resulting in A&E attendance. Children at risk of FS should be advised to receive seasonal influenza vaccination.

Introduction

Febrile seizure (FS) is a frequent cause of emergency attendance in children. Worldwide, the prevalence of FS ranges from 2% to 5% [1, 2]. The young age and sudden onset leads FS to be a major cause of parental anxiety, resulting in emergency attendance [3].

Hand, foot and mouth disease (HFMD) is a highly prevalent paediatric disease in many Asian countries, caused by enteroviruses belonging to the genus of the Picornavirus family. Enterovirus genotypes enterovirus 71 (EV71) and coxsackievirus A16 (CA16) belonging to species A are the most common causes of HFMD worldwide. Other enterovirus types such as CA2, CA4, CA5, CA6, CA8, CA10 and CA12 from species A, CB2 and CB4 from species B and some echoviruses can cause HFMD as well [4]. Most HFMD infections are mild and self-limiting [5]. However, HFMD caused by EV71 infection is relatively severe, leading to occasional neurological involvement and death [6]. The age range for infection with HFMD coincides closely with that for FS: the latter occurring between 6 months and 6 years with a median age of 18 months [7], while most cases of HFMD occur prior to the start of formal schooling in China [8], Japan [9], Singapore [10] and other countries. FS has been more commonly observed in patients infected with EV71 [11]. However, FS has also been observed among patients with infection caused by coxsackieviruses such as CA2, CA6, CA10 and so on [12].

In addition, FS has been associated with infections with influenza, parainfluenza, adenovirus, rotavirus and human herpes viruses [13]. Influenza infection is usually self-limiting with recovery within a week in most people, but among people at high risk, including pregnant women, children under the age of 6, the elderly, individuals with chronic medical conditions and individuals with immunosuppressive conditions, and influenza can cause severe illness and even death [14]. It has been estimated that healthy children under the age of 5 have the highest influenza admission rate [15]. It has previously been demonstrated that influenza A infection is an important cause of FS [16].

In tropical Singapore, both HFMD and influenza are endemic and year-round transmission occurs [10]. HFMD and influenza outbreaks are irregular, compared to the annual or bi-annual outbreaks in temperate and subtropical countries [17]. HFMD is a notifiable disease, with notification of cases mandatory by both clinicians and preschools; in the latter, children

Table 1. Detailed yearly number of A&E and inpatient FS and epilepsy, and yearly number of HFMD cases from 2004 to 2012

Year	A&E FS	A&E epilepsy	Inpatient FS	Inpatient epilepsy	HFMD
2004	1199	352	640	233	10 641
2005	1206	363	693	259	16 888
2006	1203	372	720	289	20 282
2007	1320	431	761	314	23 279
2008	1170	379	701	259	29 605
2009	1255	385	661	326	16 810
2010	1300	426	748	395	30 709
2011	1395	465	704	363	20 599
2012	1364	438	178	274	36 986

A&E, Paediatric accident and emergency; FS, febrile seizures; HFMD, hand, foot and mouth disease.

are screened daily for symptoms and isolated for 10 days if infected. Epidemics of HFMD and other respiratory infections are frequent and, as such, Singapore offers a useful opportunity to assess the relative role of EVs and respiratory viruses on FS, which also exhibit temporal fluctuations both within and between years, with less potential for confounding due to seasonal synchronicity of other viruses than in temperate countries.

To assess the magnitude of association between FS and HFMD and respiratory viruses, we conducted a retrospective analysis of paediatric accident and emergency (A&E) attendance for FS at Singapore's largest paediatric hospital, KK Women's and Children's Hospital (KKH) – which admits about 55% of paediatric patients in Singapore, using KKH laboratory respiratory virus data (on influenza A and B, parainfluenza 1, 2 and 3, respiratory syncytial virus (RSV) and adenovirus) and Ministry of Health (MOH) surveillance data on HFMD disease from 2004 to 2012. The objective was to determine the temporal relationship between occurrences of HFMD cases, respiratory virus detection and FS among children.

