Japanese encephalitis (JE) is one of the most common encephalitides worldwide, with an estimated 68,000 cases and 13,200–600,400 deaths annually (1). The disease is caused by a mosquito-borne flavivirus and is found across south and east areas of Asia (currently 24 countries with JE virus transmission) (2). JE virus is transmitted to humans through the bite of infected Culex mosquitoes, which lay their eggs in irrigated rice paddies and other pools of stagnant water. Pigs and birds serve as amplifying vertebrate hosts. Humans are believed to be dead-end hosts, as viremia is too low to infect feeding mosquitoes.

The incubation period is 4–14 days. Most JE infections are asymptomatic. Symptoms range from non-specific febrile illness to severe meningoencephalitis, occurring in about one out of 300 cases. JE virus infection can also cause aseptic meningitis or poliomyelitis-like acute flaccid paralysis. Convulsions are common in children. Case-fatality rate in severe clinical cases is estimated to be 20–30%, with young children (<10 years) having a greater risk of severe disease and death. Approximately 30–50% of survivors of severe disease have long-term neurologic or psychiatric sequelae.

Currently, three WHO-prequalified JE vaccines are available, all of which are safe and effective. WHO recommends use of JE vaccines in all countries where JE is recognized as a public health priority (3). The recommended introduction strategy is a one-time campaign in the target population (usually children <15 years), followed by introduction of JE vaccines into the routine immunization programme.

**DISEASE AND VACCINE CHARACTERISTICS**

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**RATIONALE AND OBJECTIVES OF SURVEILLANCE**

The following are the surveillance objectives for JE:

- establish JE epidemiology, including defining at risk populations and estimating JE burden in a country
- determine geographic distribution of JE in country
- provide information to inform JE vaccination policy
- evaluate vaccine impact and effectiveness after introduction.

**TYPES OF SURVEILLANCE RECOMMENDED**

**MINIMAL SURVEILLANCE**

- Sentinel hospital, case-based surveillance with laboratory confirmation should be conducted in areas where JE is suspected to be a problem. Sentinel hospitals must be located in appropriate geographic locations and should be sufficiently numerous to identify areas where JE transmission may occur. Since most acute encephalitis syndrome (AES) cases will require hospitalization, a focus on inpatient facilities is likely to capture a majority of AES cases. Sentinel hospitals should have necessary laboratory and clinical infrastructure to conduct appropriate laboratory investigations of admitted AES cases, such as the capacity to analyse cerebrospinal fluid (CSF) and ship specimens to a reference laboratory for testing if needed.
- JE surveillance should be conducted year round. While JE transmission shows some seasonality in some countries, it may occur year round in other areas. In addition, AES is caused by many other pathogens and occurs year round, which is another
CASE DEFINITIONS AND FINAL CLASSIFICATION

SUSPECTED CASE DEFINITION FOR CASE FINDING
Because JE cannot be distinguished from encephalitis due to other causes on clinical grounds alone, a syndromic approach is used in identifying cases. A suspected JE case is a person meeting the definition of AES. The AES clinical case definition is a person of any age at any time of year with the acute onset of fever and at least one of the following:

- A change in mental status (including symptoms such as confusion, disorientation, coma or inability to talk)
- New onset of seizures (excluding simple febrile seizures).

A simple febrile seizure is defined as a seizure that occurs in a child aged 6 months to < 6 years old, whose only finding is fever and a single generalized convulsion lasting less than 15 minutes, and who recovers consciousness within 60 minutes of the seizure.

AES might have reduced sensitivity for JE among children in some settings. In a study in Vietnam, some children with laboratory-confirmed JE presented with signs of meningitis only (like neck stiffness) or acute limb paralysis only. Overall, the AES case definition captured two-thirds of children with JE; sensitivity among adults was 100%, though the numbers were small (4). Countries can choose to conduct integrated surveillance for meningoencephalitis, which could also include acute flaccid paralysis (AFP) (see section on Special considerations for surveillance of Japanese encephalitis at end of chapter). Dengue virus (DENV) infections may be captured in AES surveillance due to the low specificity of the AES case definition, and case presentations can overlap.

