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# Temporal epidemiologic trends and molecular characteristics of hand, foot, and mouth disease in Zhejiang Province, 2016-2025: Evidence from a 10-year surveillance dataset

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# Temporal epidemiologic trends and molecular characteristics of hand, foot, and mouth disease in Zhejiang Province, 2016-2025: Evidence from a 10-year surveillance dataset

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## Abstract

### Background

Hand, foot and mouth disease (HFMD) is highly prevalent in the mainland China. It can be caused by more than 20 different enteroviruses (EVs). The dominant enterovirus serotypes for HFMD cases have been undergoing continuous shifts. Zhejiang has been reported as high-incidence province. This study aims to explore on its epidemiology, etiology characteristics in recent years.

### Methods

Epidemiological and laboratory data on hand, foot, and mouth disease (HFMD) cases from 2016 to 2025 were collected. Age, gender, periodicity, and enterovirus (EV) serotypes were analyzed in this study. RT-PCR or real-time fluorescent quantitative RT-PCR was used to confirm the serotype of each laboratory-confirmed case. The age-period-cohort (APC) model was applied to examine the temporal trends of incidence from three dimensions—age, period, and birth cohort, while wavelet analysis was used to characterize the periodic patterns of the major HFMD serotypes, including enterovirus A71(EV-A71), Coxsackievirus A16(CV-A16), Coxsackievirus A6(CV-A6), Coxsackievirus A10(CV-A10) and other EV.

### Results

A total of 1,223,560 HFMD cases were reported in Zhejiang Province from 2016 to 2025, including 1,223,414 mild cases, 137 severe cases and 9 fatal cases. The 2-<5 years age group constituted the largest share of mild cases, whereas the 1-<2 years and 0-<1 years age groups accounted for the highest proportions of severe and fatal cases, respectively. A total of 60,620 HFMD cases were enrolled for etiological surveillance, of which 38,282 showed positive results. Among the 38,161 positive mild cases, CV-

A6 was the predominant serotype among the positive mild cases, while EV-A71 prevailed among positive severe and fatal cases. Children aged 1-<2 years and infants aged 0-<1 year represented the highest risk for mild HFMD and fatal outcomes, respectively. A distinct 12-month annual periodic epidemic cycle was identified for all enterovirus serotypes. During 2016-2025, the predominant pathogens causing mild HFMD have shifted from other EVs to CV-A6 and CV-A16. EV-A71 has been progressively superseded by CV-A6, CV-A16 and other EVs among severe and fatal HFMD cases.

**Conclusion**

The evolving etiology of pediatric HFMD and the increasing age of susceptibility highlight the need for continuous surveillance, as well as the urgent development of a multivalent HFMD vaccine to enhance prevention and strengthen public health responses.

**Key words:** Hand, foot and mouth disease, enteroviruses, Epidemiology, Periodicity

Hand-foot-mouth disease (HFMD) is a common infectious disease in children caused by various human enteroviruses, which predominantly affects children under 5 years old. Most infected individuals present with mild symptoms, mainly manifested as fever, rashes and herpes on the hands, feet and mouth. The clinical phenotypes of HFMD consist of asymptomatic infections and symptomatic cases. A small number of infections may develop neurological complications including aseptic meningitis, encephalitis, acute flaccid paralysis and neurogenic pulmonary edema. In particular, children under 3 years of age infected with enterovirus 71 (EV-A71) are prone to severe complications involving the central nervous system, cardiopulmonary system, and even death[1, 2].

Since the 1990s to the 2000s, severe HFMD cases caused by enterovirus EV-A71 have been reported in Japan, Singapore, Malaysia and other regions and countries[3]. Thereafter, the prevention and control of HFMD had become a major public health concern in the Asia-Pacific region. The per capita economic burden is 25.58 US dollars for untreated individuals, 366.50 US dollars for patients with mild symptoms, and 2355.89 US dollars for those with severe cases[4]. In addition, the hospitalization rate among symptomatic HFMD cases was 6% (95%CI: 2.8%–14.9%), and 18.7% (95%CI: 6.7%–31.5%) of these cases developed complications[5]. It is estimated that HFMD causes 96,900 (95%CI: 40,600–259,000) age-weighted disability-adjusted life years (DALYs) annually across eight high-burden countries in East and Southeast Asia.[5]，Most of these DALYs were attributable to years of life lost due to premature death. In

2008, China designated HFMD as a category C notifiable infectious disease for statutory surveillance and reporting management[6]. According to data from the Chinese Center for Disease Control and Prevention (China CDC), both the annual incidence and mortality rates of HFMD in China ranked among the top three among all Category C infectious diseases from 2009 to 2022[7].

In 2008, mainland China experienced the first outbreak of severe, highly clustered and rapidly fatal pediatric cases caused by EV-A71, with approximately 12.4% of hospitalized cases progressing to severe illness or death[8]. This situation aroused great attention from the Chinese government, which further promoted the research and application of HFMD vaccines. At present, the EV-A71 vaccine is the only officially licensed vaccine against HFMD in China. Three inactivated EV-A71 vaccines were approved and launched for population vaccination in 2015, January 2016 and late 2016, respectively[9-11]. Meanwhile, these vaccines were gradually rolled out in Zhejiang Province.

Zhejiang Province is located in southeastern China, spanning from 118°E to 123°E in longitude and 27°N to 32°N in latitude. It has 11 prefecture-level cities and 90 counties. Situated in the central subtropical zone, Zhejiang features a humid monsoon climate with mild temperatures, abundant sunshine and plentiful rainfall. Owing to its warm climatic conditions and sound economic development, the number of HFMD cases has risen rapidly in Zhejiang, making it one of the high-prevalence provinces of HFMD across mainland China[12].

Several studies have demonstrated that the EV-A71 vaccine confers a high protective efficacy against HFMD. Real-world studies have further verified its satisfactory protective effect against severe HFMD cases[13]. With the implementation of vaccination and the influence of other factors, the epidemiological characteristics of HFMD have changed in Zhejiang Province. In this study, we adopted HFMD case data and etiological surveillance data over the past decade (2016–2025) to identify predominant enterovirus serotypes and comprehensively explore the temporal dynamics of age-specific disease patterns. The findings will provide valuable insights for optimizing integrated prevention and control strategies as well as facilitating the development of multivalent HFMD vaccines.

## Materials and Methods

### 1. Study Design

This retrospective study was conducted based on data obtained from the National Notifiable Disease Surveillance System. All HFMD cases diagnosed in outpatient and

inpatient departments of community hospitals, secondary or tertiary hospitals across Zhejiang Province from January 1, 2016 to December 31, 2025 were included. Available data encompassed demographics (sex, birth date, residential location), clinical grouping (mild/severe), onset, confirmation and death dates (when recorded). All cases were classified as either clinically diagnosed or laboratory-confirmed.

**2. Case Definition**

In accordance with the Diagnostic Criteria for Hand-foot-mouth Disease (WS588—2018), clinically diagnosed HFMD cases were defined as patients with acute onset, maculopules and herpes on the hands, feet and buttocks, scattered herpes on the oral mucosa or isthmus faucium, accompanied by symptoms such as cough, rhinorrhea, poor appetite and diarrhea, with or without fever, and excluding other related diseases. Laboratory confirmed cases were defined as cases meeting the above clinical diagnostic criteria and testing positive via etiological or serological examinations.

Severe cases were defined as those presenting with central nervous system involvement, including lassitude, lethargy, startle response, delirium, headache, vomiting, limb tremor, myoclonus, nystagmus, ataxia, acute flaccid paralysis and convulsions, as well as visible meningeal irritation signs and decreased or absent tendon reflexes. Mild cases referred to all confirmed HFMD cases excluding severe and fatal cases.

To minimize duplicate reports, repeated records of the same child (identified by ID number) reported by different medical institutions during a single illness episode were excluded.

**3. Specimen Collection and Detection**

Serotyping data for laboratory-confirmed HFMD cases were obtained from Zhejiang's provincial HFMD-specific surveillance network. In accordance with the Guidelines for Prevention and Control of Hand-foot-mouth Disease (2009 Edition)[14]. All severe and fatal HFMD cases were mandatorily subjected to laboratory testing. For mild HFMD cases, local CDC selected a certain number of cases to collect clinical specimens for detection, including throat swabs, anal swabs, stool specimens, vesicle fluid swabs or other relevant samples. Laboratory tests were completed within 24 hours after specimen collection using conventional RT-PCR or real-time fluorescent quantitative RT-PCR.

In this study, all specimens were subtyped for EV-A71, Coxsackievirus A16(CV-A16) and other EVs by HFMD surveillance network laboratories in 2016. From 2017 to 2025, laboratory typing was performed to identify EV-A71, CV-A16, Coxsackievirus A6(CV-A6), Coxsackievirus A10(CV-A10) and other EVs.

## 4. Data Analysis

Cases were stratified into five age groups: under 1 year, 1 to less than 2 years, 2 to less than 5 years, 5 to less than 10 years, and 10 years and above. Categorical variables including gender, age group and pathogen serotype were described by case number (n) and constituent ratio (%). Continuous variables were presented as median and interquartile range [M (Q1, Q3)]. Chi-square test was applied to compare differences among mild, severe and fatal case groups.

The age-period-cohort (APC) model is widely used in sociological and epidemiological studies. Based on the Poisson distribution, the model reflects the temporal trends of incidence or mortality from three dimensions: age, period and birth cohort. Age effect refers to the differences in HFMD incidence across age groups caused by aging-related factors. Period effect represents the impact of artificial factors such as advances in diagnostic techniques on disease incidence. Cohort effect indicates incidence variations among populations born in different years due to diverse exposure levels to risk factors. In this model age was divided into 26 groups ranging from 0–10 years(1-year strata) to 15–85 years(5-year grouping). Intrinsic estimator (IE) method was adopted to estimate the net effects of the three dimensions[15], relative risk (RR) and 95% confidence intervals were calculated based on estimated coefficients to quantify the impacts of age, period and cohort factors on HFMD incidence.