Materials and methods

This was a retrospective time-series analysis of three sets of existing databases from January 2004 to December 2012: KKH A&E attendance and hospital admissions for FS; KKH Laboratory data of respiratory viruses and cases of HFMD notified to MOH.

Data sources

Three age-structured time-series data sets were created and merged. For the hospitalisation data set, raw, abstracted data from de-identified individual cases was grouped into weekly count data of A&E and hospital admission for FS from KKH records, together with epilepsy as a control condition that should not be affected by circulating viruses. The second data set consists of weekly counts of HFMD cases from the MOH data. The third data set contains weekly counts of specific respiratory viruses from KKH laboratory.

FS: KKH A&E created a database of extracted cases having the following principal or secondary diagnosis based on International Classification of Diseases (ICD) (9th edition) codes: 780.32 (complex FS); 780.31 (simple FS); 345.9 (epilepsy) and 345.8 (other forms of epilepsy and recurrent seizure) from 2004 to 2011. In 2012 where the more recent ICD-10 edition was used to code discharge data, cases

were extracted based on the corresponding ICD-10 code: R 56.8 (seizure/fit), R 56.0 (FS (simple or complex)), G 40.90, G 40.91, G 40.10, G 40.40, G 40.80 and G 40.11 (other epilepsy and recurrent seizures). Because the inpatient FS data for 2012 was not consistent with that of previous years, presumably due to the switch to ICD-10, we excluded it from analysis.

HFMD surveillance: HFMD cases were reported by physicians, educational institutions and laboratories. Age-stratified aggregate weekly counts of notified HFMD cases were obtained from the MOH.

Other virus surveillance: KKH laboratory had data on respiratory viruses detected from clinical specimens from children who were admitted to KKH for various conditions. The antigen detection tests identified a panel of respiratory viruses including Influenza A, B, parainfluenza 1,2,3, adenovirus and RSV tested by immunofluorescence and obtained by nasopharyngeal swabs of patients admitted for upper or lower respiratory tract infections. The sensitivity and specificity of immunofluorescence have been estimated to be 85.9% and 87.1%, respectively, when compared to standard viral culture [18].

Statistical analysis

Four multivariate linear regression analyses were conducted to investigate how much each virus contributed to FS or epilepsy hospital cases. The dependent variables were taken to be weekly counts of FS or epilepsy at A&E or inpatient admissions. Weekly counts of HFMD and other viruses were used as independent variables. The dependent variables were regressed with all potential viruses and significant viruses were identified at a significance level of 0.05. Only the significant viruses were included in the final multivariate regression models to measure the effect size of each virus.

Relative risks of FS and epilepsy by age were calculated using Bayesian approaches [19]. We approximated the number of children in each age group assuming the population to be constant (the number being unknown: the distribution at exact ages is known for the resident population at census years only, but the non-resident population is large in Singapore). We assigned a non-informative, conjugate Dirichlet prior for the risk of infection, then used Monte Carlo sampling to draw samples from the resulting Dirichlet posterior whereby the relative risk and corresponding 95% credible intervals were obtained.