FINAL CASE CLASSIFICATION

- **Laboratory-confirmed JE**: An AES case that has been laboratory-confirmed as JE.
- **Probable JE**: An AES or suspected JE case that occurs in close geographical and temporal relationship to a laboratory-confirmed case of JE, in the context of an outbreak.
- **AES – other agent**: An AES case in which diagnostic testing is performed and an etiologic agent other than JE virus is identified.
- **AES – unknown**: An AES case in which no diagnostic testing is performed or in which testing was performed but no etiologic agent was identified, or in which the test results were indeterminate.

reason for year-round surveillance when the surveillance platform is used to detect other causes of AES besides JE.

- JE surveillance can be integrated with Integrated Disease Surveillance and Response (IDSR), where possible.
- Although JE is commonly seen in children younger than 15 years of age in Asia, cases can occur in the older age groups especially when the virus enters a new area. Furthermore, in areas with a JE vaccination programme, there is frequently a shift to a greater proportion of cases in older, unvaccinated age groups. While the recommended surveillance includes all age groups, in countries that are in the early implementation stage of a JE vaccination programme, it is cost-effective to conduct surveillance among children < 15 years of age, or in the group with the preponderance of cases.

ENHANCED SURVEILLANCE

- Nationwide, case-based surveillance for JE (and AES) might be considered, where possible, for the following reasons: 1) data from nationwide surveillance of AES with laboratory confirmation provides the best source of information for complete JE disease burden; and 2) in countries where a high level of JE control has been achieved, nationwide surveillance in all hospitals with laboratory confirmation of all suspected cases can capture the most cases. However, this kind of surveillance requires extensive resources.
The attending clinician, nurse or surveillance officer identifies cases fulfilling the AES clinical case definition. The case investigation form should be completed and specimens drawn as soon as possible. It is important to exclude other causes of AES, such as bacterial or tuberculous meningitis or cerebral malaria, which require specific timely treatment.

* An adequate sample is one with a volume greater than 0.5 ml and transported to the laboratory under reverse cold chain.

Source: WHO manual for the laboratory diagnosis of Japanese encephalitis virus infection, 2007 (5)
SPECIMEN COLLECTION

SPECIMEN TYPES
CSF is the preferred specimen for laboratory confirmation of JE and should be gathered from every case when possible. Furthermore, CSF is also important in identifying other causes of AES and meningitis. The attending clinician should perform lumbar puncture as soon as possible after admission to hospital or case identification. Depending on the hospital set-up, CSF should be allocated into at least three separate sterile screw-cap collection tubes. Laboratory confirmation of JE requires at least 0.5 to 1.0 ml of CSF. CSF samples must be received in the laboratory within one hour of collection or stored at 2–8°C before transport (-20°C for longer-term storage). Receipt more than one hour after collection may result in false-negative results.

If a CSF sample is unable to be collected, a blood specimen can be collected. Blood should be collected via venipuncture as soon as possible after admission. Because JE-specific IgM antibodies may not be present at the time the first blood sample is taken, a second blood sample must be obtained on day 10 of illness (usually on the seventh day of hospitalization), at the time of discharge or at the time of death. These blood samples will be processed in the hospital laboratory to obtain the serum for testing for JE antibodies. Obtain at least 3–5 mL of blood for older children and adults, and 1–2 mL of blood for infants and younger children.

A second blood specimen for IgM is not needed if the first blood or CSF specimen is positive. However, IgM ELISA laboratory testing is frequently conducted at a location other than where the sample was collected (such as a national laboratory or provincial laboratory). Taking into account specimen transport and testing time, the results are often not available by the time of discharge or death, so a second sample should generally be collected.

STORAGE AND TRANSPORT
CSF and serum samples should be stored in a refrigerator (at 2–8°C) prior to transport, if the specimens can be transported within one to three days. However, for longer-term storage, specimens should be kept at or below -20°C. Avoid repeated freezing and thawing of samples. At least two tubes should be sent to the national or regional reference laboratories for JE-specific testing, if JE testing is not available at the sentinel hospital.

