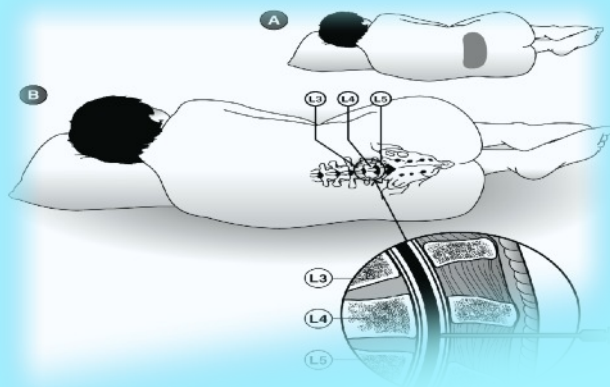


# NATIONAL GUIDELINE ON MENINGOCOCCAL MENINGITIS SURVEILLANCE AND OUTBREAK MANAGEMENT



Ethiopian Health and Nutrition Research Institute  
Federal Democratic Republic of Ethiopia



1<sup>st</sup> Edition  
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# Guideline on Meningococcal Meningitis Surveillance and Outbreak Management



**Ethiopian Health and Nutrition Research Institute  
Public Health Emergency Management Center**

**Federal Democratic Republic of Ethiopia**

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## ACRONYMS

AR	Attack Rate
CSF	Cerebrospinal Fluid
EPR	Epidemic Preparedness and Response
EPI	Expanded Program of Immunization
ICG	International Coordination Group
IFRC	International Federation of Red Cross and Red Crescent Societies
MSF	Medecins Sans Frontieres
NGO	Non Governmental Organization
PCR	Polymerase Chain Reaction
TI	Trans-isolate
UNICEF	United Nations Children's Fund
WHO	World Health Organization

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We particularly thank WHO for financial contribution and facilitation of the printing of this guideline.

## SECTION 1. INTRODUCTION

Meningococcal meningitis is a contagious disease caused by Gram-negative diplococcal bacteria called, *Neisseria meningitidis* (Nm). There are two clinical forms of meningococcal disease. The first clinical form is meningococcal meningitis, which is more common, especially during outbreaks; outcomes are good if appropriately treated. The second clinical form is meningococcal septicemia, in which bacteria are found in the blood stream, less common but highly fatal, even in intensive care treatment setup. Patients who have meningitis and septicemia simultaneously are usually regarded as cases of meningitis.

Meningococcal meningitis, commonly known as cerebrospinal meningitis, is the only form of bacterial meningitis that causes outbreaks. The largest outbreaks occur mainly in the semi-arid areas of sub-Saharan Africa, designated the African “meningitis belt”. Epidemics of meningococcal meningitis previously occurred every 8-12 years, however in recent years they have been occurring yearly.

The purpose of this guideline is to help regional, zonal and woreda public health emergency management officers, health workers and others involved in surveillance and control of outbreak of meningococcal meningitis to guide them to do surveillance, early detect outbreaks, get prepared, coordinate and take appropriate outbreak responses.

The guideline has eight basic components: Introduction to meningococcal meningitis, Meningococcal meningitis surveillance, Outbreak investigation, Specimen collection, transport and processing, Control and prevention measures, Information, education and communication (IEC), Coordination, Monitoring and evaluation.

Based on the mandate given to the Federal Ministry of Health to prepare and distribute health and health related guidelines and standards, this guideline has been prepared by the Ethiopian Health and Nutrition Research Institute (EHNRI), Public Health Emergency Management (PHEM) Center.



## 1.1. Epidemiology of the Disease

Meningitis is an infection and inflammation of the meninges (the membranes that cover the brain and the spinal cord). Meningitis can be caused by many different pathogens including bacteria, viruses, fungi and parasites. Meningococcal meningitis is a contagious disease caused by Gram-negative diplococcal bacteria called; *Neisseria meningitidis* (Nm). At any time, 5-10% of the population may be nasopharyngeal carriers of *N. meningitidis*. Invasive disease is rare in non-outbreak areas, occurring at a rate of 0.5-10 cases per 100,000 populations per year, but can occur at a rate of up to 1,000 cases per 100,000 populations per year in outbreak prone areas. In endemic areas, during outbreaks, children over age 6 months, adolescents and young adults are the most at risk groups with 80- 90% of cases occurring under the age of 30 years, <sup>1</sup>.

Outbreaks can occur in any part of the world. However, the highest burden of meningococcal disease occurs in sub-Saharan Africa, which is known as the "Meningitis Belt", an area that stretches from Senegal in the west to Ethiopia in the east. This hyper-endemic area is characterized by particular climate and social habits. During the dry season, between December and June, because of dry windy conditions and higher incidence of upper respiratory tract infections, the local immunity of the pharynx is diminished thereby increasing the risk of meningitis. At the same time, the transmission of *N. Meningitidis* is favored by overcrowding and large population displacements. These factors help explain some of the large outbreaks that occur during this season in the meningitis belt area. Due to herd immunity (whereby transmission is blocked when a critical percentage of the population had been vaccinated, thus extending protection to the unvaccinated), these outbreaks occur in a cyclic mode.

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1. WHO. 2005. Communicable Disease Control in Emergencies. A field Manual. WHO, Geneva, 2005.

There are about 12 Serogroups causing meningococcal meningitis outbreak in Africa. In 1996, Africa experienced the largest recorded outbreak of outbreak meningitis in history, with over 250,000 cases and 25,000 deaths registered. The most affected countries were Burkina Faso, Chad, Ethiopia and Niger. In 2002, outbreaks occurring in Burkina Faso, Ethiopia and Niger accounted for about 65% of the total cases reported in the African continent.

In Ethiopia, meningitis outbreaks have been described in written reports since 1901<sup>2</sup> Outbreaks were reported in 1935, 1940, 1950, 1964, 1981 and 1989. The 1981 and 1989 outbreaks were the largest ever recorded in Ethiopia with 50,000 and 45,806 cases, and 990 and 1686 deaths respectively. The 1981 outbreak affected the northern and western part of Ethiopia. The 1988-1989 meningococcal meningitis outbreaks affected all regions. Since these major outbreaks a number of smaller outbreaks have occurred in the country most notably outbreaks in Amhara, Tigray and Gambella Regions in February 2000. Between March and August 2000 there was an outbreak in Addis Ababa with 850 cases and 33 deaths.

During 2001 major epidemic was recorded with 6964 cases and 330 deaths followed by another epidemic during 2003-2004 epidemic seasons which recorded a total of 3326 cases and 160 deaths from all regions and was not limited to the traditional meningitis belt areas of North West and South Western part of the country.

In the epidemic season 2005 a total of 1061 cases with 46 deaths were reported from four regions while epidemic in the year 2006 affected all Regions with a report of close to 3000 cases. Out of these cases 1300 cases (45%) with 43 deaths were reported from three regions, namely Oromiya, SNNPR and Tigray.