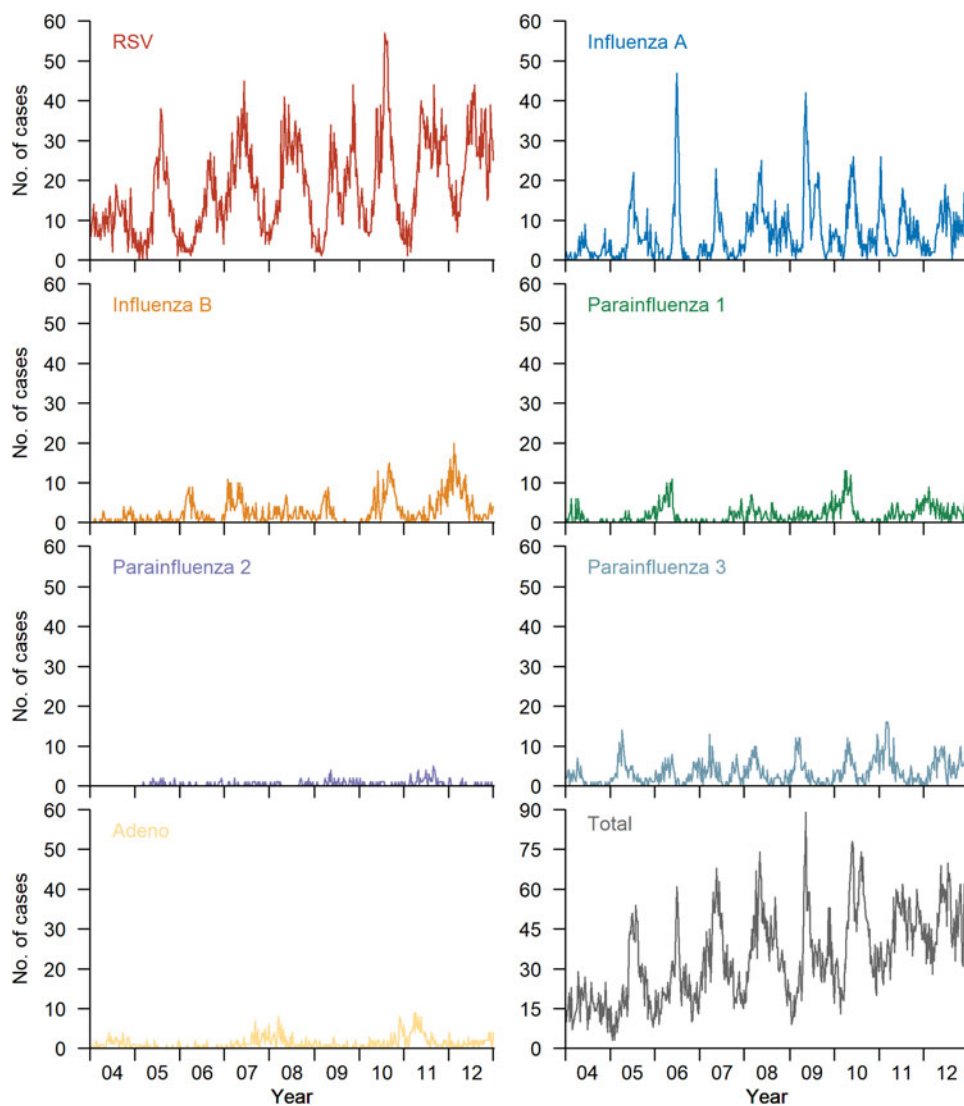Statistical analyses were performed using the R Statistical Software (version 3.1.1) [20].

Fig. 1. Weekly number of various respiratory viruses confirmed by KKH laboratory from 2004 to 2012. The bottom right panel shows the total number of viruses detected. All other panels show the number detected for each virus, respectively.

Table 2. Summary of A&E FS cases explained by all viruses, 2004–2012

Virus	Excess FS per case (95% CI)	P value
Influenza A	0.47 (0.39–0.55)	<0.0001*
Influenza B	0.32 (0.12–0.51)	0.001*
RSV	0.03 (−0.03 to 0.09)	0.35
Parainfluenza 1	0.17 (−0.08 to 0.43)	0.18
Parainfluenza 2	−0.33 (−1.08 to 0.43)	0.40
Parainfluenza 3	0.36 (0.17–0.56)	0.0003*
Adenovirus	0.034 (−0.33 to 0.40)	0.85
HFMD	−0.0010 (−0.004 to 0.002)	0.45

A&E, Paediatric accident and emergency; FS, febrile seizures; HFMD, hand, foot and mouth disease; RSV, respiratory syncytial virus.