LABORATORY TESTING

CONFIRMATION METHODS
The recommended method for laboratory confirmation of a JE virus infection is to demonstrate the presence of JE virus-specific IgM antibody in a single sample of CSF or serum, as detected by an IgM-capture enzyme-linked immunosorbent assay (ELISA) specifically for JE virus (5). JE-specific IgM antibody detected by IgM-capture ELISA in the CSF or serum reaches > 95% sensitivity 10 days after onset of first symptoms. It is important to differentiate true JE virus infections from false-positive JE results because of cross-reactive epitopes among flaviviruses. For example, a patient with a dengue virus infection might have a positive JE IgM result because of flavivirus cross-reactivity. Current commercial JE IgM detection assays have low specificity for JE. Therefore, to rule out false-positive results, a validated dengue-specific assay has to be carried out on all JE-positive specimens.

In addition, any of the following laboratory criteria is confirmatory for JE:

- detection of JE virus antigens in brain tissue by immunohistochemistry or immunofluorescence assay
- detection of JE virus genome in CSF, serum, plasma, blood, or brain tissue by reverse transcription polymerase chain reaction (RT-PCR) or an equally sensitive and specific nucleic acid detection assay, such as loop mediated isothermal amplification (LAMP) or whole genome sequencing
Japanese Encephalitis

- isolation of JE virus in CSF, serum, plasma, blood or brain tissue
  
  OR

- detection of a four-fold or greater rise in JE virus-specific IgG antibody as measured by plaque reduction neutralization test (PRNT) in serum collected during the acute and convalescent-phase of illness. The two IgG specimens should be collected at least 14 days apart.

Detection of virus genome or virus isolation in serum, plasma, or blood is very specific for JE diagnosis; however, it is not sensitive, as virus levels are usually undetectable in clinically ill JE cases. Therefore, a negative result by these methods should not be used to rule out JE in a suspected case. Similarly, detection of virus genome or virus isolation in CSF is usually only found in fatal cases, and is therefore not very sensitive and should not be used for ruling out a diagnosis of JE. JE cannot be ruled out in cases with IgM negative results when the sample was collected less than seven days post-onset of illness because the IgM may not have risen to detectable levels, and a second sample should be collected.

SPECIAL LABORATORY CONSIDERATIONS

- The large majority of JE infections are asymptomatic. Therefore, in areas that are highly endemic for JE, it is possible to have AES due to a cause other than JE virus and have JE virus-specific IgM antibody present in serum from a recent, possibly subclinical, infection. Therefore, testing of a CSF sample from all persons with encephalitis is recommended when feasible. Even in the presence of a positive CSF specimen, further confirmatory tests should be carried out (such as, looking for cross-reactivity with other flaviviruses circulating in the geographical area) in any of these situations:
  - there is an ongoing dengue or other flavivirus outbreak
  - JE vaccination coverage is very high
  - the areas do not have epidemiological and entomological data supportive of JE transmission.

- For persons vaccinated with JE vaccine within six months of illness onset, testing a single serum sample for JE IgM may not be diagnostic because any IgM detected may be vaccine-related and not disease-related. In such cases, a diagnosis can only be confirmed by demonstrating JE IgM in the CSF, JE virus isolation, a positive nucleic acid amplification test, immunohistochemistry or a four-fold or greater rise in antibody titer in acute- and convalescent-phase serum samples.

- There may be other causes of acute disease with impaired brain function, such as meningitis (viral and bacterial including tuberculosis), encephalopathy due to toxins, cerebral malaria, viral encephalitis due to herpes simplex virus, mumps virus or neurovirulent enteroviruses (such as enterovirus 68 or enterovirus 71 with hand, foot and mouth syndrome) or post-infectious meningoencephalitis (such as post-measles or post-varicella). Distinguishing between encephalitis and encephalopathy and between encephalitis and meningitis requires CSF examination with cell count, cell morphology and standard biochemistry. CSF cell count of < 10 cells/mm³, and especially < 6 cells/mm³, is indicative of encephalopathy or early bacterial infection. CSF cell count of 10–100 cells/mm³, predominantly lymphocytic, is indicative of encephalitis or viral meningitis. CSF cell count > 100 cells/mm³ with predominantly polymorphs is indicative of bacterial meningitis; CSF cell count > 100 cells/mm³ with predominantly lymphocytes is indicative of viral meningitis. High protein concentration (> 100 mg/dL), low glucose (< 40 mg/dL), or both must alert the clinician of possible tuberculous meningitis or other bacterial cause.