Morlet wavelet function was applied to conduct wavelet analysis on monthly HFMD cases in Zhejiang Province. Monthly case counts were logarithmically transformed and standardized to a mean of 0 and variance of 1. Continuous wavelet transform (CWT) was performed on processed data, followed by significance test[16]. Wavelet analysis enables multi-scale decomposition of non-stationary time series in both time and frequency domains. It can effectively identify the dominant epidemic cycles, seasonal characteristics and dynamic variations of HFMD prevalence intensity, and clearly reveal long-term trends and short-term fluctuation patterns.

Analyses of wavelet analysis, APC model and the map figure were performed using R software (version 4.2.0, http://www.r-project.org). The base layers of the map were extracted by the National Geographic Information Public Service Platform (Tianditu): https://cloudcenter.tianditu.gov.cn/administrativeDivision/. The employed packages included WaveletComp (version 1.2), Epi (version 2.64) and sf (version 1.0-20). $P< 0.05$ was considered statistically significant.

# Results

## 1. The temporal, geographical distribution and population differences

A total of 1,223,560 HFMD cases were reported in Zhejiang Province from 2016 to 2025, including 1,223,414 mild cases, 137 severe cases and 9 fatal cases. The highest annual incidence rate reached 433.57 per 100,000 population in 2018, while the lowest rate of 95.91 per 100,000 population occurred in 2022. The severe case rate (defined as the proportion of severe cases among all reported annual cases) peaked at 0.047% in 2016 and dropped to zero in 2024. Except for 2016 and 2017, the annual severe case rate remained below 0.01% (Figure 1-A). Single autumn epidemic peaks (weeks 40-50) were observed in 2017, 2020 and 2025, whereas summer peaks (weeks 20-35) appeared in the remaining years (Figure 1-B). Among reported HFMD cases, the proportion of children aged 5–10 years and over 10 years increased continuously after 2020, rising from 11.75% in 2020 to 28.58% in 2025 (Figure 1-C). In terms of occupational distribution, the percentage of student cases grew annually from 5.12% in 2020 to 17.16% in 2025. By contrast, the proportion of home-based children declined gradually, dropping from 66.54% in 2016 to 32.14% in 2024 (Figure 1-D).

Males predominated over females among mild, severe and fatal cases (Table 1). The median age of mild cases was 2 years old, which was higher than that of severe and fatal cases, both with a median age of 1 year old. The 2-<5 years age group accounted for the largest proportion in mild cases, while the 1-< 2 years group and the 0-<1 years group had the highest proportion in severe and fatal cases respectively. Statistically significant differences in age distribution were observed among groups ($P<0.001$). A total of 60,620 HFMD cases were included in etiological surveillance, of which 38,282 tested positive in laboratory examinations. CV-A6 was the predominant serotype among 38,161 positive mild cases. EV-A71 dominated in positive severe cases (114 cases) and fatal cases (7 cases), accounting for 39.42% and 66.67% respectively. Significant differences were found in serotype distribution among the three groups ($P<0.001$) (Table 1).

From 2016 to 2025, the annual HFMD incidence rate of prefecture-level cities in Zhejiang Province ranged from 20.23 per 100,000 population(Zhoushan City in 2020) to 608.89 per 100,000 population (Ningbo City in 2018). From 2016 to 2019, the high-incidence areas mainly concentrated in eastern Zhejiang. The high-prevalence regions shifted to southern and central areas in Zhejiang between 2023 and 2025. Ningbo ranked first in incidence rate in three years including 2016, 2018 and 2021. Jinhua reported the highest annual incidence in 2024 and 2025. As an island city, Zhoushan maintained the lowest incidence in most years except 2017 and 2023 (Figure 2).

**2. Age-Period-Cohort (APC) Effects**

Figure 3 presents the age, period and cohort effects on HFMD morbidity and mortality,

with shaded areas representing 95% confidence intervals. The HFMD infection risk peaked in the 1-year-old group, then declined rapidly and leveled off near zero after the age of 10 (Figure 3-A). The period effect fluctuated throughout the study period, reaching a peak in 2018, followed by an overall fluctuating downward trend and a slight rebound after 2021 (Figure 3-B). The cohort effect indicated that the effect of HFMD incidence was close to zero among populations born before 2010, then effect size rose rapidly in cohorts born between 2010 and 2018, and reached the maximum in 2018, followed by a sharp decline. It maintained a fluctuating upward trend at a high level from 2020 to 2025, implying that newly born cohorts had markedly higher infection risk than earlier cohorts (Figure 3-C).

**3. Seasonal and Periodic Characteristics**

Figure 4 displays the wavelet power spectra of total cases, EV-A71, CV-A16, CV-A6, CV-A10 and other EVs cases. The epidemic intensity of various enterovirus serotypes varied across years. EV-A71 predominated from 2016 to 2018, other EVs prevailed during 2016 to 2020, while CV-A6, CV-A16 and CV-A10 became the dominant serotypes from 2021 to 2024, 2023-2025 and 2021-2025, respectively. The annual periodicity of EV-A71 cases was no longer statistically significant after 2018(Figure 4-B). CV-A16, CV-A6 and CV-A10 cases exhibited distinct annual periodicity from 2020 to 2025 (Figure 4-C, 4-D, 4-E). Strong signals were detected for other enteroviruses before 2020 (Figure 4-F).

Wavelet transformation results of total cases and cases caused by five pathogens revealed a significant 12-month annual cycle in all groups ($P<0.05$). All serotypes presented a 6-month semi-annual cycle except CV-A6. EV-A71 and CV-A10 cases were dominated by the 12-month cycle, followed by the semi-annual cycle, with weak medium and long periodic(16-32 months) signals. Total cases, CV-A16, CV-A6 and other pathogens showed obvious medium-and-long-term epidemic patterns of 16–32 months ($P<0.05$) (Figure 5).

**4. Evolutionary Characteristics of Hand, Foot and Mouth Disease Pathogen Spectrum**

The predominant pathogen serotypes of mild HFMD cases varied annually. EV-A71 serotypes prevailed from 2016 to 2018, during which EV-A71 cases accounted for 93.60%(1740/1859) of all EV-A71 infections in the whole study period. Other EVs dominated in 2019, CV-A6 became the dominant serotype from 2020 to 2023 and 2025, and CVA16 was predominant in 2024 (Figure 6-A, 6-B). Among severe cases, 104 cases were reported between 2016 and 2017, accounting for 75.91% of the total severe cases during the study period. The proportions of severe cases caused by EV-A71 were

47.73%, 68.75% and 100% in 2016, 2017 and 2019 respectively. Severe cases declined markedly after 2020, with an annual average of three cases from 2020 to 2025(Figure 6-C). EV-A71 was the leading pathogen responsible for fatal HFMD cases, making up 66.67% of all deaths. Among the remaining three fatal cases, one was infected with other EVs and two belonged to clinically diagnosed untyped cases (Figure 6-D).

**Disscussion**

This study analyzed the etiological epidemiology, characteristics and epidemic patterns of different pathogen serotypes among mild, severe and fatal HFMD cases in Zhejiang Province over the past decade. The average annual HFMD incidence rate was 203.49 per 100,000 population from 2016 to 2025, higher than the national average of 105.72 per 100,000 population. The proportions of severe and fatal cases were 0.011% and 0.001% respectively, both lower than the national levels of 0.69% and 0.014% recorded from 2008 to 2023.Noticeable shifts were observed in the HFMD pathogen spectrum, and young children constituted the high-risk population. The dataset provides updated evidence for the evolutionary trend of HFMD pathogen spectrum in eastern China.

Three inactivated EV-A71 vaccines, developed by the Institute of Medical Biology at the Chinese Academy of Medical Sciences, Sinovac Biotech Co., Ltd., and the Wuhan Institute of Virology, were successively launched in China. These enterovirus 71 (EV-A71) vaccines can effectively prevent mild and severe cases of HFMD caused by EV-A71 infection[17]. This study found that starting in 2018, the proportion of EV-A71 infection cases in Zhejiang Province decreased significantly, and concurrently, the number of severe cases caused by EV-A71 infection also declined sharply. Vaccination against EV-A71 has driven a shift in the serotype composition of HFMD pathogens in Zhejiang Province. The relative proportions of CV-A6 and CV-A16 in the HFMD pathogen spectrum increased, replacing EV-A71 as the main causative agents of HFMD, this trend is consistent with the findings of relevant studies conducted in Henan Province[17] and Japan[18]. However, due to the lack of cross-immunity conferred by the EV-A71 vaccine, the overall incidence of hand, foot, and mouth disease (HFMD) in Zhejiang Province remains at a relatively high level, indicating that an effective immune barrier has not yet been established in the population. This finding is consistent with the results of studies conducted in Shandong Province[19] and Shanghai[21].

Two seasonal epidemic peaks were observed in most years during the study period, which is consistent with the seasonal epidemic characteristics reported in regions such as Sichuan, Yunnan, and Xinjiang[20-22].In this study, summer (typically weeks 20–35) was the major peak season for HFMD, while in individual years (specifically 2017,

2020, and 2025 in this study), the autumn peak (typically weeks 40–50) became the dominant epidemic season, which may be attributed to the influence of multiple complex factors during those particular years. Of which, the single autumn epidemic peak observed in 2017 was possibly attributable to large-scale population-based EV-A71 vaccination, which drove shifts in the circulating HFMD serotypes. Besides, the substantial decline in the incidence of HFMD from 2020, as well as the delay in the epidemic peak, were most likely attributed to the stringent non-pharmaceutical interventions implemented to prevent the spread of COVID-19, the closure of kindergartens and schools effectively contained HFMD outbreaks[23]. Additionally, the autumn peak of HFMD becoming the single epidemic peak in 2025 may be attributable to abnormal climatic conditions such as higher monthly average temperature and monthly cumulative rainfall[21].