Table 3. Summary of A&E FS cases explained by significant viruses, 2004–2012

Virus	Excess FS per case (95% CI)	P value
Influenza A	0.47 (0.40–0.55)	<0.0001
Influenza B	0.32 (0.16–0.49)	0.0001
Parainfluenza 3	0.35 (0.17–0.52)	0.0001

A&E, Paediatric accident and emergency; FS, febrile seizures.

Results

Yearly numbers of A&E attendance and admission for FS and epilepsy, as well as yearly number of HFMD cases, are detailed in Table 1. Among all incidences of FS, a total of 95% were younger than 6 years old, of whom, 12% were in the <1 year age group, 62% in the 1–2 year age group and 21% in the 2–5 year age group.

Weekly numbers of various respiratory viruses detected during 2004 and 2012 are shown in Figure 1. The bottom right panel shows the total number of viruses detected, whereas all other panels show the number detected for each virus, respectively.

FS was significantly associated with influenza A, influenza B and parainfluenza 3 epidemic activity (Table 2). No association was observed with parainfluenza 1 and 2, RSV, adenovirus and HFMD. The three contributing viruses were then used to form
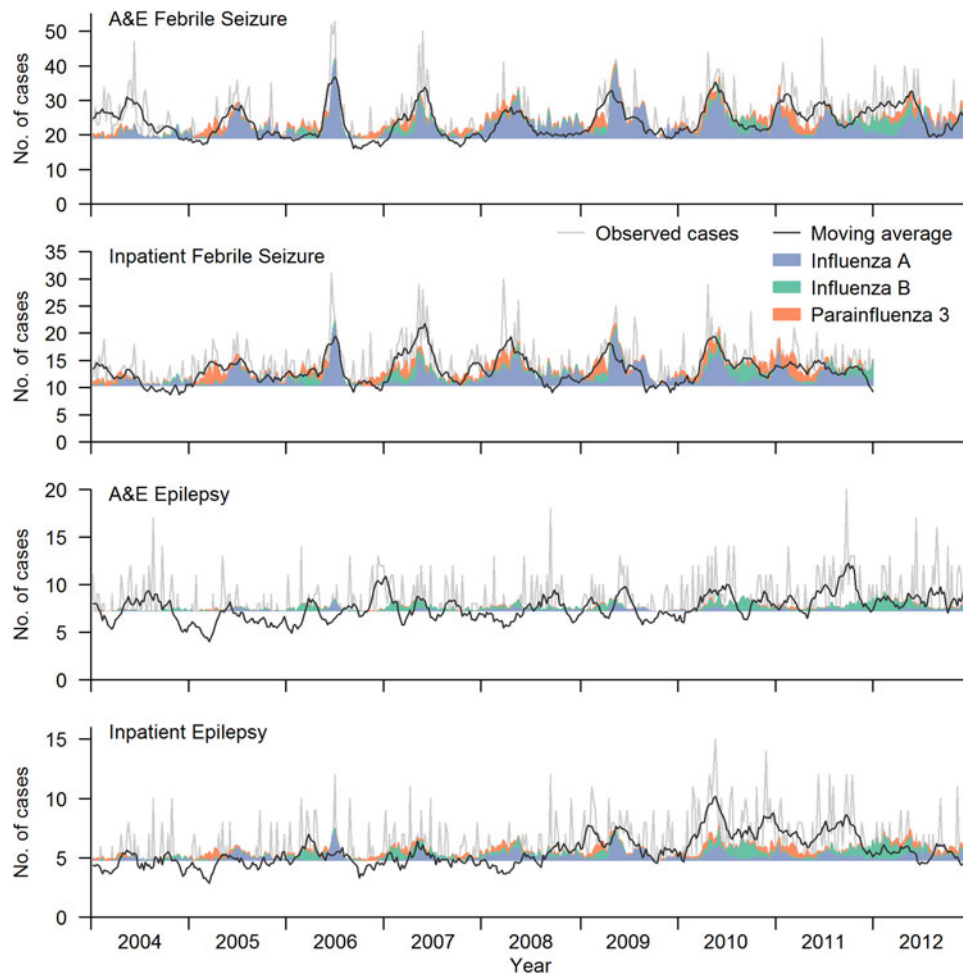
Fig. 2. Number of excess FS cases above baseline that can be explained by one extra case of each virus. Baseline level is indicated by the horizontal line. The excess number explained by influenza virus A, B and parainfluenza virus 3 are shown in blue, green and orange shaded regions, respectively. Observed time series is shown in light grey line in each panel, and a moving average of the number of cases (up to 4 weeks before and after) is overlaid as the black line on top.