LABORATORY NETWORKS

In 2008–2009, the JE laboratory network was established to improve the capability for JE case confirmation among countries either known or suspected to be endemic for JE in the WHO Western Pacific Region and the WHO South East Asia Region. The JE laboratory network was formed based on the WHO polio and measles-rubella multi-tiered laboratory network models. The purpose of the JE network is to improve and standardize the capability of JE diagnosis in countries where the disease is endemic, and to determine disease burden with a view to a more targeted vaccine introduction. The network in the WHO Western Pacific Region consists of one global specialized laboratory (GSL) in Japan, two regional reference laboratories (RRLs) in China and the Republic of Korea, and seven national laboratories (NLs) in Cambodia, the Lao People’s Democratic Republic, Malaysia, Papua New Guinea, the Philippines and Viet Nam (northern and southern). In the South East Asia Region, the JE laboratory network includes one RRL in India and 14 NLs in nine countries: Bangladesh, Bhutan, India (6), Indonesia, Myanmar, Nepal, Sri Lanka, Thailand and Timor Leste.
DATA COLLECTION, REPORTING AND USE

RECOMMENDED DATA ELEMENTS

- Unique case identifier
- Date of birth (or age if date of birth not available)
- Sex
- Place of residence (city, district and province)
- Travel history over the past two weeks
- Ever immunized against JE
- Number of JE vaccine doses received
- Dates of all JE vaccine doses (if card available)
- If vaccinated, type of JE vaccine (most recently received)
- Symptoms (fever, change in mental status, and seizure)
- Date of onset of first symptoms (fever, change in mental status, or seizure)
- Type of specimen collected (CSF, serum, autopsy)
- Type(s) of testing methodology (IgM, PRNT, PCR, virus isolation, etc.)
- Date(s) of specimen collection (include serum sample one and two)
- Date(s) specimen(s) received in laboratory
- Date(s) specimen(s) tested (for each different type of test)
- Date(s) of laboratory results reported from laboratory
- Laboratory results for each specimen
- Final classification: laboratory confirmed JE, probable JE, AES unknown, AES other agent
- Status at discharge: alive, dead, unknown
- Date of death or discharge

RECOMMENDED DATA ANALYSES

- Number of suspected JE (AES) cases and confirmed JE cases by week, month, year, age group, geographical area, and immunization status
- Incidence of suspected JE cases (AES) and confirmed JE cases by week, month, year, age group and geographical area (if population-based or nationwide surveillance is conducted)
- Case fatality rate
- Final classification of all suspected JE (AES) cases
- Proportion of AES cases attributed to JE

USING DATA FOR DECISION-MAKING

The principal uses of surveillance data for decision-making are to do the following:

- guide policy and strategies on JE control
- assess the impact of vaccination
- identify geographic areas or populations at high risk to further guide where immunization coverage should be improved
- monitor the performance of surveillance
- monitor the performance of the laboratory
- monitor vaccine effectiveness.

In countries where a high level of JE control has been achieved, case-based data should be reported. Reporting should be weekly or monthly and include “zero-reporting” (i.e., no blanks left in the reporting forms – a zero should be indicated when there are no cases detected).

Although reporting of JE cases is not required by International Health Regulations (IHR), JE is included on the WHO/Unicef Joint Reporting Form, which should be submitted annually.