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2. Tedla T. Meningococcal meningitis. In Kloos H and Zein AZ (eds), the Ecology of health and Disease in Ethiopia. Westview Press, Boulder and Oxford, 1993: 285-293.

Between the year 2005 and 2010 foci of epidemics occurred in few areas which were managed timely and contained at a local level.

During 2010, the country reported 1611 cases with 21 deaths from 23 woredas in Oromia, SNNPR, Amhara and Tigray while close to 1200 cases with 30 deaths (2.5%) from Oromia, SNNPR, Amhara and Gambella were recorded during the year 2011 and major epidemic was reported in 2013 from all zones of SNNP and central and south parts of Oromia region with report of 1466 cases with 40 deaths (CFR- 2.7%).

## 1.2. Infectious Agent

*Neisseria meningitidis* is classified into different serogroups on the basis of the composition of the capsular polysaccharide (currently 12 serogroups have been identified). The 5 major meningococcal serogroups associated with disease are A, B, C, Y and W135. About 90% of infections are caused by serogroup A, B and C<sup>3</sup>. Serogroup A predominates in the meningitis belt and accounts for about 80 to 85 percent of all cases. Serogroups C, Y and W135 are also found.

## 1.3. Mode of Transmission

*Neisseria meningitidis* only infects humans; there is no animal reservoir. It is transmitted through close contact with infected persons through respiratory secretions or saliva while sneezing or coughing. Then the bacteria colonize the nasopharynx of susceptible individuals.

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3. Medecines Sans Frontieres. 2008. Management of Epidemic Meningococcal Meningitis 4<sup>th</sup> edition, Medecines Sans Frontieres Medical Department , France, January 2008

This infection usually goes unnoticed or manifests as simple pharyngitis, and the majority of infected individuals develop protective antibodies and become healthy carriers. Asymptomatic carriers play a major role in meningococcal transmission.

#### 1.4. Risk Factors

The risk factors for invasive disease and for outbreaks are not completely understood. However, a combination of conditions such as the environment, the host and the organism are necessary for an outbreak to occur.

Dry, hot season and dust storms climatic conditions, overcrowded living and working conditions such as schools, prisons Immunological susceptibility of the population i.e. accumulation of unvaccinated and susceptible population, etc. All age groups are at risk of meningitis, but age groups 2 to 30 years are more at risk.

The type and the virulence of the strain is also a risk factor for an outbreak to occur. Acute respiratory tract infections may also contribute to the development of meningococcal disease.

#### 1.5. Pathogenesis

*Neisseria meningitidis* attaches to the microvilli of non-ciliated columnar epithelial cells that reside in the nasal region of humans. The bacteria are able to multiply and form a colony because of its ability to acquire iron from the host. The bacteria are then able to invade the mucous membrane that lines the nasopharynx. *Neisseria meningitidis* then has access to the blood stream of the individual and from there can move into the meninges of the brain.

## 1.6. Symptoms and Signs

Acute meningitis is characterized by a sudden onset of:

- intense headache,
- high grade fever,
- nausea,
- Vomiting,
- photophobia (fear to see light) and
- Stiff neck.

In addition, neurological signs can be observed, such as lethargy, delirium, coma, and/or convulsions.

Infants may have illness without sudden onset stiff neck and bulging fontanels may be observed.

Physical examination will typically reveal:

- meningeal rigidity (stiff neck with Kerning's and/or Brudzinski's signs),
- purpura (sometimes extensive and necrotic, usually localized in the extremities, or generalized, coetaneous or mucosal (conjunctival)),
- Low blood pressure and symptoms of shock. Shock associated with purpura indicates fulminating meningococce-mia, the most severe form of meningococcal disease.

## 1.7. Incubation Period and Period of Infectivity

The incubation period of meningococcal meningitis is 2-10 days, most commonly 3-4 days. The period of infectivity extends until the organism no longer exists in mouth and nasal discharges. However, infectivity stops in 24 hours after starting appropriate antibiotics.

## 1.8. Differential Diagnosis

In endemic situations, acute meningitis or meningoencephalitis is associated with purulent or cloudy CSF only in a minority of cases, the CSF is clear. Acute meningitis with clear CSF could also be due to one of numerous viral agents. Other bacteria (*Mycobacterium tuberculosis*, spirochetes) or fungi (*Cryptococcus*) also cause acute meningitis.

The other most important differential diagnosis of meningococcal meningitis is other acute febrile illnesses such as severe falciparum malaria, relapsing fever, typhoid fever etc. However, the association of acute fever, purpura and shock is very suggestive of meningococcal disease.

## 1.9. Complications

The complications of meningitis can be severe. The longer the disease goes without treatment, the greater the risk of seizures and permanent neurological damage, including: hearing loss, blindness, memory difficulty, loss of speech, learning disabilities, behavior problems, shock, brain damage and paralysis.

## 1.10. Outcome

Early treatment improves the outcome. If untreated the case-fatality rate may exceed 50%. However, even in appropriately treated patients the case-fatality rate can be higher than 10%. Young children and adults over 50 have the highest risk of death. Among those who survive the meningococcal disease, 10-20% experience neurologic sequelae.

## SECTION 2. MENINGOCOCCAL MENINGITIS SURVEILLANCE

Implementation of strengthened meningitis surveillance activities at all levels of the health system with prompt laboratory confirmation of circulating pathogen is an essential strategy for early detection of meningococcal meningitis outbreaks. These surveillance activities must be scaled-up at an early stage in the meningitis season before an outbreak has occurred. They can help detect the first few cases, identify the pathogen and the serogroups of the Nm and serve as a trigger to launch a rapid response operation. Standard case definitions can be used to recognize early cases and these should then be confirmed by laboratory tests. Standard reporting mechanisms are needed in order to analyze the incoming data and determine the extent and evolution of an outbreak.

Meningococcal meningitis is an epidemic prone disease, and is one of the 21 priority diseases and conditions that are under the Public Health Emergency Management integrated disease surveillance system. It is a weekly reportable disease.

### 2.1 Standard Case Definitions

#### **Suspected case:**

Any person with sudden onset of fever ( $>38.5$  °C rectal or  $38.0$  °C axillary) and one of the following signs: neck stiffness, altered consciousness, or other meningeal signs such as bulging fontanel, convulsion.

#### **Probable case:**

Any suspected case with turbid or purulent CSF or with microscopic examination showing Gram-negative diplococci.

#### **Confirmed case:**

A suspected or probable case confirmed by isolation of *Neisseria meningitidis* from CSF or blood by culture, PCR or agglutination test.

## 2.2. Reporting

The flow of surveillance data is usually from reporting sites to the next level up to the central level. The community and health facilities, especially health posts are the main source of information. Routinely, all suspected and confirmed cases of meningitis should be reported on weekly basis. The absence of cases should also be reported as zero (zero reporting) on weekly bases, to allow public health personnel to distinguish an area that is truly unaffected from the one in which the surveillance or communication system has not functioning well.