The analysis of age distribution showed that the peak incidence of HFMD in Zhejiang Province in this study was concentrated in the children aged 0–5 years, which is generally consistent with the overall epidemic situation in mainland China[24]. The results of APC model revealed that the effect on the incidence of HFMD decreased with increasing age. The risk of incidence peaked in the 1-year-old age group, whereas the risk of death was highest in the 0-year-old group, indicating that the highest risk of death occurs during early infancy, this is attributable to the relatively low immunity levels in infants, making them more susceptible to fatal outcomes following HFMD infection[25]. Additionally, this study found that the proportion of HFMD cases aged over 5 years and the proportion of student cases exhibited an increasing trend, which was consistent with the findings of a previous study in Shandong Province[19]. This may be attributed to the fact that children under 3 years old are more susceptible to EV-A71, while CV-A6, CV-A10 have stronger infectivity among older and school-age children, thereby increasing the proportion of cases aged over 5 years[26, 27]. On the other hand, during the COVID-19 pandemic (2020–2022), young children in childcare facilities stayed at home under quarantine and failed to develop protective immunity, leading to a growing pool of susceptible individuals. When these children entered primary school, HFMD infections occurred, which ultimately drove the increase in the proportion of patients aged over 5 years.

Central nervous system symptoms are common in severe and fatal HFMD cases, which may lead to long-term sequelae[28]. Previous studies have demonstrated that EV-A71 infection elevates the level of exosomes in the serum of patients with severe HFMD. These exosomes can facilitate viral replication and trigger cytokine storms, thereby worsen disease progression[29, 30]. This study revealed that EV-A71 was the predominant serotype among severe and fatal HFMD cases in 2016–2017 and 2019,

whereas other EVs prevailed during 2018 and 2020–2025. Accordingly, monovalent EV-A71 vaccines can hardly meet the current demands of HFMD prevention. It is imperative to explore the genetic recombination and evolution of diverse enteroviruses and develop multivalent vaccines for effective control of severe and fatal HFMD cases.

Highly transmissible diseases require a continuous influx of susceptible individuals to sustain semi-annual or annual epidemics. In regions with low fertility rates, the number of young susceptible children increases slowly, which slows down viral circulation and tends to produce long epidemic cycles of 2 to 3 years. In areas with extensive population mobility and dense susceptible children, viral transmission accelerates and epidemic frequency rises remarkably. Studies have shown that Japan and Malaysia[31] exhibit annual epidemic patterns, which are consistent with those observed in northern China. By contrast, semi-annual epidemic cycles have been reported in Hong Kong of China[32],southern Taiwan of China[33] and Vietnam[34] in line with the epidemiological characteristics of southern China. Our study demonstrated that HFMD displays stable annual (12-month) seasonal epidemics, distinct short-term (6-month) and medium-to-long-term (16–32 month) epidemic cycles were observed among specific serotypes. These variations may stem from differences in viral shedding duration and transmission efficiency across serotypes. Infections caused by CV-A6 and CV-A10 are characterized by prolonged viral shedding, a higher proportion of asymptomatic and mild cases, and accordingly more persistent epidemic activity[35]. Other studies have suggested that the periodic patterns of HFMD may also be complicated by interferences between different serotypes of pathogenic enteroviruses[31].

In summary, we conducted a retrospective study on the demographic characteristics, serotype distribution and epidemic periodicity of HFMD in Zhejiang Province over the past decade. Our results indicated that HFMD remained highly prevalent in this region, while the rates of severe cases and mortality stayed relatively low. Children aged 1-<2 years and infants age 0-<1 year were the high-risk groups for mild HFMD and fatal cases, respectively. A prominent 12-month annual epidemic cycle was observed across all enterovirus serotypes. While a 6-month semi-annual cycle was present in all serotypes except CVA6. The dominant enterovirus serotypes for mild and severe HFMD cases have been undergoing continuous shifts during the study period. The predominant pathogens of mild cases have transitioned from other EVs to CV-A6 and CV-A16. For severe and fatal infections cases, EV-A71 has been gradually replaced by CV-A6, CV-A16 and other EVs.

Accordingly, targeted research and development of multivalent combined HFMD vaccines are essential to match the currently circulating serotypes and further reduce

the overall incidence as well as the risks of severe illness and death. These changes can further alter viral transmissibility and epidemic patterns in the population. Therefore, population-based surveillance and in-depth research on virological characteristics are of great scientific significance.

## Abbreviations

HFMD, Hand Foot and Mouth Disease, EV-A71, enterovirus 71, CV-A16, Coxsackievirus A16, CV-A6, Coxsackievirus A6, CV-A10, Coxsackievirus A10.

## Ethics approval and consent to participate

This investigation was undertaken in response to a public health emergency and approved by the ethics committee of Zhejiang Provincial Centre for Disease Control and Prevention (protocol code: 2016-020). Informed consent was obtained from all participants or their legal guardians. The study was conducted in accordance with the Declaration of Helsinki.

## Consent for publication

Consent for publication was not applicable because all data were fully anonymized and no identifiable individual information was included in this manuscript.

## Availability of data and materials

The datasets generated and analysed during the current study are not publicly available due to the confidential nature of notifiable infectious disease surveillance data, but are available from the corresponding author on reasonable request.

## Competing Interests

All authors declare no competing interests.

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## Authors' contributions

Wanwan Sun: Conceptualization, Data curation, Formal analysis, Software, Visualization, Writing – original draft

Yijuan Chen: Data curation, Investigation

Ziping Miao: Investigation, Resources Fudong Li: Validation, Methodology

Qiong Ge: Investigation, Data collection

Yanli Cao: Resources, Validation

Jimin Sun: Methodology, Writing – review & editing

Juan Hou*: Supervision, Project administration, Writing – review & editing

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**Captions**

Figure 1 Population and seasonal distribution of hand, foot and mouth disease from 2016 to 2025. (A) Epidemic trends of mild and severe cases annually, (B)Weekly epidemic trend of total cases, (C)Age-group distribution characteristics of annual total cases, (D)Population classification characteristics of annual total cases.

Figure 2 The incidence rates of HFMD across cities in Zhejiang Province, China, from 2016 to 2025. The numbers represent the following cities: 1. Hangzhou, 2. Ningbo, 3. Wenzhou, 4. Jiaxing, 5.Huzhou,6.Shaoxing,7.Jinhua,8.Quzhou,9.Zhoushan,10.Taizhou,11.Lishui.

Figure 3 Age-period-cohort analysis on incidence and mortality of HFMD. (A-C)The effects on incidence, (D-F) The effects on incidence mortality.

Figure 4 Wavelet spectrum analysis of HFMD cases. (A) Total cases, (B) EV-A71 cases, (C) CV-A16 cases, (D) CV-A6 cases, (E) CV-A10 cases, (F) Other enterovirus cases.

Figure 5 The average power of HFMD cases. (A) Total cases, (B) EV-A71 cases, (C) CV-A16 cases, (D) CV-A6 cases, (E) CV-A10 cases, (F) Other enterovirus cases.

Figure 6 Serotype composition of enteroviruses in HFMD cases by severity in Zhejiang, China, 2016–2025. (A) All cases by month. (B) Mild cases. (C) Severe cases. (D) Fatal cases.

**Table 1 Demographic and serotype characteristics of hand, foot, and mouth disease cases by clinical type.**

| Variable | Mild | Severe | Death | P value |
|---|---|---|---|---|
| | **N=1,223,414** | **N=137** | **N=9** | |
| **Gender** | | | | |
| Male | 721,000(58.93) | 83(60.58) | 5(55.56) | 0.906 |
| Female | 502,414(41.07) | 54(39.42) | 4(44.44) | |
| **Age (median, IQR)** | 2(1,4) | 1(1,2) | 1(0.92,2) | <0.001 |
| 0-<1y | 89,707(7.33) | 23(16.79) | 4(44.44) | <0.001 |
| 1-<2y | 310,339(25.37) | 55(40.15) | 2(22.22) | |
| 2-<5y | 590,851(48.30) | 54(39.42) | 3(33.34) | |
| 5-<10y | 199,118(16.28) | 4(2.92) | 0(0.00) | |
| >=10y | 33,399(2.73) | 1(0.72) | 0(0.00) | |
| **Pathogen typing of laboratory-confirmed cases** | **N=60474** | **N=137** | **N=9** | |
| EV-A71 | 1693(2.80) | 54(39.42) | 6((66.67) | <0.001 |
| CV-A16 | 10,439(17.26) | 6(4.38) | 0(0.00) | |
| CV-A6 | 14,404(23.82) | 7(5.11) | 0(0.00) | |
| CV-A10 | 1,353(2.24) | 0(0.00) | 0(0.00) | |
| Other EVs | 10,272(16.98) | 47(34.31) | 1(11.11) | |
| Negative | 22313(36.90) | 23(16.79) | 2(22.22) | |

EV7-A1 = enterovirus A71. CV-A16 = Coxsackievirus A16. CV-A6 = Coxsackie virus A6. CV-A10 = Coxsackie-virus A10, Other EVs= Other enterovirus.