REPORTING REQUIREMENTS AND RECOMMENDATIONS

Aggregate JE case counts (confirmed and probable) to track disease burden are sufficient to identify clusters and monitor trends. Aggregate case counts should be reported at least monthly to public health authorities.
### Surveillance performance indicators for Japanese encephalitis/AES

<table>
<thead>
<tr>
<th>SURVEILLANCE ATTRIBUTE</th>
<th>INDICATOR</th>
<th>TARGET</th>
<th>HOW TO CALCULATE (NUMERATOR/ DENOMINATOR)</th>
<th>COMMENTS</th>
</tr>
</thead>
<tbody>
<tr>
<td>COMPLETENESS OF REPORTING</td>
<td>Percentage of surveillance units reporting to the national level, even in the absence of cases</td>
<td>≥ 80%</td>
<td># of surveillance units in the country reporting / # of surveillance units in the country x 100</td>
<td>None</td>
</tr>
<tr>
<td>TIMELINESS OF REPORTING</td>
<td>Percentage of surveillance units reporting to the national level on time, even in the absence of cases (e.g., monthly)</td>
<td>≥ 80%</td>
<td># of surveillance units in the country reporting by the deadline / # of surveillance units in the country x 100</td>
<td>This should be at least quarterly reporting. At each level reports should be received on or before the requested date.</td>
</tr>
<tr>
<td>SPECIMEN COLLECTION</td>
<td>Percentage of all suspect cases for which at least one specimen was collected</td>
<td>≥ 90%</td>
<td># of AES cases with specimen collected / # of AES cases x 100</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td>Percentage of suspected AES cases that have a lumbar puncture performed</td>
<td>≥ 90%</td>
<td># of suspected meningitis cases that had a lumbar puncture performed / # of suspected meningitis cases x 100</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td>Percentage of serum samples taken a minimum of 10 days after onset</td>
<td>≥ 80%</td>
<td># of serum samples obtained at least 10 days after onset of illness / # of serum samples received in the laboratory x 100</td>
<td>This applies to where the testing methodology is IgM capture ELISA</td>
</tr>
<tr>
<td>SPECIMEN ADEQUACY</td>
<td>Percentage of CSF and serum samples reaching laboratory in adequate condition</td>
<td>≥ 80%</td>
<td># CSF and serum samples reaching laboratory in adequate condition / all CSF and serum samples received in the laboratory x 100</td>
<td>Adequate is defined as 1) the specimen is transported using reverse cold chain and 2) the sample volume is greater than 100 µL</td>
</tr>
<tr>
<td>TIMELINESS OF REPORTING LABORATORY RESULTS</td>
<td>Percentage of laboratory results reported to national public health authorities 7 days after receipt of specimen</td>
<td>≥ 80%</td>
<td># of laboratory test results reported &lt; 1 month of specimen receipt / # of specimens received by lab x 100</td>
<td>Indicator only applies to public laboratories.</td>
</tr>
<tr>
<td>SENSITIVITY</td>
<td>Minimum AES rate per 100,000 population</td>
<td>&gt;2/100,000</td>
<td># of AES cases captured by the surveillance / # of target population in the country x 100,000</td>
<td>This applies to enhanced (nationwide) surveillance and not minimal (sentinel) surveillance.</td>
</tr>
</tbody>
</table>
CLINICAL CASE MANAGEMENT

No specific antiviral treatments have been found to benefit patients with JE, but similar to other cases of AES, hospitalization for supportive care and close observation is generally required. Potentially preventable factors often associated with death include raised intracranial pressure, status epilepticus, hypoglycaemia, aspiration pneumonia, and secondary infections (6). Treatment is supportive. Rest, fluids and use of pain relievers and medication to reduce fever may relieve some symptoms. Supportive care such as intravenous fluids and vasopressors might be needed in severe cases.

CONTACT TRACING AND MANAGEMENT

No contact investigations are done for JE as the disease is vector-borne.

SURVEILLANCE, INVESTIGATION AND RESPONSE IN OUTBREAK SETTINGS

DEFINITION OF AN OUTBREAK
An outbreak of JE can be defined as an occurrence of the disease in excess of the expected frequency in a given area among a specific group of people over a particular period of time, or two or more epidemiologically linked cases of the illness in a short period (7). Major outbreaks of JE occur every 2–15 years in endemic areas, especially in areas with low use of JE vaccine. JE transmission normally intensifies during the rainy season, when vector populations increase. Studies have shown a strong association between agricultural practices, including rice cultivation, and density of the vector mosquitoes for JE; therefore, an increase in abundance of rice fields or establishment of rice cultivation in new areas is likely to increase JE risk (8).