a final multivariate model with similar estimates to the full model containing all the viruses (Table 3). In contrast, HFMD activity was not associated with an increase in A&E FS.

The multivariate model using influenza A, B and parainfluenza 3 was able to explain the majority of the excess FS cases above baseline for both inpatient and A&E FS (Fig. 2). Each influenza A case was associated with 0.47 (95% CI (0.40–0.55)) excess A&E FS (P-value<0.001), slightly more than influenza B (0.32, 95% CI (0.16–0.49), P-value = 0.001) and parainfluenza 3 (0.35, 95% CI (0.17–0.52), P-value = 0.001). In contrast, and as expected, there was no association between epilepsy attendance and any of the other viruses.

Relative risks for children of different ages are shown in Figure 3, using age 0 as baseline group. The relative risk of FS is higher than epilepsy for 1-year-old children, but it decreased rapidly as age increased. The incidence of epilepsy decreased slightly with increasing age and stabilised after 3 years of age.

Discussion

In contrast to our initial expectations, we did not find an association between HFMD incidence and FS. The data instead revealed a relatively strong association between FS and three respiratory viruses, two of which are vaccine preventable: influenza A and B and parainfluenza 3.

Possible reasons for children developing FS include the structural immaturity of a child's brain which is therefore more susceptible to seizures triggered by minor insults such as systemic viral infections [21]. This is supported by the age distribution of FS cases in our data, the incidence of which was the highest among younger patients, in contrast to epilepsy cases for which the relative risk was almost constant with age.

Although neurological involvement is one of the hallmarks of severe EV71 infection [22], and one of the major motivations behind the recent development of a vaccine in China, where more than 15 00 000 children are infected annually [23]. Cases of neurological complications due to EV71 infection have also been reported in Japan, Taiwan, South Korea and Australia [11, 24–26]. Indeed, the risk of severe outcomes has led to the implementation of school closures, and mandatory screening and isolation of infected cases in Singapore [27]. As a result of this well-established connection, we expected that FS might be associated with HFMD given the high incidence in Singapore, where between 24% and 32% are estimated to be infected with EV71 by the time they reach school age [28], and the ostensible presence of epidemics of FS cases over time, a feature indicative
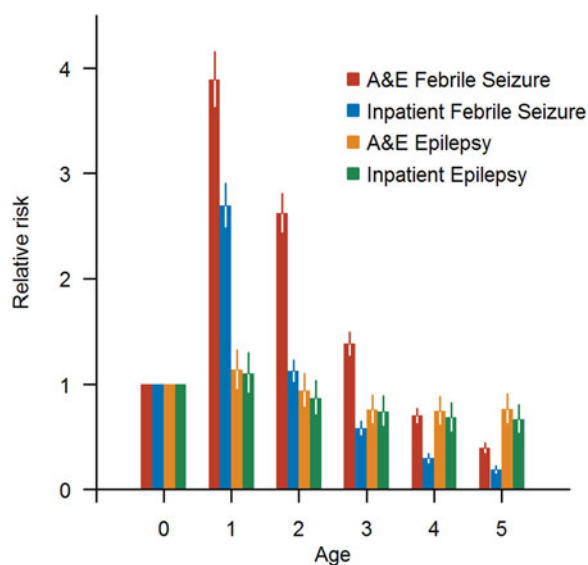
Fig. 3. Relative risk for children of different ages to have A&E/inpatient FS and A&E/inpatient epilepsy, respectively.

of an infectious aetiology. This association, however, was not established from our data.