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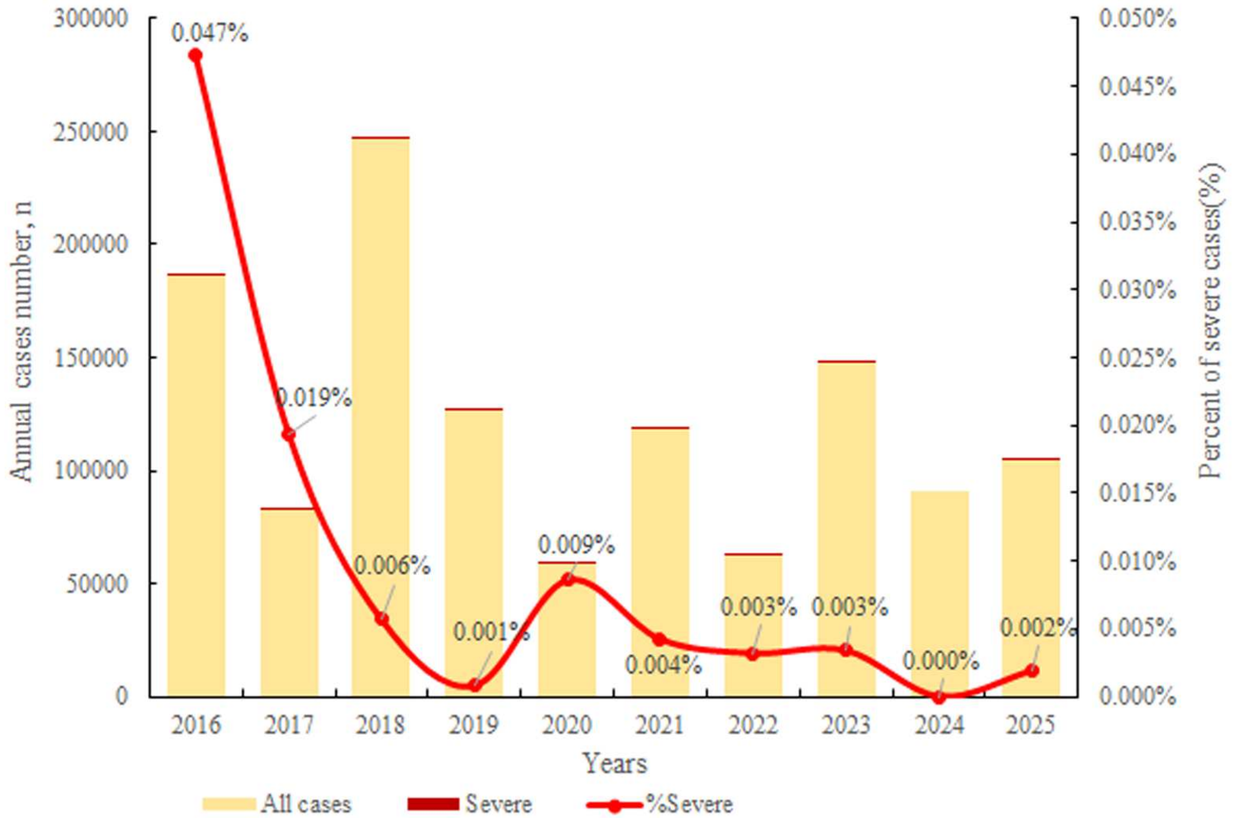
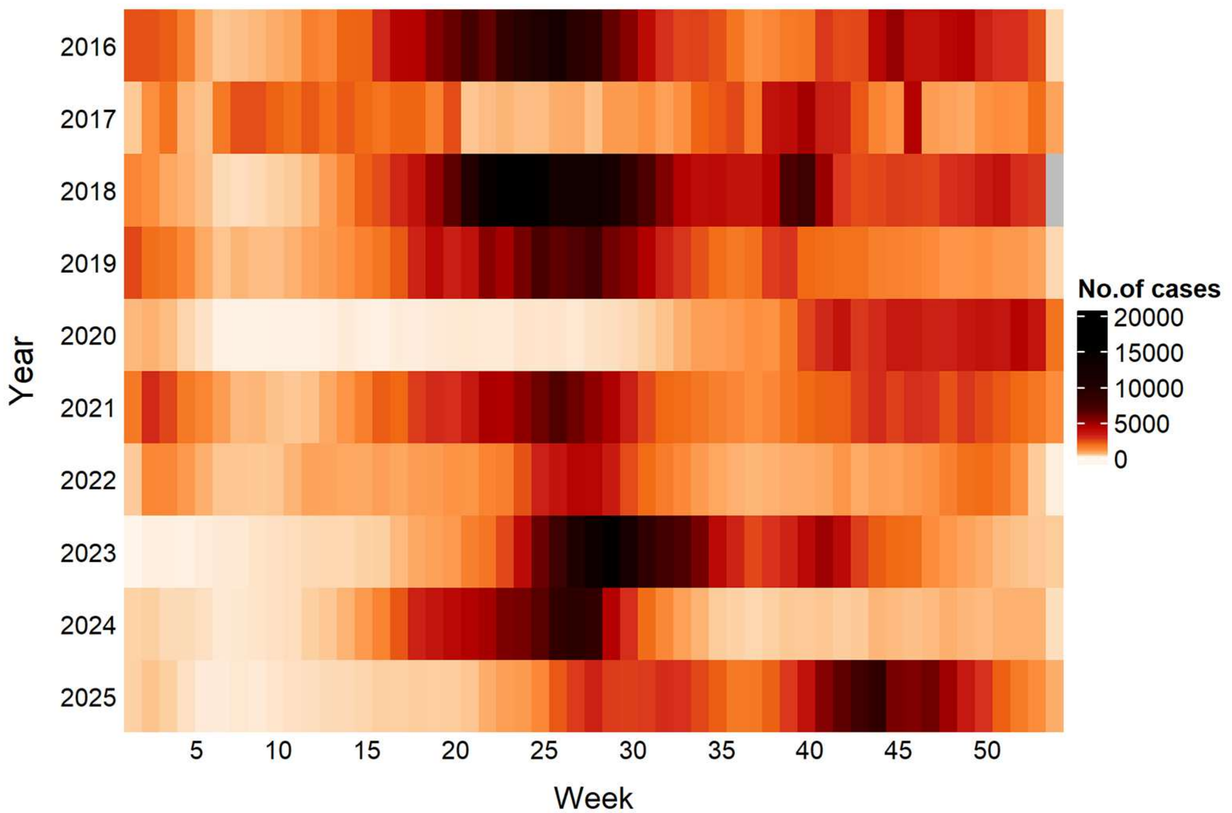
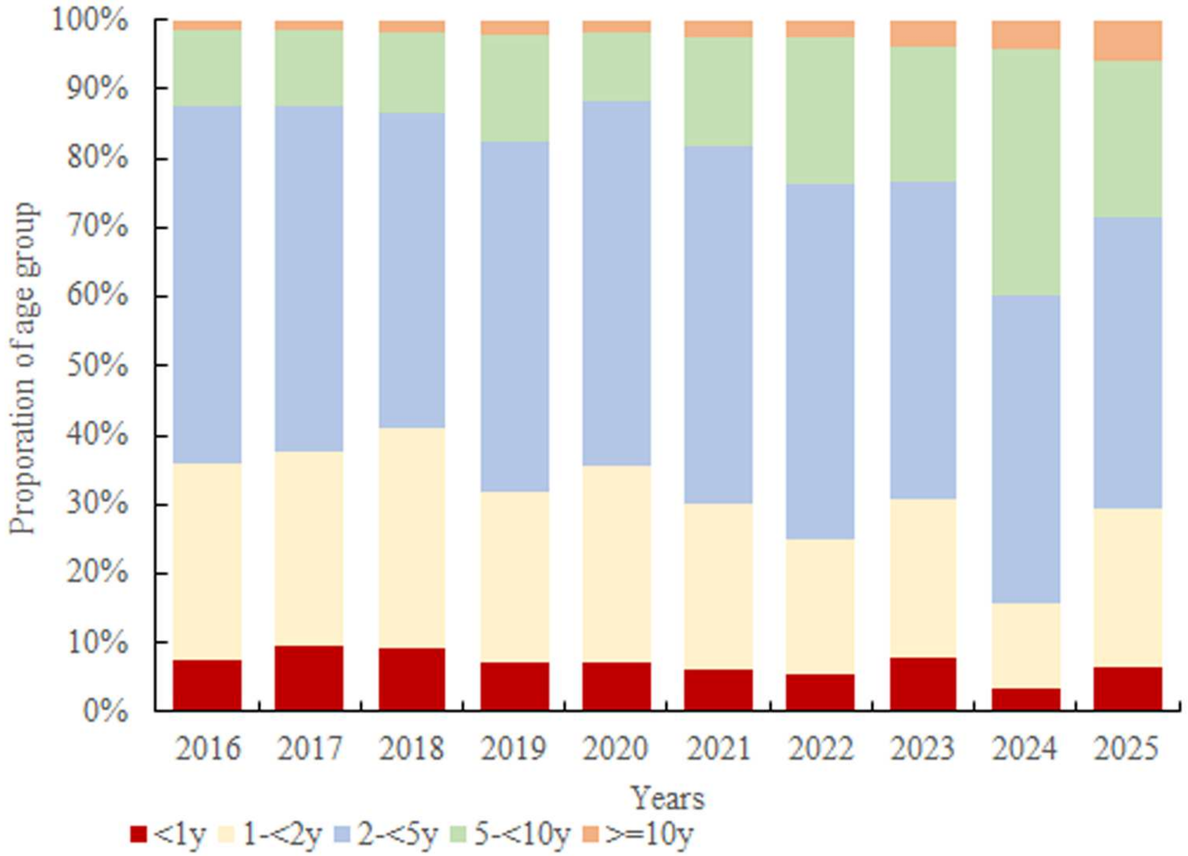
A


300000
250000
200000
150000
100000
50000
0
Annual cases number, n
0.050%
0.045%
0.040%
0.035%
0.030%
0.025%
0.020%
0.015%
0.010%
0.005%
0.000%
Percent of severe cases(%)
0.047%
0.019%
0.006%
0.001%
0.009%
0.004%
0.003%
0.003%
0.000%
0.002%
2016
2017
2018
2019
2020
2021
2022
2023
2024
2025
Years
All cases
Severe
%Severe