If a health facility suspects a meningococcal meningitis outbreak, according to the PHEM guideline, the health facility should notify the woreda within 30 minutes. The woreda should notify the zone within another 30 minutes. The zone should notify the region within another 30 minutes. And the region should notify the Federal Public Health Emergency Management (PHEM) center within another 30 minutes.

Report the first 10 cases of suspected meningitis cases using the Case-based Reporting Format (CRF) during the suspected outbreak with CSF samples to determine the Nm serogroups. If the outbreak is confirmed and the outbreak threshold is surpassed then start reporting using line list and daily epidemic reporting format. Keep testing or sending 5-10 CSF samples every week for further follow up and characterization.

## 2.3. Laboratory Based Surveillance

Laboratory based surveillance is the key part of the overall surveillance as the detection and control of outbreaks requires rapid identification of the pathogens and their source of infection. Suspected meningitis outbreaks should be confirmed by laboratory investigation at health centers, hospitals and regional and national laboratories. Further and advanced testing of the meningococcal meningitis samples can be performed at regional and at the national reference laboratory levels.



Before the beginning of the outbreak season, an adequate stock of lumbar puncture kits, rapid latex identification tests (Pastorex) and trans-isolate (TI) bottles should be available. These materials should be readily prepositioned at national and regional levels.

### 2.3.1. Collection of specimens

Collection of CSF is an invasive technique and should be performed by experienced personnel under aseptic conditions. CSF should be collected in sterile containers. CSFs are high priority specimen. Once CSF is obtained from the lumbar puncture, it should be immediately transported to the laboratory (within one hour) and plated. CSF specimen should never be refrigerated.

During the meningitis outbreak, health personnel and field investigation teams should collect CSF specimens for laboratory confirmation. Collecting a total of 20 to 30 CSF samples is recommended for an affected area to determine the circulating causal pathogens, drug susceptibility trends for selection of appropriate antibiotics, to guide the choice of the vaccines to be used if new serogroups are isolated and to limit the number of invasive medical practices.

The number of CSF specimen to be collected may vary according to local circumstances and human resources available. Once the outbreak has been confirmed, regular collection of CSF specimens (5-10 per week) should be continued in selected areas throughout the outbreak season, in order to monitor circulating serogroups.

Health personnel at health facilities should be trained on performing lumbar puncture, specimen collection, TI media utilization and specimen handling and transportation to the reference laboratory.

## Collecting CSF for laboratory analysis

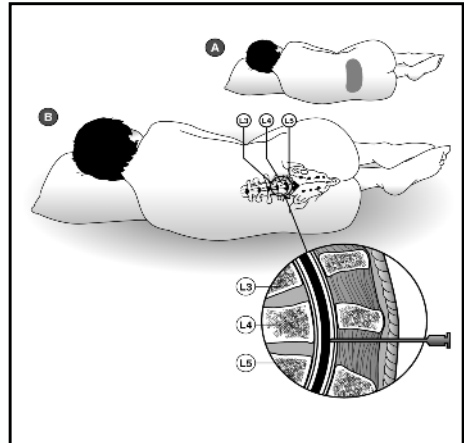
### How to perform a lumbar puncture:

#### What you need:

- lumbar puncture needles
- sterile alcohol swabs
- sterile gauze pad
- latex gloves
- iodine
- adhesive labels

#### Step by step:

- Wash your hands
- Put on sterile gloves
- Disinfect the puncture site
- Locate the puncture site between Lumbar Vertebrae 4 and 5 or 3 and 4 ( L4-5 or L3-4)
- Use a spinal needle to collect 1 to 3 ml of cerebrospinal fluid (CSF) in the sterile tube
- Dress the puncture site and allow the patient to lie flat for a minimum of 30 minutes



## 2.3.2. Transport of Specimens

### For Culture

Preparing CSF samples for transportation for culture, sensitivity and serology tests as follows:

You are expected to place 1ml of CSF sample into a TI bottle

#### **Procedure:**

- Remove a bottle of TI medium from the refrigerator at least 30 minutes before inoculating it with the specimen.
- Before inoculating the vial, check to see if there is any visible growth or turbidity. If there is visible growth or turbidity, discard the media, because it may be contaminated.
- Lift up the small lid in the middle of the metal cap on top of the TI bottle.
- Disinfect the top of the TI bottle with alcohol and allow drying.
- With a new, sterile needle and syringe transfer 1 ml of CSF from the sterile tube into the TI bottle.
- If not transported immediately, puncture the top of the TI bottle with a sterile needle to ventilate and ensure bacteria growth.
- Keep the sample at room temperature away from light and cold.
- Remove the needle to avoid contamination before packaging for travel.
- Forward the TI bottle to the woreda authority for transportation to a reference laboratory within 24 hours without cold chain.
- Label the TI bottle with patient name and identification number and complete the appropriate form.

### Always remember that:

- TI vials should never be frozen.
- Before inoculation and transportation TI bottle should be kept in the refrigerator.
- Once inoculated, TI bottle should be kept at room temperature and ventilated until transportation not more than 24 hours.

The TI bottles containing CSF specimen will be sent from the health facility to the woreda within 48 hours. The woreda team should send the collected TI bottles to the regional and national reference laboratory at least twice per week. The woreda team should ensure that the collected specimens are maintained at room temperature.

### For Polymerase Chain Reaction (PCR):

Next collect 1 ml of CSF in cryotube for PCR testing and send along with the TI bottle, in order to detect etiological pathogens in case of no growth of the specimens sent by TI.

Note that CSF specimens in cryotubes should preferably be stored in a freezer (-20°C) or in sterile dry tubes in the refrigerator (+4°C for few weeks), and shipped in a cool box to national or regional reference laboratories with PCR capacity for the determination of causal pathogen and genotype.

### 2.3.3. Processing of specimens and laboratory procedures

All CSF specimens collected should undergo a Gram stain at the nearest laboratory for Gram reaction determination of presence of bacteria. The identification of the Nm serogroup is also crucial for deciding on the most appropriate polysaccharide (PS) vaccine to be used for outbreak control. Nm should be confirmed from CSF specimens by either:

- Rapid latex agglutination tests that can be used at the peripheral laboratories and allow the identification of most common pathogens/serogroups; or

- Culture and serogrouping at national or regional reference laboratories.

The following lab test will be conducted depending on the health services organizational levels (national, regional, woreda) and the technical capacity of the laboratory at that level.

**A. Gram stain:** it is important to strengthen laboratory capacities at woreda levels to perform Gram stain and cell counts at health facility with appropriate infrastructure (see annex 2).

**B. Latex test (Pastorex®):** Rapid latex tests are performed at the field or peripheral health facility levels. Latex tests confirm the pathogen and Nm serogroups and hence the use of a latex test is highly recommended during the initial phase of an outbreak as it substantially reduces the delay for bacteriological confirmation and decision-making.