On the other hand, for influenza, FS constitutes the majority of neurologic complications [29], followed by encephalopathy, encephalitis, acute necrotising encephalitis, Reye's syndrome, acute disseminated encephalomyelitis, transverse myelitis and Guillain–Barré syndrome [30]. Both mild and severe neurological complications have been reported to occur in children in other settings during the 2009 influenza A H1N1 pandemic [31]. In other case series, about 5–19% of children with pandemic H1N1 Influenza presented with neurologic manifestations [32]. FS is also common among children with seasonal influenza infection, occurring in ~20% of children hospitalised with influenza [30], while the incidence of A&E presentations for seizures increased by 6.7/1 00 000 among children under 6 years and that of ambulance calls for seizures increased by 3.2/1 00 000 [33]. Chiu *et al.* [16] found in a retrospective hospital-based study in Hong Kong, China, that the risk of FS was high with influenza and not with other respiratory viruses and was associated with repeated seizures in the same febrile episode.

Although less well studied, parainfluenza has also been associated with FS in a historic paper from England [34], and more recently [13] has been demonstrated to have a similar risk profile to influenza and adenovirus for FS, though in our study, adenovirus epidemic activity was not positively associated with FS. Given the lower incidence of parainfluenza 3 (among hospitalised cases), it has relatively lower impact than influenza A and B, both of which are widespread in Singapore and capable of causing epidemics throughout the year [35].

The incidence of FS has previously been estimated to vary across settings, from 0.5 to 1.5% in China, 2.2 to 2.3% in North America and 8.8% in Japan [36], and while relatively uncommon, the distress caused to families can be substantial. Given the not-insubstantial relationship with influenza epidemic activity demonstrated in this paper, future cost-effectiveness analyses for influenza vaccination or other interventions should consider paediatric FS as an outcome, both for the health and financial impacts. Influenza vaccination is recommended for young children

[37] but uptake rates are low – 27% for children between 6 months and 8 years old in the United States [38] and even lower in Singapore [39]. In Singapore, substantial effort is put into surveillance and control of HFMD outbreaks, but rather less on influenza among children. This study did not find any evidence that the former contributed to FS incidence. Given the mismatch between mortality and complications attributed to influenza and due to HFMD, there is an argument for reviewing control policies that include mandatory isolation of infected children for 10 days, daily screening, 'naming and shaming' schools with sustained outbreaks online and closing schools with outbreaks for disinfection.

The primary limitation of this study is that the HFMD data were based on notifications to MOH, and thus were not proven to be positive for enterovirus and may have included some cases of herpangina or misidentified herpes simplex stomatitis or other mistaken febrile exanthems. In addition, some HFMD cases may not be accounted for due to misdiagnosis of HFMD. As a retrospective study, the FS diagnoses extracted from ICD coding may potentially have led to some missing or inaccurate diagnoses. Another limitation is that the laboratory method for diagnosis of respiratory viruses was based on antigen detection test, which is less accurate than the gold standard nucleic acid amplification test [40]. The analysis was ecological, which limits the strength of inference, but this is countered by the close correlation of the fitted model to the FS data. It is unlikely this is due to overfitting as the same concordance is not observed for epilepsy incidence. Another limitation of this study was our inability to differentiate between first episodes and recurrent FS. We did not look at underlying neurologic disease or structural abnormalities in this cohort, and this would be a useful avenue for further research.

Conclusions

Excess FS was associated with influenza A and B and parainfluenza 3, but neither with HFMD nor other respiratory viruses. There was no association between epilepsy and any of the viruses. This age group is already recommended to receive seasonal vaccination and the association with FS strengthens the argument for routine seasonal influenza vaccination. A valuable extension to the work presented herein would be a prospective study that includes laboratory diagnosis of enterovirus and enterovirus typing to confirm the conclusion that HFMD associated with different genotypes was not associated with FS and to validate the association between influenza, parainfluenza 3 and FS.