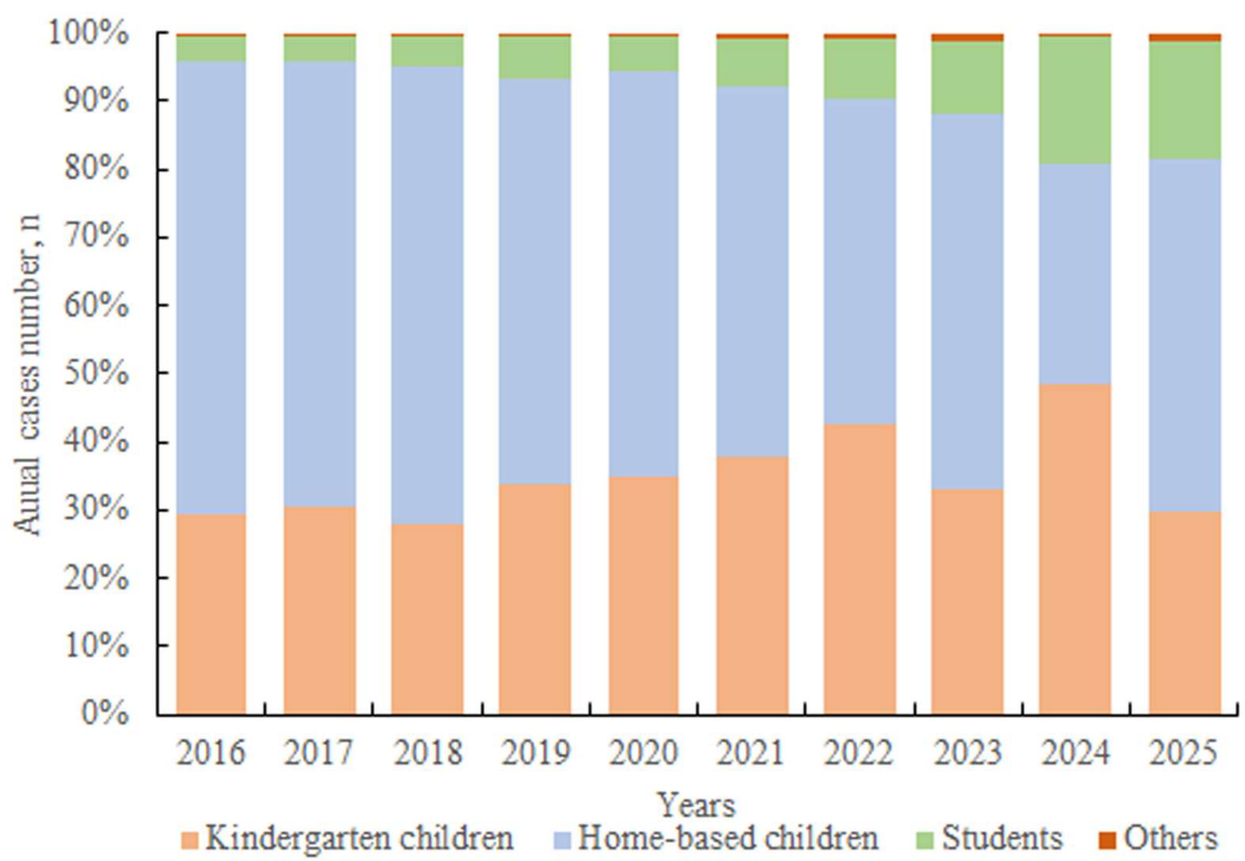
B

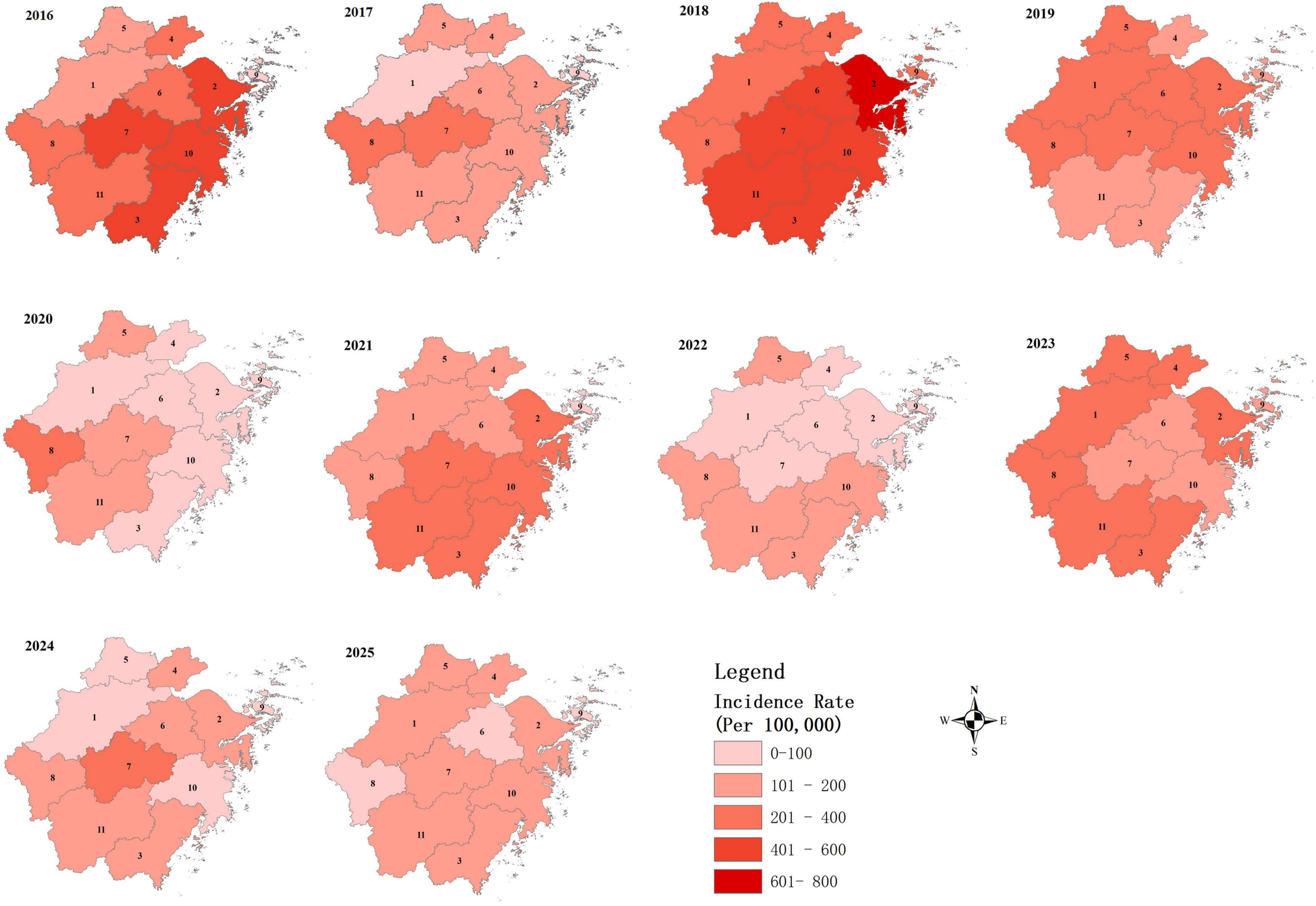

Year
2016
2017
2018
2019
2020
2021
2022
2023
2024
2025
5
10
15
20
25
30
35
40
45
50
Week
No.of cases
20000
15000
10000
5000
0