### ***General Method for Performing Latex Agglutination Tests:***

Several commercial kits are available. Follow the manufacturer's instructions precisely when using these tests.

General recommendations and instructions typical for the detection of soluble bacterial antigens are provided here. For best results, test the supernatant of the centrifuged CSF sample as soon as possible. If immediate testing is not possible, the sample can be refrigerated (between 2°C and 8°C) up to several hours, or frozen at -20 °C for longer periods. Reagents should be kept refrigerated between 2°C and 8°C when not in use. Latex suspensions should never be frozen.

## Performance of the Test

- Heat the supernatant of the CSF in a boiling water bath for 5 minutes.
- Shake the latex suspension gently until homogenous.
- Place one drop of each latex suspension on a ringed glass slide or a disposable card.
- Add 30-50  $\mu\text{l}$  of the CSF to each suspension.
- Rotate by hand for 2-10 minutes. Mechanical rotation at 100 rpm, if available, is recommended.

## Reading the Test Results

**Read under a bright light without magnification.**

- Positive reaction: agglutination (visible clumping) of the latex particles occurs within 2 minutes.
- Negative reaction: the suspension remains homogenous and slightly milky in appearance.
- Non interpretable results: give a reaction with negative control or with more than one latex reagent in the kit. In this case, it is advisable to repeat the test with another sample and wait for **result of the culture.**

**C. Culture and Sensitivity:** Confirms pathogen and Nm Serogroups. Culture is the gold standard for laboratory confirmation and also confirms antibiotic sensitivity. The culture, serogrouping and drug sensitivity tests are done at regional or national reference laboratories.

**Note:** A discordance between a positive antigen test and a negative culture can be explained by the absence viable bacteria in the sample inoculated (i.e. antibiotic therapy instituted before samples were taken or transport conditions not adapted to survival of fragile bacteria).

**D. Polymerase Chain Reaction (PCR):** Performed at national level to confirm the causal agent by bimolecular (DNA) test. PCR can be used to confirm the bacteria if there is a negative TI (i.e. no growth by culture).

**E. Additional Tests:** Additional investigations performed on CSF may include:

- Measurement of glucose concentration (<0.40 g/l is suggestive of bacterial infection).
- Blood cell counts may show an increase of polymorph nuclear cells.
- Counter immunoelectrophoresis may also be used for direct antigen detection in CSF.

The laboratory results should be sent by the performing laboratory focal officers to the highest level and to the facility that sent the sample(s) within 48 hours upon completion. Remaining specimen has to also be sent to regional and national laboratories for further investigation.

- Woreda (Health facility): send to regional laboratory within 48 hours upon reception of the samples.
- Regional laboratory: within 5 days upon reception of the samples from woreda or health facilities.
- National level: give feedback within 7 days upon reception of the samples.

#### 2.3.4. Storage of CSF isolates frozen at minus 20°C

Storage of CSF isolates is recommended to allow further testing Serogrouping/stereotyping and susceptibility testing should be done in a National reference laboratory, due to the complexity of such tests.

Storage is done by freezing of CSF isolates at -20°C or at a -70°C. Freezers with automatic defrosters should never be used. When stored, isolates should be easily identifiable with a label, in order to link the frozen stored isolate with the patient information recorded.

## SECTION 3. MENINGOCOCCAL MENINGITIS OUTBREAK INVESTIGATION

### 3.1. Definition and Thresholds

Outbreaks of cases of meningococcal infection present some of the most challenging situations for public health authorities. There is an intense public concern and media interest due to the potential for severe morbidity and mortality among patients and the limited published evidence to guide best practice.

A meningococcal meningitis outbreak can be suspected when:

- A health facility reports an increase in the number of cases;
- An unusual increase in the number of cases is noted during data analysis; or
- Communities report an unusual number of cases or deaths;

Meningococcal meningitis thresholds and outbreak are defined as indicated in the table below:

**Table 1. Determining Alert and Outbreak Thresholds**

Threshold	In area with population of <30,000	In area with population of >30,000
<b>Alert threshold</b>	2 cases in one week  OR Greater number of cases than over the same period in non-epidemic years	AR = 5 cases/100,000 Population/week
<b>Epidemic threshold</b>	5 cases in one week  OR doubling in number of cases over 3 weeks period  Special situations should be studied on a case-by-case basis <sup>a</sup>	AR = 15/100,000 population/week In certain conditions indicating higher epidemic risk <sup>b</sup>  AR = 10/100,000 population/week Special situations should be studied on a case-by-case basis <sup>a</sup>



Epidemic threshold should be studied on a case-by-case basis

**AR** = 15/100,000 population/week

In certain conditions indicating higher epidemic risk

**AR** = 10/100,000 population/week

Special situations should be studied on a case-by-case basis

- a. For mass gatherings, refugees and displaced persons, 2 confirmed cases in 1 week are enough to warrant vaccination of the population.
- b. No epidemic in previous 3 years or vaccination coverage <80 % or alert threshold crossed early in season.

In order to guide appropriate response measures to a suspected outbreak, an outbreak investigation should be conducted. In investigating an outbreak, both the speed of the investigation and getting the correct answers are essential. To satisfy both requirements follow these steps:

### **3.2. Prepare for Field Work**

In order to enhance the capacity to respond to outbreaks the Public Health Emergency Management taskforce or an outbreak coordination committee at woreda level should immediately get together to agree on the need to investigate the suspected meningococcal meningitis outbreak. The committee then has to activate the multidisciplinary outbreak investigation team (rapid response team) and initiate outbreak investigation within 3 hours of receipt of a report of a suspected outbreak.

### **The rapid response team might consist of:**

- A clinician who will both verify patients' clinical symptoms, take CSF and also train health care workers in case management;
- A lab technician who will take samples and train healthcare workers in correct sampling and analysis procedures;
- An expert in information, education and communication who will assess how the community reacts to the disease and define and disseminate key health education messages; and
- An epidemiologist who will initiate data collection and assess surveillance procedures.

### **Before leaving for the field, the rapid response team should:**

#### **Read and know about the suspected disease;**

- Gather the supplies and equipment that are needed.
- Required formats (PHEM case based form, line list, Daily Epidemic Reporting Format for woreda and region).
- This guideline and other reading materials(PHEM guideline).
- Transport media(TI media).
- Material for Gram stain.
- Materials for specimen collection(needles, syringes, sterile tubes, gloves, aseptic agent).
- Communication equipment.
- Drugs for initial response if needed .
- Data analysis tools such as laptop.
- Make necessary administrative and personal arrangements for travel,
- Consult with all parties to determine each team members' role in the investigation and who your local contacts will be once you arrive on the scene.

### 3.3. Establish the Existence of an Outbreak

In order to establish the existence of the outbreak:

- Review trends in cases and deaths due to the disease over the last 1-5 years (if available),
- Determine a baseline number to describe the current extent of the disease in the catchment area,
- Know the epidemic threshold for that particular disease,
- Compare the reported case versus the baseline and the threshold per month or week under that particular catchment area,
- Take into account factors influencing disease occurrences such as seasonal variations in some of the diseases such as malaria and meningitis,
- Based on the finding, decide whether the outbreak exists or not.