References

1. **Verity CM, Butler NR and Golding J** (1985) Febrile convulsions in a national cohort followed up from birth. I--Prevalence and recurrence in the first five years of life. *British Medical Journal (Clinical Research Ed.)* **290**, 1307–1310.
2. **Kjeldsen MJ et al.** (2002) Genetic and environmental factors in febrile seizures: a Danish population-based twin study. *Epilepsy Research* **51**, 167–177.
3. **Baumer JH et al.** (1981) Many parents think their child is dying when having a first febrile convulsion. *Developmental Medicine and Child Neurology* **23**, 462–464.

4. **Xu M** *et al.* (2015) Genotypes of the enterovirus causing hand foot and mouth disease in Shanghai, China, 2012–2013. *PLoS ONE* **10**, e0138514.

5. **WPRO|Hand, Foot and Mouth Disease Information Sheet**. Available at http://www.wpro.who.int/emerging_diseases/hfmd.information.sheet/en/. (Accessed 23 May 2016).

6. **Lee T-C** *et al.* (2009) Diseases caused by enterovirus 71 infection. *The Pediatric Infectious Disease Journal* **28**, 904–910.

7. **Offringa M** *et al.* (1994) Risk factors for seizure recurrence in children with febrile seizures: a pooled analysis of individual patient data from five studies. *The Journal of Pediatrics* **124**, 574–584.

8. **Zhang Y** *et al.* (2010) An emerging recombinant human enterovirus 71 responsible for the 2008 outbreak of hand foot and mouth disease in Fuyang city of China. *Virology Journal* **7**, 94.

9. **Momoki ST** (2009) Surveillance of enterovirus infections in Yokohama city from 2004 to 2008. *Japanese Journal of Infectious Diseases* **62**, 471–473.

10. **Hand, Foot & Mouth Disease|Ministry of Health**. Available at https://www.moh.gov.sg/content/moh_web/home/diseases_and_conditions/h/hfmd.html. (Accessed 18 May 2016).

11. **McMinn P** *et al.* (2001) Neurological manifestations of enterovirus 71 infection in children during an outbreak of hand, foot, and mouth disease in Western Australia. *Clinical Infectious Diseases: An Official Publication of the Infectious Diseases Society of America* **32**, 236–242.

12. **Francis JR** *et al.* (2016) An observational study of febrile seizures: the importance of viral infection and immunization. *BMC Pediatrics* **16**, 202.

13. **Chung B and Wong V** (2007) Relationship between five common viruses and febrile seizure in children. *Archives of Disease in Childhood* **92**, 589–593.

14. **Ghebrehewet S, MacPherson P and Ho A** (2016) Influenza. *British Medical Journal* **355**, i6258.

15. **Cromer D** *et al.* (2014) The burden of influenza in England by age and clinical risk group: a statistical analysis to inform vaccine policy. *The Journal of Infection* **68**, 363–371.

16. **Chiu SS** *et al.* (2001) Influenza A infection is an important cause of febrile seizures. *Pediatrics* **108**, E63.

17. **Viboud C, Alonso WJ and Simonsen L** (2006) Influenza in tropical regions. *PLOS Medicine* **3**, e89.

18. **Matthey S** *et al.* (1992) Rapid detection of respiratory viruses by shell vial culture and direct staining by using pooled and individual monoclonal antibodies. *Journal of Clinical Microbiology* **30**, 540–544.

19. **Gelman A** *et al.* (2013) *Bayesian Data Analysis*. 3rd Edn. Boca Raton, FL: Chapman & Hall/CRC.

20. **R Core Team** (2014) *R: A Language and Environment for Statistical Computing*. Vienna, Austria: R Foundation for Statistical Computing.