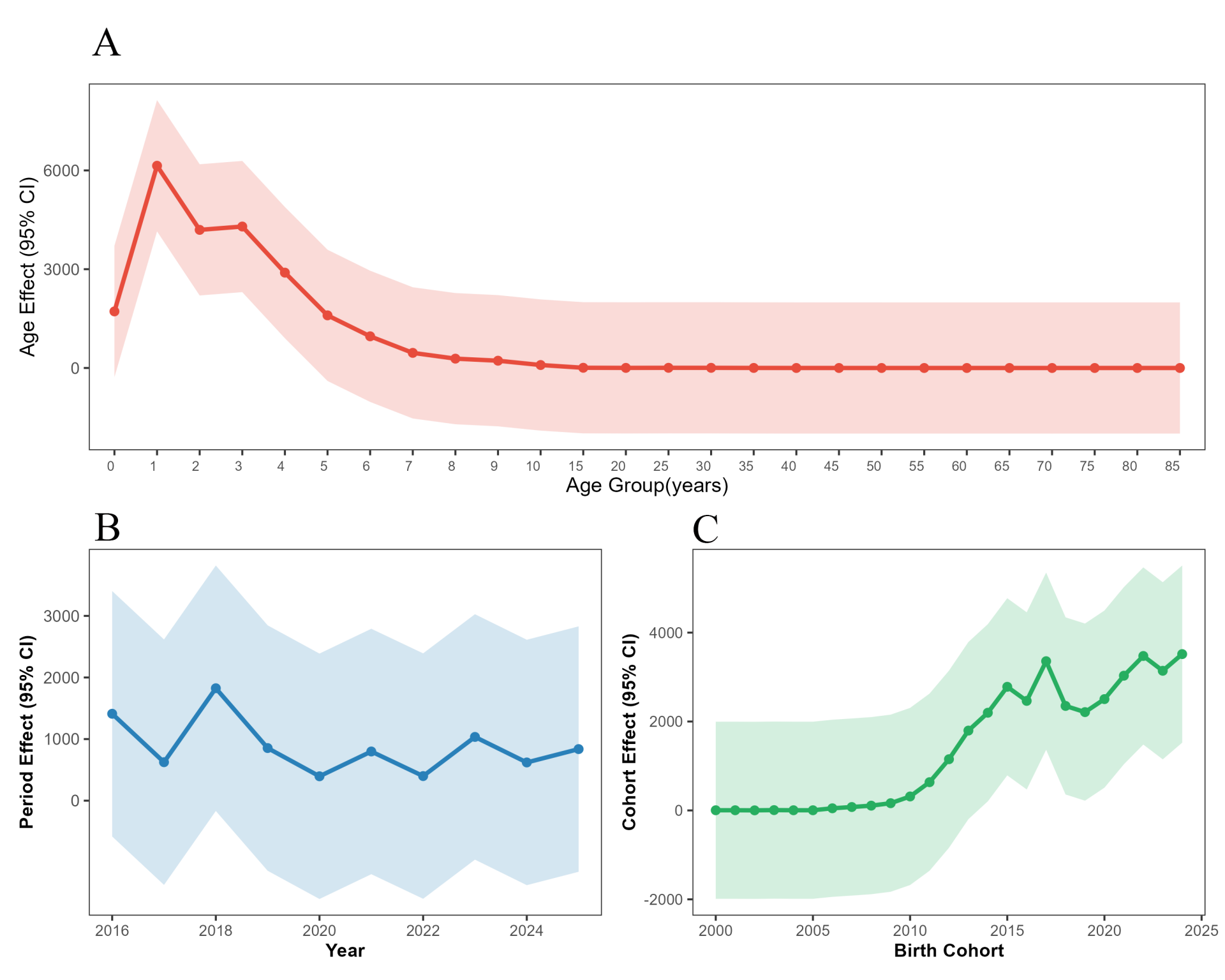
C

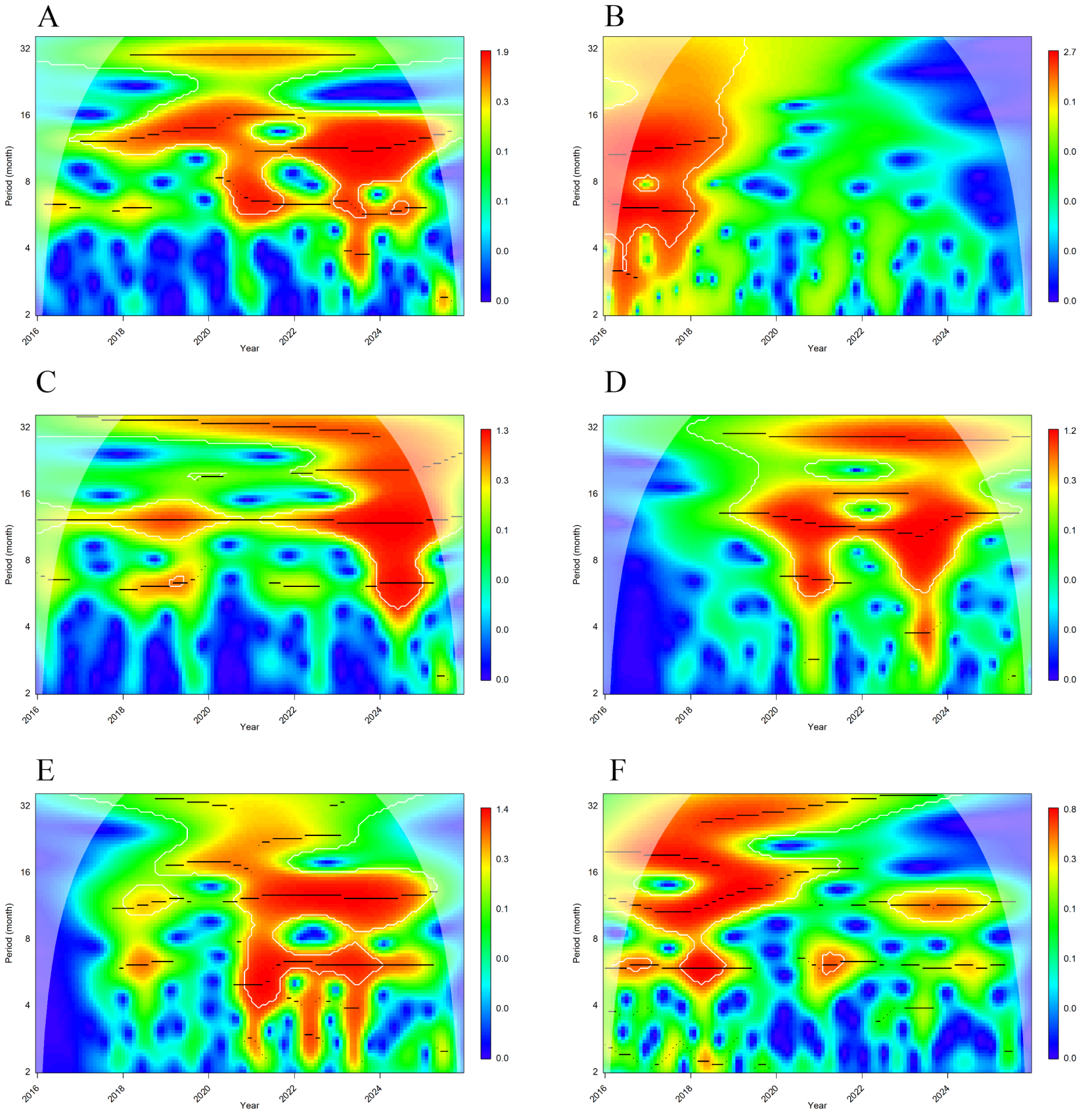

100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
Proporation of age group
2016
2017
2018
2019
2020
2021
2022
2023
2024
2025
Years
<1y
1-<2y
2-<5y
5-<10y
>=10y

D

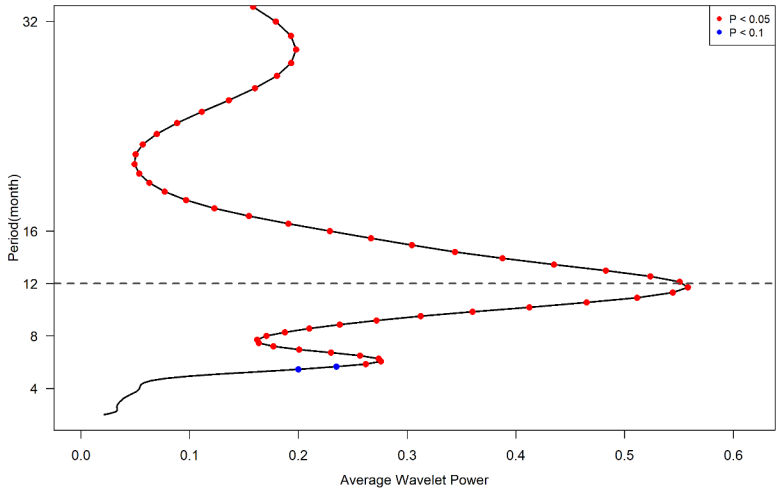

100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
Anual cases number, n
2016
2017
2018
2019
2020
2021
2022
2023
2024
2025
Years
Kindergarten children
Home-based children
Students
Others


2016
2017
2018
2019
2020
2021
2022
2023
2024
2025
Legend
Incidence Rate
(Per 100,000)
0-100
101 - 200
201 - 400
401 - 600
601- 800
N
W
E
S

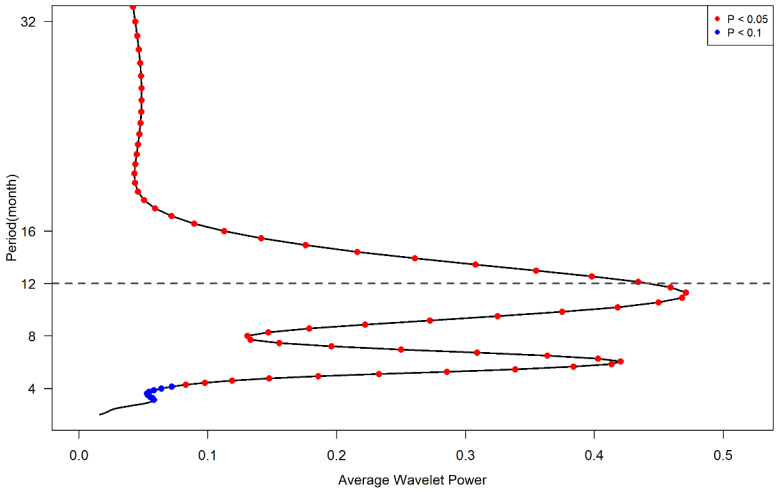

A
Age Effect (95% CI)
6000
3000
0
0 1 2 3 4 5 6 7 8 9 10 15 20 25 30 35 40 45 50 55 60 65 70 75 80 85
Age Group(years)
B
Period Effect (95% CI)
3000
2000
1000
0
2016 2018 2020 2022 2024
Year
C
Cohort Effect (95% CI)
4000
2000
0
-2000
2000 2005 2010 2015 2020 2025
Birth Cohort


A
B
C
D
E
F
Period (month)
Year
2
4
8
16
32
2016
2018
2020
2022
2024
1.9
0.3
0.1
0.1
0.0
0.0
2.7
0.1
0.0
0.0
0.0
1.3
0.3
0.1
0.0
0.0
0.0
1.2
0.3
0.1
0.0
0.0
0.0
1.4
0.3
0.1
0.0
0.0
0.0
0.8
0.3
0.1
0.1
0.0
0.0