### 3.4. Verify the Diagnosis

The goals in verifying the diagnosis are:

- Ensure that the problem has been properly diagnosed.
- Rule out laboratory error as the basis for the increase in diagnosed cases.

When verifying the existence of an outbreak early in the investigation, you must also identify as accurately as possible the specific nature of the disease. Examine patients at the health facility and review records to confirm that the signs and symptoms meet the standard case definition. Review laboratory results for the people who are affected. If you are at all uncertain about the laboratory findings, the techniques being used should be reviewed. Collect sample to isolate the organism or identify evidence of infection.

### 3.5. Identify Additional Cases

Conduct active searches for additional cases. In health facilities where cases have been reported, search for additional suspected cases and deaths in the registers. Look for other patients who may have presented with the same or similar signs and symptoms as the disease or condition being investigated. Do the search in neighboring health facilities too. Collect information on suspected cases using the line listing form and make sure that the first ten cases have PHEM case-based reporting forms completed and CSF collected. Contact tracing should be conducted in the communities where cases are identified to determine if there are additional symptomatic individuals.

### 3.6. Analyze Surveillance Data

Surveillance data analysis is essential to:

- Describe the characteristics of cases in order to understand the reasons for the occurrence of the disease and develop appropriate control measures;
- Predict potential outbreaks and implement vaccination strategies in order to prevent outbreaks;
- Detect and investigate outbreaks timely to ensure proper case management, and determine why outbreaks have occurred;
- Monitor progress towards achieving meningococcal meningitis control goals;

Using the routine surveillance data, conduct detailed analysis looking into all meningococcal meningitis cases that are confirmed by laboratory by person, place and time and look for trends and unusual changes.

## **The minimum expected data handling and analysis includes:**

Monitoring of the timeliness and completeness of surveillance reporting at all levels;

Following the trends of meningococcal meningitis using the basic epidemiological dimensions:

**Time:** Date patient develops symptoms - allows the creation of the epi-curve.

**Person:** What are the characteristics of the cases (e.g. age, sex)?

**Place:** Where do cases live? Where are the most affected areas/localities? Allows the mapping of the geographical extent of the outbreak (e.g. spot map or table with attack rates by woreda).

### **3.6.1. Data completeness**

Every week start data analysis by assessing the data completeness using the following formula:

#### **Completeness**

=  $\frac{\text{(total number of health facilities reported in that week)}}{\text{(total number of health facilities expected to report)}} \times 100$

Health facilities in your area are all the institutions that are required to report to you i.e. hospitals, health center and health posts. In order to get a correct epidemiological meaning completeness should be 80% or above.

### **3.6.2. Describe and orient the data in terms of person, place and time**

Once you have collected some data, you can begin to characterize an outbreak by person, place, and time. You may perform this step several times during the course of an outbreak. Characterizing an outbreak by these variables is called descriptive epidemiology. The data collected during an investigation should be analyzed to determine why the outbreak occurred. The data will also help to identify which group are the most susceptible individuals. Spot maps, demographic information distribution of the cases while attack rates can help to identify severity of the outbreak.

### ***In general, data analysis at this step will help you:***

- Learn what information is reliable and informative (e.g., the same unusual exposure reported by many of the people affected) and what may not be as reliable (e.g., many missing or "don't know" responses to a particular question).
- Provide a comprehensive description of an outbreak by showing its trend over time, its geographic extent (place), and the populations (people) affected by the disease. This description lets you begin to assess the outbreak in light of what is known about the disease (e.g., the usual source, mode of transmission, risk factors, and populations affected) and to develop causal hypotheses.

### ***Characterizing by time:***

Traditionally, the time course of an outbreak is shown by drawing a graph of the number of cases by date of onset. This graph, called an epi-curve gives a simple visual display of the outbreak's magnitude and time trend.

An outbreak curve provides a great deal of information.

- You will usually be able to tell where you are in the course of the outbreak, and possibly, to predict its future course.
- You have identified the disease and know its usual incubation period; you may be able to estimate a probable time period of exposure and can then develop a questionnaire focusing on that time period.

To draw an epi-curve, you first must arrange the data on a weekly basis using the date of onset of illness for each person. The number of cases seen in a week are plotted on the y-axis of an epi-curve while the unit of time (week number) on the x-axis. Also show the threshold lines (action threshold) so that you can clearly see whether the numbers cross the line. Do not forget to label the chart properly.

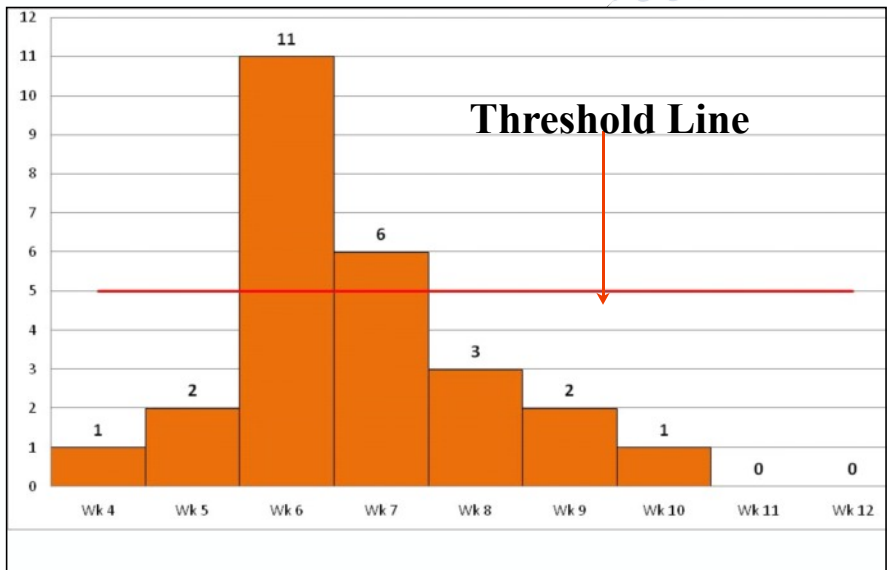


Figure 1. Example of an epi-curve showing outbreak threshold line

### Type Characterizing by person:

The following calculations and their interpretations should be used to see patterns of disease and determine what populations are at risk for the disease according to their host characteristics (age, sex) or exposures (vaccination status, residence, etc.). Such an analysis is also used to estimate an attack rate (by age grouping as in age specific attack rates, or by geographic area), the case fatality rate (as a measure of the quality of case management), and weekly incidence rate. The quality and reliability of the data is most important.

### Case Fatality Rate (CFR)

The case fatality rate is the proportion of cases that resulted in death. To find the case fatality rate, divide the number of deaths by the number of cases, and multiply by 100. CFR can be calculated on weekly, monthly or yearly bases. CFRs should be estimated by age-group, if possible.