CHANGES TO SURVEILLANCE DURING AN OUTBREAK
Only the first 5–10 cases of an outbreak need to be confirmed through laboratory testing. If an outbreak continues over a protracted period, another 5–10 samples should be collected every two to three months to ensure that the outbreak is still due to JE. If the outbreak is not an expected seasonal outbreak, or there are unusual epidemiological features (such as age distribution of cases not consistent with pattern of JE infection or absence of typical vectors or hosts), testing of CSF is especially important, as an encephalitis outbreak could be due to other etiologies.

PUBLIC HEALTH RESPONSE
Public health response to JE should include the following:

- **Vaccination.** JE vaccination should be integrated into national immunization schedules in all areas where JE is recognized as a public health priority. The value of reactive vaccination campaigns during outbreaks of JE has not been studied. If an outbreak occurs in a country or region where JE vaccination has not been introduced, assess whether it is appropriate to implement an immediate vaccine response, considering factors such as size of the outbreak, timeliness of the response, population affected and programmatic capacity. Due to the need for rapid production of protective antibodies, live attenuated or live recombinant vaccines should be used. When outbreak response vaccination is conducted, planning for introduction into the routine immunization schedule should follow (3).
Japanese Encephalitis

\[ \text{Japanese Encephalitis} \]

\[ \begin{align*}
&\text{SPECIAL CONSIDERATIONS FOR JAPANESE ENCEPHALITIS SURVEILLANCE} \\
&\text{INTEGRATED SYNDROMIC SURVEILLANCE} \\
&\text{The AES case definition emphasizes sensitivity over specificity in detecting suspected cases of JE. However, as mentioned, some cases of JE, particularly among children, might only present with signs of meningismus or AFP, which are not captured by AES. Conversely, many other diseases, including bacterial meningitis and other flavivirus infections, can present as AES. Because of the overlap in clinical syndromes, there is reason to consider integrating AES surveillance for JE with surveillance for meningitis (and possibly AFP) to maximize sensitivity and efficiency of surveillance. Approaches to encephalitis and meningitis surveillance are similar, with collection of a CSF specimen for definitive diagnosis. Integration of syndromic surveillance may be appropriate to help streamline programme logistics, ensure case detection is as complete as possible, and make the best use of available resources (see chapters on pneumococcus, meningococcus, and \textit{Haemophilus influenzae}). Integrated meningoencephalitis surveillance, for example, could enable data collection for a variety of central nervous system infections for which an effective public health control measure such as immunization is available (examples include JE, meningitis due to \textit{Haemophilus influenzae} type b, meningococcus and pneumococcus).}
&\text{A challenge in integrating JE and bacterial meningitis surveillance is that different laboratory capacity is needed for confirmation of viruses and bacteria. From a laboratory standpoint, JE surveillance might be more easily integrated with surveillance of other viral vaccine-preventable diseases. As such, JE surveillance has been successfully integrated with polio and measles surveillance, including use of the same laboratory networks as acute meningitis-encephalitis surveillance in China and the same networks as AES surveillance in India (9). The public health priorities in a country, availability of viral and bacterial diagnostics and access to testing may all determine the appropriateness of an integrated approach.}
\end{align*} \]

\[ \text{Health education and community involvement. It has been observed that there is a direct relationship between the time lag in onset of symptoms and initiation of medical care. Immediate supportive management of cases reduces fatality considerably.} \]

\[ \text{Interruption of transmission. There is little evidence to support a reduction in JE disease burden from interventions other than vaccination of humans, such as vaccination of pigs, environmental management for vector control, and chemical control of vectors (3).} \]

\[ \text{WHO Vaccine-Preventable Diseases Surveillance Standards} \]
REFERENCES CITED

ADDITIONAL REFERENCES