21. **Jensen FE and Sanchez RM** (2002) Chapter 11 – Why does the developing brain demonstrate heightened susceptibility to febrile and other provoked seizures? In Baram TZ and Shinnar S (eds), *Febrile Seizures*. San Diego: Academic Press, pp. 153–168.

22. **Koh WM** *et al.* (2018) Severity and burden of hand, foot and mouth disease in Asia: a modelling study. *BMJ Global Health* **3**, e000442.

23. **Chinese Center for Disease Control and Prevention**. Available at http://www.chinacdc.cn/en/. (Accessed 20 May 2016).

24. **Komatsu H** *et al.* (1999) Outbreak of severe neurologic involvement associated with Enterovirus 71 infection. *Pediatric Neurology* **20**, 17–23.

25. **Ho M** *et al.* (1999) An epidemic of enterovirus 71 infection in Taiwan. Taiwan Enterovirus Epidemic Working Group. *The New England Journal of Medicine* **341**, 929–935.

26. **Ryu W-S** *et al.* (2010) Enterovirus 71 infection with central nervous system involvement, South Korea. *Emerging Infectious Diseases* **16**, 1764–1766.

27. **Siegel K, Cook AR and La H** (2017) The impact of hand, foot and mouth disease control policies in Singapore: a qualitative analysis of public perceptions. *Journal of Public Health Policy* **38**, 271–287.

28. **Ang L-W** *et al.* (2011) The changing seroepidemiology of enterovirus 71 infection among children and adolescents in Singapore. *BMC Infectious Diseases* **11**, 270.

29. **Ekstrand JJ** (2012) Neurologic complications of influenza. *Seminars in Pediatric Neurology* **19**, 96–100.

30. **Toovey S** (2008) Influenza-associated central nervous system dysfunction: a literature review. *Travel Medicine and Infectious Disease* **6**, 114–124.

31. **Blanton L** *et al.* (2012) Neurologic disorders among pediatric deaths associated with the 2009 pandemic influenza. *Pediatrics* **130**, 390–396.

32. **Calitri C** *et al.* (2010) Clinical features of hospitalised children with 2009 H1N1 influenza virus infection. *European Journal of Pediatrics* **169**, 1511–1515.

33. **Polkinghorne BG** *et al.* (2011) Relationship between the population incidence of febrile convulsions in young children in Sydney, Australia and seasonal epidemics of influenza and respiratory syncytial virus, 2003–2010: a time series analysis. *BMC Infectious Diseases* **11**, 291.

34. **Downham MA, McQuillin J and Gardner PS** (1974) Diagnosis and clinical significance of parainfluenza virus infections in children. *Archives of Disease in Childhood* **49**, 8–15.

35. **Influenza|Ministry of Health**. Available at https://www.moh.gov.sg/content/moh_web/home/diseases_and_conditions/i/influenza.html. (Accessed 3 April 2018).

36. **Hauser WA** (1994) The prevalence and incidence of convulsive disorders in children. *Epilepsia* **35**(Suppl. 2), S1–S6.

37. **Vaccination: Who Should Do It, Who Should Not and Who Should Take Precautions | Seasonal Influenza (Flu)|CDC**. Available at http://www.cdc.gov/flu/protect/whoshouldvax.htm. (Accessed 20 May 2016).

38. **Lu P** *et al.* (2013) Surveillance of influenza vaccination coverage--United States, 2007-08 through 2011-12 influenza seasons. *Morbidity and Mortality Weekly Report. Surveillance Summaries (Washington, D.C.: 2002)* **62**, 1–28.

39. **Ang LW** *et al.* (2014) Influenza-associated hospitalizations, Singapore, 2004–2008 and 2010–2012. *Emerging Infectious Diseases* **20**, 1652–1660.

40. **Ginocchio CC and McAdam AJ** (2011) Current best practices for respiratory virus testing. *Journal of Clinical Microbiology* **49**, S44–S48.