**CFR**= (Total number of M.meningitis deaths during the week)/(Total number of M.meningitis cases during the same week) x 100

Note: total number of cases is equal to total cases alive plus total deaths.

### **Attack Rate (AR)**

Attack rate is an incidence rate (usually expressed as a percent), used only when the population is exposed to meningitis risk for a limited period of time such as during an outbreak. It relates the number of meningitis cases in the population at risk and reflects the extent of outbreak. Attack rate is calculated as follows :

**AR**= (Total number of M.meningitis cases seen)/(Total number of at risk population in the area) x 100

If age-specific data are available for the area of the outbreak, age-specific attack rates can also be calculated by restricting the numerator and denominator to persons within specific age groups (e.g., persons 2–30 years of age).

### **Weekly Incidence Rate (WIR)**

Incidence shows the rate at which new cases occur within a given period of time (usually one week). WIR can be expressed per hundred persons (percentage) or per 1000 persons.

**WIR**= (Total number of new meningitis cases during the week)/(Total number of at risk population during the same week) x 1000



## Characterizing by place:

Construct a spot map to using the place of residence on the case reporting forms or line lists. Then study the map and:

- Describe the geographic extent of the problem.
- Identify and describe any clusters or patterns of transmission or exposure.

Depending on the organism that has contributed to this outbreak, specify the proximity of the cases to likely sources of infection

## 3.7. Implement Control and Prevention Measures

Control measures, which can be implemented early if you know the source of an outbreak, should be aimed at specific links in the chain of infection, the agent, the source, or the reservoir.

### 3.7.1 Case Management

#### Principles

- Meningococcal disease (either meningitis or septicemia) is potentially fatal or will end in severe and permanent sequel and should always be managed as a medical emergency.
- Admission to a hospital or health centre is necessary for diagnosis (lumbar puncture and CSF examination) and for treatment.
- Antimicrobial therapy is essential and should be combined with supportive treatment.
- As contagiousness of patients is moderate and disappears quickly following antimicrobial treatment, isolation of the patient is not necessary.

Antimicrobial treatment must be instituted as soon as possible. Lumbar puncture should be performed, if possible, prior to the administration of antibiotics, which should be given immediately after the puncture, without waiting for laboratory results. Treatment of a suspected case of meningococcal meningitis with an antibiotic should not be delayed when lumbar puncture cannot be done on initial presentation.

A range of antibiotics can treat the infection, including Penicillin G, Ampicillin, chloramphenicol and Ceftriaxone. Note that it is important to determine the sensitivity of these drugs on a regular basis.

The recommended drug of choice for the treatment of cases of meningococcal meningitis during outbreaks is ceftriaxone or oily chloramphenicol as a single dose.

### ***Pre-Outbreak***

#### **At the woreda level: ,**

- Detailed data on the suspected cases should be recorded on a line list.
- Ensure CSF sample collection should be strengthened and samples sent to the nearest reference laboratory for bacteriological tests.
- Plan and implement training courses for health-workers on outbreak treatment protocols;
- Print and distribute national treatment protocols to all health-facility.
- Calculate the amount of antibiotics and material that may be needed during an outbreak, pre-position stocks in high-risk areas and establish smooth lines for distribution throughout the district.

Drug	Dosage	
	Children (0-23 Months)	Over 2 years of age and Adults
Ceftriaxone	<p>100mg/kg/day once a day for 5 days (age &lt;2 months) and for 7 days (age 2-23 months)</p> <p>Refer if no improvement within 48hrs or in coma or convulsion</p> <p>Adapt treatment according to patient's age and most likely causative pathogen if no improvement after 48hrs</p>	<p>100mg/kg IM</p> <p>2<sup>nd</sup> dose if no improvement after 24 hrs</p> <p>If no improvement after 48 hrs, treat for 5 days or refer.</p>
Oily Chloramphenicol	-	<p>Single dose as presumptive treatment</p> <p>100mg/kg/IM</p> <p>2<sup>nd</sup> dose if no improvement after 24 hrs</p>
Penicillin G	400,000 U/kg IV	3-4 MUq 4-6h IV
Ampicillin	250mg/kg IV	2-3gq.6h IV

## In the health facilities (hospitals and health centers):

- following lumbar puncture, treat every new patient who is suspected of having meningitis with antibiotics as soon as possible;
- ensure any child under 2 years of age or any patient with severe symptoms is admitted to the health center for in-patient treatment and adjust the treatment as necessary;
- Record details of all patients in the registry.

### During an Outbreak

- Instruct all health facilities to switch to the outbreak treatment protocol – single dose ceftriaxone or oily chloramphenicol.
- Launch a public information campaign informing communities of the availability of free treatment in government health centers;
- Monitor supplies of antibiotics and restock health facilities as stocks become limited.

## Table 2: Antimicrobials to treat bacterial meningitis

### Supportive therapy

Fluid and electrolyte balance should be monitored and fluid replaced accordingly. If the patient is unconscious or vomits, and if an intravenous line cannot be placed, a nasogastric tube should be inserted.

### Planning for Antibiotic and other supplies required for case:

The antibiotics and the other supplies that are required for the treatment of meningococcal meningitis should be estimated and calculated based on the previous history of outbreak of the disease and also the current outbreak (see annex 9 for detail information).

### 3.7.2 Vaccination

In order to interrupt the spread of the outbreak and prevent an outbreak, WHO recommends large-scale vaccination of population groups that are at risk with the appropriate polysaccharide vaccine for the meningococcal serogroup that is responsible for the outbreak.

As soon as the epidemic threshold is reached in a woreda, it is recommended to conduct mass immunization campaign targeting the entire woreda, using the appropriate polysaccharide bivalent (AC) or trivalent (ACW) vaccine, and immunize all 2-30 years old population of the woreda. It is also recommended to vaccinate any contiguous woreda in alert phase.

#### What should be done during the epidemic phase:

1. Vaccinate immediately the epidemic woreda with the appropriate vaccine as well as any contiguous woreda in alert phase.
2. Continue data collection, transmission and analysis.
3. Maintain regular collection of 5 to 10 CSF specimen per week through out the epidemic season in the epidemic woredas in order to detect any serogroup shift.
4. Treat all case with the appropriate antibiotic as recommended by the National protocols.
5. Prepare and communicate micro plan and budget for each area targeted for mass vaccination.
6. Establish a cold chain to distribute the vaccines to the target areas.
7. Launch a public information campaign to inform all the communities in the target areas.
8. Vaccination targeted age group (2-30), which means 70% of the population.
9. Monitor adverse events following vaccination.
10. Prepare to manage the waste from the campaign.
11. Coordinate and oversee each vaccination campaign.

## Criteria for vaccine choice

The decision on the type of vaccine to be used should ideally be based on the results from at least 10 Nm positive specimens.