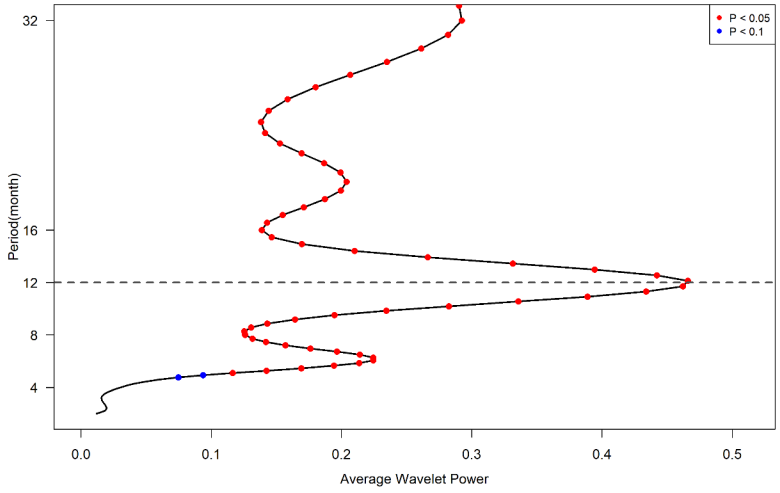
A


P < 0.05
P < 0.1
Period(month)
32
16
12
8
4
0.0
0.1
0.2
0.3
0.4
0.5
0.6
Average Wavelet Power

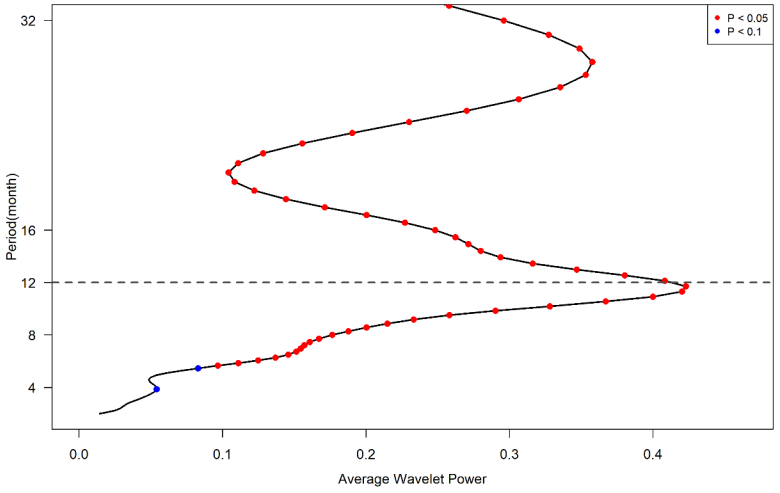
B


P < 0.05
P < 0.1
Period(month)
32
16
12
8
4
0.0
0.1
0.2
0.3
0.4
0.5
Average Wavelet Power

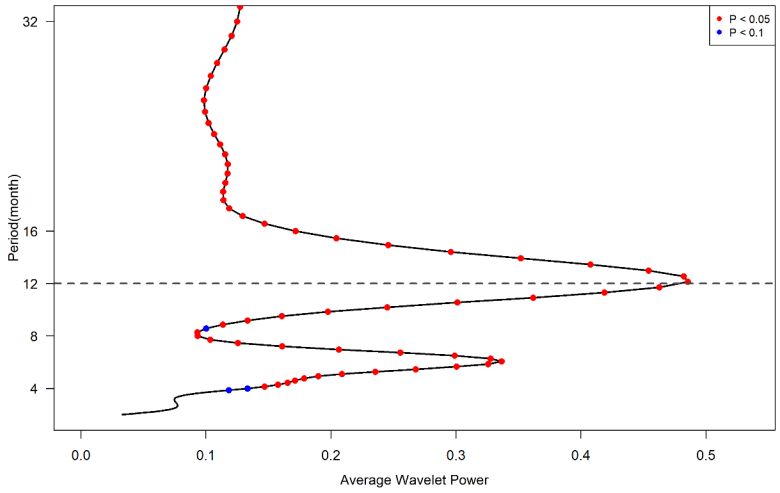
C


P < 0.05
P < 0.1
Period(month)
32
16
12
8
4
0.0
0.1
0.2
0.3
0.4
0.5
Average Wavelet Power

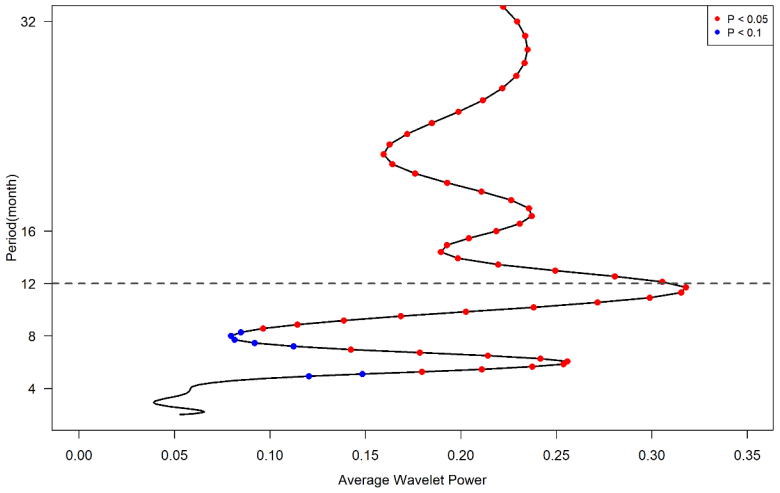
D


P < 0.05
P < 0.1
Period(month)
32
16
12
8
4
0.0
0.1
0.2
0.3
0.4
Average Wavelet Power

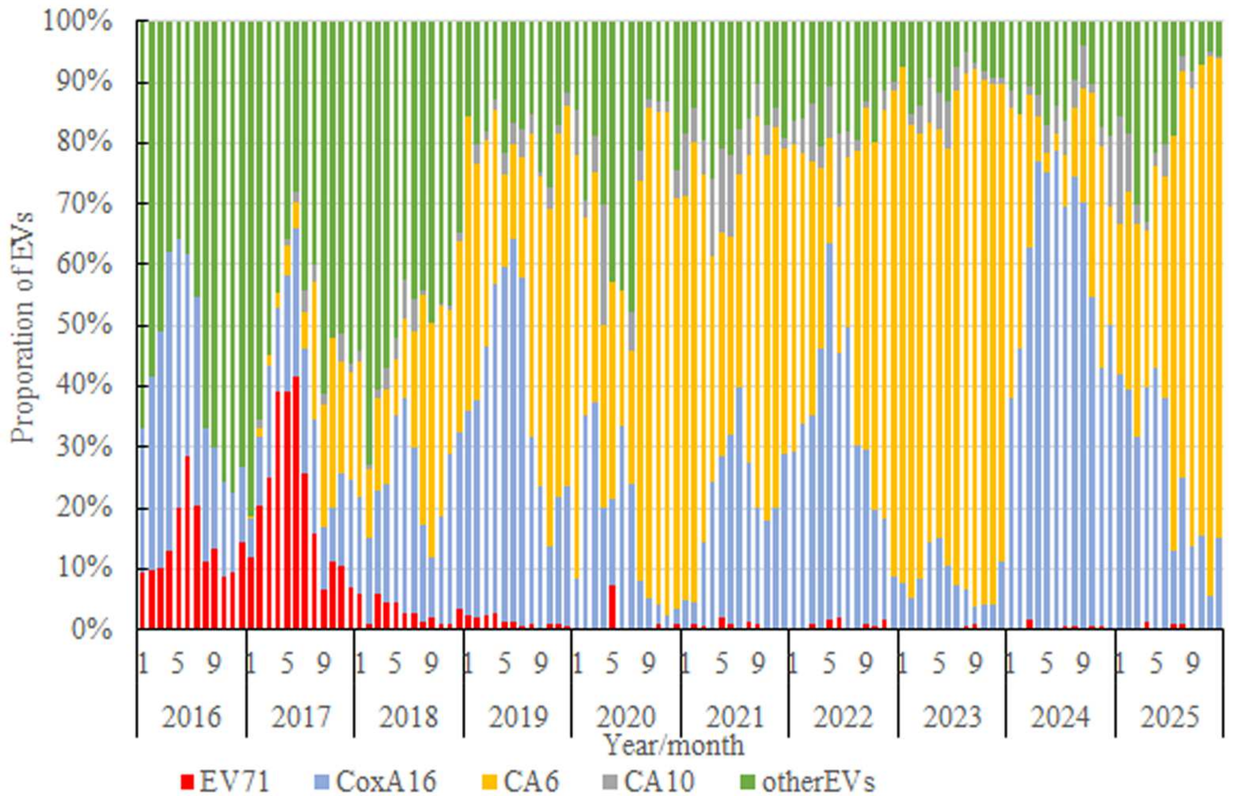
E


P < 0.05
P < 0.1
Period(month)
32
16
12
8
4
0.0
0.1
0.2
0.3
0.4
0.5
Average Wavelet Power

F


P < 0.05
P < 0.1
Period(month)
32
16
12
8
4
0.00
0.05
0.10
0.15
0.20
0.25
0.30
0.35
Average Wavelet Power