In order to obtain that number of Nm positive specimens, it is estimated that 20 to 30 CSF specimens should be collected from the affected area. Efforts should be made to collect and test CSF specimens in the field as early as possible.

The proportion of Nm W135 required warranting the use of ACW trivalent vaccine could be defined according to the number of Nm positive samples available from a given affected area.

The following criteria could be suggested:

- 30% of W135 out of 10-19 Nm positive samples.

OR

- 20% of W135 out of 20 or more Nm positive samples.

In the total absence of laboratory evidence of Nm W135 the use of trivalent ACW vaccine should be strongly discouraged.

In the above-mentioned situation, vaccination with bivalent AC vaccine should be recommended (provided that some laboratory evidence of Nm A is available).

In situations where a full blown outbreak is reported and where the minimum percentage of Nm W135 was not reached, the identification of one or more Nm W135 in the concerned area(s) and concurrent W135 outbreak in contiguous area(s) will justify the use of the trivalent vaccine.

In any other situation, decisions to use vaccine should be evaluated on a case-by-case basis and should take into account all epidemiological and laboratory information available (see Annex 7).

## Micro-planning reactive vaccination campaign

Micro planning is the critical step to be taken before starting vaccination campaigns. The following assumptions can be used during preparation of this plan.

## 1. Calculating target population for vaccination:

- a. For the general population target those of age 2-30 years (which is nearly 70 % of the general population). Hence target population = 70% x total population of the target kebeles or woreda)
- b. For closed settings like prisons, orphanage, universities, schools etc, the target population is equal to total population of that setting except for age less than 2 years old. i.e, approximately 100% of that setting;

## 2. Calculating vaccine and other supplies required:

The demand of vaccine and supplies depends on the target population, the coverage rate intended (100%) and potential wastage rate. These calculations can be done (automated) by using computers on excel spread sheet.

- a. Vaccine = target population + 17% wastage.
- b. AD syringe = target population + 10% wastage.
- c. Mixing syringe = number of vials.
- d. Safety box = (AD + mixing syringes) /100 (1 box can hold 100 syringes of AD and mixing syringes).
- e. Vaccine carrier = at least 1 per vaccination posts.
- f. Cold box = 1 for 1200 vial.
- g. Tally sheet = 3 sheets per day for each vaccine posts.
- h. Megaphone = one per kebele.
- i. Batteries for Megaphone = 8 dry cell per day.
- j. Supervisors sheet = 1 sheet per day.
- k. Cotton = at least 1 role for each vaccine posts.
- l. Waste collection big plastic bags(Biohazard) = 2 per day for each vaccine posts.

**Table 3 example on how to calculate vaccine required**

Main assumptions	How to calculate
For an estimated population of about	50,000 people
Target population of 2 - 30 years of age (70% of total population)	$50,000 \times 0.7 = 35,000$ people
Goal of vaccine coverage 100%	$35,000 \times 1.0 = 35,000$ people
Number of doses to administer (1 dose per person)	$35,000 \times 1 = 35,000$ doses
Number of doses needed assuming wastage rate of 17%	$35,000 \times 1.17 = 40,950$ doses
Number of doses needed assuming need for a reserve of 25%	$40,950 \times 1.25 = 51,875$ doses Approximately 52,000 doses should be ordered.
For the vial of 10 doses	5,200 vials will be needed

(For additional information see Annex 1)

- 3. Human resource:** for the operation to be successful the following minimal number of personnel are required for each team considering active participation of the local authorities and police.
- a. Assumption 1 team vaccinates = 1000 people per day
  - b. Coordinator = 1 at woreda level
  - c. Cold chain professionals management = 2 per woreda
  - d. Vaccination team supervisors = 1 supervisor for 5 teams (vaccination posts)
  - e. Vaccination team members in each post = 5
    - i. vaccinators = 2 (1 preparing and the other injecting),
    - ii. record keeper = 1
    - iii. crowd controller = 1 (additional support from the local police station)
    - iv. social mobilizer = 1
  - f. Drivers- minimum 1 for each supervisor



4. **Operational Cost:** the operation cost may include perdiems, transportation or fuel and maintenance cost, training costs, social mobilization and other costs.

### 3.7.3 Information, Education & Communication

An outbreak of meningococcal meningitis can be more quickly controlled when the public understands how to help to limit the spread. Health education is crucial to ensure the participation of the community.

- Select the best way to disseminate messages to the community:
- Communication through radio, posters, talks, etc.
- In the local language.
- Give clear information – but not too many messages.
- Adapt messages to the social, cultural, and economic circumstances of the community and to its ability to cope with a change of behavior (for example, care for cases).
- Organize talks in places where people are usually waiting (health care facilities, hairdressers, etc.).

Informing the public is an integral and important part of meningococcal meningitis control strategies: the population must be informed of the outbreak and of the measures to be taken, including the importance of early case identification. Consult local authorities to adapt the messages to the local context and to know which media methods are most appropriate in each specific context. Local language should be used.

Rumors regarding sources of the disease and individual protection are frequent during meningococcal meningitis outbreaks, especially in areas not previously hit by the disease. Public information should clarify these rumors with specific, adapted messages – aimed both at those potentially spreading false information (e.g. religious leaders, traditional healers), and those who may receive it.

For such rumors, it is important to coordinate with local authorities before discussing with target groups.

1. Come to the health care facility as soon as possible in case of meningococcal meningitis.
2. Use infection prevention measures at all levels.

### 3.8. Communicate Findings

Your final task in an investigation is to communicate your findings to others who need to know. This communication usually takes two forms: 1) an oral briefing for local health authorities and 2) a written report.

## SECTION 4. COORDINATION

The overall planning and coordination strategy for outbreak meningitis control should take place at the woreda level. It is the responsibility of the local health authorities but requires the input of a wide range of partners. Experience has shown that establishing a committee for epidemic preparedness and response (EPR Committee), well in advance of the outbreak season, is the most effective way to plan, coordinate and supervise the activities of multiple partners to ensure outbreaks are detected early and an appropriate response is launched promptly. The EPR Committee should be led by respective representatives from each level, and should include staff from key hospitals in the area, reference laboratories and other partners who may be involved in treating patients and monitoring outbreaks. The EPR Committee should meet regularly prior to and throughout the outbreak season.

### EPR Committee

The committee may consist of representatives from the following organization and the representation depends on the availability of organizations present at each level:

- Ministry of Health (including personnel from the PHEM center and the Health Promotion and Diseases Prevention directorate).
- Referral hospital for meningitis.

- Reference laboratory.
- Other hospitals in the affected area(s).
- Head of national logistic and drug supply.
- Expanded Programme on Immunization (EPI)
- Mobile teams.
- Non-governmental organizations (NGOs) involved in health care.
- Technical assistants as needed.