A


100%
90%
80%
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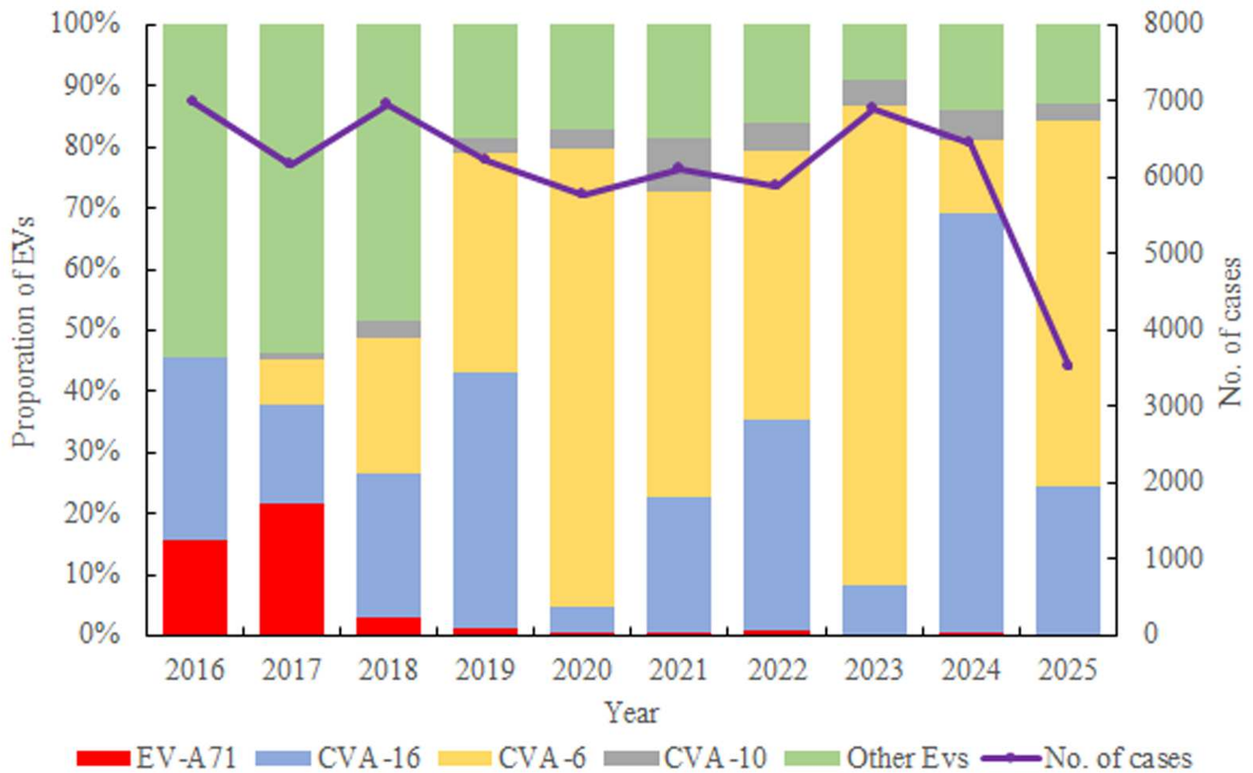
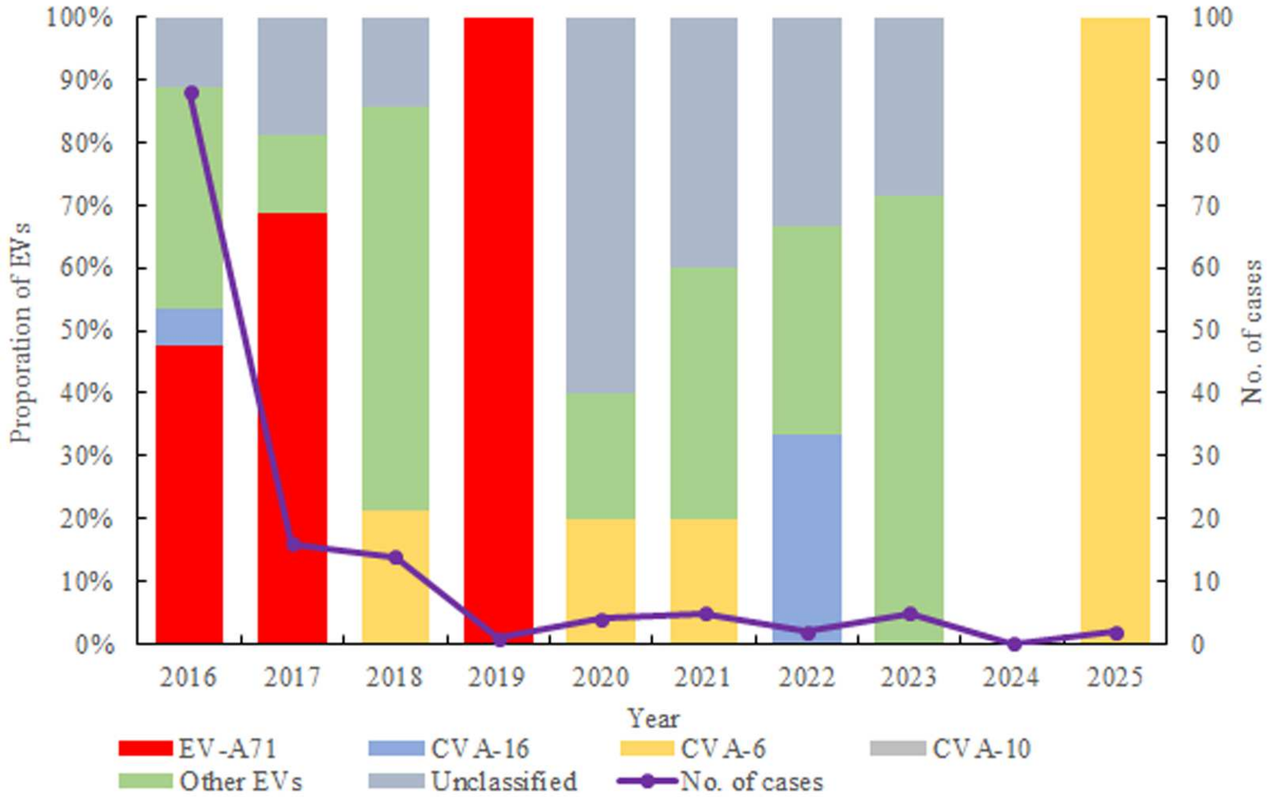
10%
0%
Proporation of EVs
1 5 9 1 5 9 1 5 9 1 5 9 1 5 9 1 5 9 1 5 9 1 5 9 1 5 9 1 5 9
2016
2017
2018
2019
2020
2021
2022
2023
2024
2025
Year/month
EV71
CoxA16
CA6
CA10
otherEVs

B


100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
Proporation of EVs
8000
7000
6000
5000
4000
3000
2000
1000
0
No. of cases
2016
2017
2018
2019
2020
2021
2022
2023
2024
2025
Year
EV-A71
CVA-16
CVA-6
CVA-10
Other Evs
No. of cases

C


100%
90%
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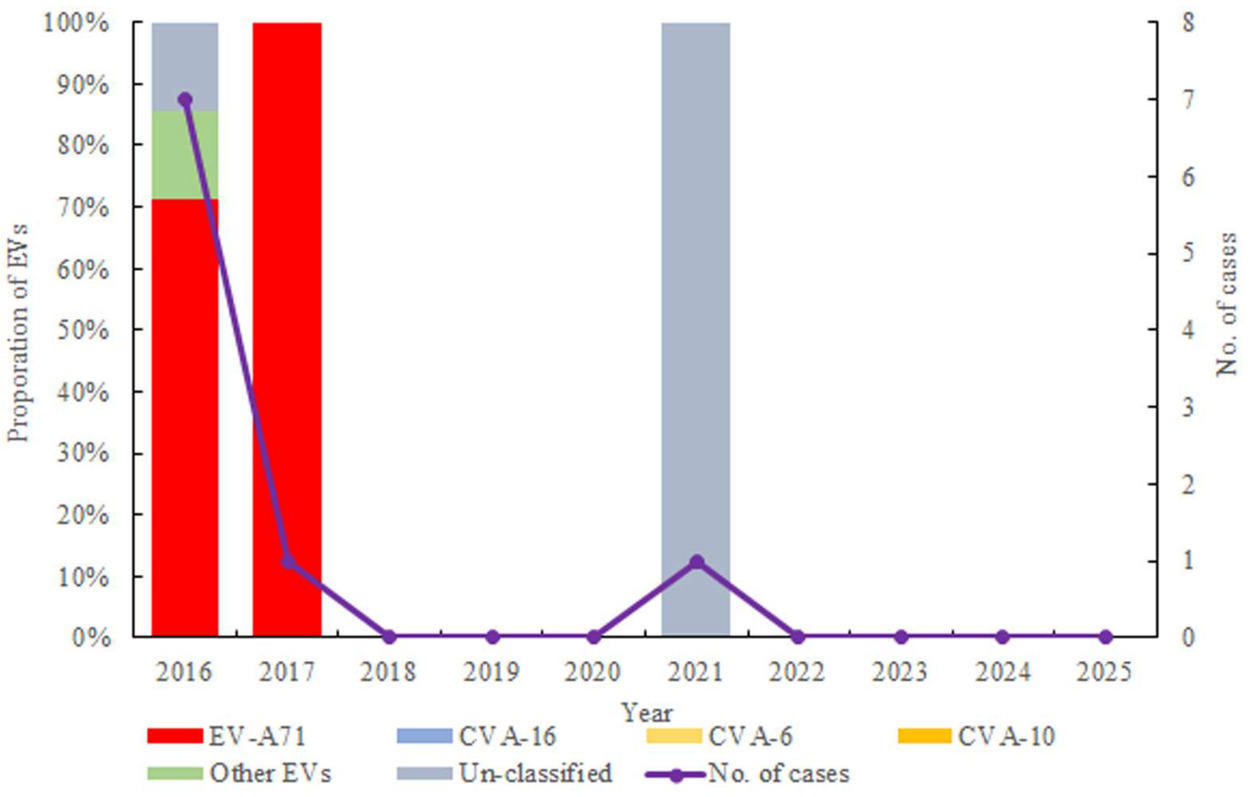
70%
60%
50%
40%
30%
20%
10%
0%
Proporation of EVs
100
90
80
70
60
50
40
30
20
10
0
No. of cases
2016
2017
2018
2019
2020
2021
2022
2023
2024
2025
Year
EV-A71
CVA-16
CVA-6
CVA-10
Other EVs
Unclassified
No. of cases

D


100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
Proporation of EVs
8
7
6
5
4
3
2
1
0
No. of cases
2016
2017
2018
2019
2020
2021
2022
2023
2024
2025
Year
EV-A71
CVA-16
CVA-6
CVA-10
Other EVs
Un-classified
No. of cases