### **The Role of EPR Committee:**

- Plan control strategies.
- Define populations at risk.
- Develop policies and sustain executive structures with clear responsibilities for Emergency health response.
- Assign specific responsibilities to individuals or units for outbreak detection and response.
- Establish procedures to rapidly mobilize mass immunization programs.
- Identify the important resources needed for rapid outbreak response, and update information on these resources at local and national levels.
- Estimate the requirements to control the outbreak (drugs, vaccine, human resources, transport, financial resources).
- Establish procedure for accessing funds.
- Identify and ensure that competent laboratory support remains available in the country.

- Coordinate communication with and education of the health care community And the general public.
- Supervise and coordinate implementation and achievement of control measures.
- Evaluate and follow up the results, adjust strategy if necessary, draw up the post-outbreak review.
- Keeps the general public informed on the risks of meningitis, where they can seek treatment and any plans for vaccination in their areas.

## SECTION 5. MONITORING AND EVALUATION

### 5.1 Monitoring

Monitoring of activities enables correct outbreak management, analysis of results and identification of any problems. It should be carried out from the start to the end of the outbreak on a daily (e.g. monitoring AR, CFR, woreda that has crossed an alert or epidemic threshold, curative case management, vaccination activities) basis. Data with which to calculate indicators are routinely collected and analyzed in detail at different levels, more specifically at woreda level. This includes vaccination coverage, vaccine utilization rate availability of treatment etc.

Vaccination tally sheets are filled in every day on vaccination sites in order to estimate the vaccination coverage. Thus daily analysis is used to evaluate the results. If the results are not satisfactory, identify the causes (i.e. shortage of vaccine or medical supplies, too few teams, lack of public information, poor choice of sites (access) etc) and if necessary, take corrective measures (in areas of supplies, duration of campaign on a site, number of teams, public message information etc).

### 5.2 Evaluation

A final evaluation report of the intervention (evaluation of surveillance, case management and vaccination activities) should provide constructive recommendations for future outbreak responses.

## Post outbreaks follow up

Meningococcal meningitis outbreak is declared to be over when the attack rate descends below the alert threshold over two consecutive weeks. Once that point has been reached, a number of follow-up activities are needed:

- Continue weekly reporting of both cases and laboratory results to monitor decreasing trends;
- Gather remaining stocks of antibiotics or reposition for use in treatment for other conditions;
- Return any remaining stocks of vaccines to woreda stockpiles;
- Conduct a vaccination coverage survey;
- Evaluate the outbreak response and complete a report on the outbreak;
- Provide feedback to stakeholders and document lesson learned.

## SECTION 6. ANNEXES

### Annex 1. Preparing Micro-plan for Vaccination Campaign

Preparing a vaccination micro-plan

A micro-plan must be prepared for every woreda targeted for a vaccination campaign. It is the responsibility of the woreda health authorities to complete and submit the plan in order to prepare thoroughly for the campaign and to secure the necessary vaccines.

#### The micro-plan should include:

- Names of kebeles targeted for vaccination.
- Total population currently in target areas.
- Population targeted for vaccination.
- Quantity of vaccine needed.
- Quantity of additional supplies need – AD syringes, safety boxes, dilution syringes (10 ml), cotton wool, gloves.
- Number of teams conducting the campaign (each team requires vaccinators, recorders, crowd controllers and a supervisor).
- Number of supervisors at team, woreda, zone, region and central levels.
- Training of vaccination teams.
- Logistic needs – cold chain equipment, vehicles.
- Waste management.
- Plans for vaccination campaign coverage surveys.

#### The budget should include:

- Allowances for members of the vaccination team.
- Social mobilization costs (including allowances for staff).
- Costs of logistic equipment.
- Costs of waste management.

## Organizing a vaccination session

### Social mobilization

Success of the campaign will be guaranteed by simple elements such as:

- Adequate communication with the public.
- Good organization.

Creation of a mobilization committee improves efficiency. The committee can include:

- Political and administrative representatives.
- Neighborhood representatives.
- health authorities.
- A police chief.

Depending on the size of the outbreak and the location, information will be transmitted by:

- The media (radio, television).
- Megaphones in the villages.
- Woreda chiefs.
- Community health workers in health facilities.
- Religious leaders.

The message will describe:

- The illness and its complications.
- The importance to detect cases and their referral to a hospital.
- The advantages of vaccination and expected side-effects.
- The age groups to be vaccinated.
- The location and time of vaccinations.
- The importance to bring one's EPI vaccination card.

## Annex 2. Materials and Techniques Needed for Gram Stain and Methylene Blue Stain

### Materials

95% ethyl alcohol 100 ml

Working solution:

Safranin stock solution 10 ml

Distilled water 90 ml

**Ziehl-Nielsen carbol-fuchsin (considered by many to be a more effective counter stain than safranin)**

Basic fuchsin 0.3 g

95% ethyl alcohol 10 ml

Phenol crystals, melted 5 ml

Distilled water 95 ml

Dissolve the fuchsin in alcohol. Add the 5% phenol solution.

Let stand overnight. Filter through paper.

Gram stain kits or individual reagents are also available commercially from several laboratory supply companies.

**Methylene blue stain (if Gram stain is not possible):**

Methylene blue 0.3 g

95% ethyl alcohol 30 ml

Distilled water 100 ml

Dissolve methylene blue in alcohol. Then add distilled water



## Handling of cerebrospinal fluid (CSF):

At least 20 drops (1 ml) of CSF should be collected in a sterile tube. Do not refrigerate but hold at room temperature before staining. Processing should take place as soon as possible after collection.

1. Centrifuge CSM at 2000 rpm for 10 minutes.
2. Draw off the supernatant and reserve for antigen detection, or other tests.
3. Use a drop of the sediment to make a smear on a glass slide. Air dry, fix gently with heat by passing through a flame.
4. Stain the smear.

## 2. Techniques for Gram stain and methylene blue stain

### Gram stain:

- a. Flood the smear with ammonium oxalate-crystal violet and let stand for 1 minute.
- b. Rinse gently with tap water. Drain off excess water.
- c. Flood smear with Gram iodine solution and let stand for 1 minute.
- d. Rinse with tap water as in step b.
- e. Decolorize with 95% ethanol (5-10 seconds may be enough).
- f. Counter stain with safranin 20-30 seconds or carbol-fuchsin 10-20 seconds.
- g. Rinse the slide with tap water and blot dry.

### **Methylene blue stain:**

- a. Flood the smear with methylene blue solution and let stand for 1-3 minutes.
- b. Rinse and blot dry.

### **Results:**

Examine the smear under oil-immersion with a microscope equipped with a bright-field condenser. Meningococcal may occur intra- or extra-cellularly, and appear as Gram-negative, coffee bean shaped diplococci.

# Annex 3. Case –based Report Form (CRF)

Reporting Health Facility:				Reporting Woreda: _____ Zone _____				
Disease Type (Put Click mark ✓)				REGION:				
Anthrax	Cholera	Measles	Meningitis	Neonatal Tetanus	Hemorrhagic Fever	Yellow Fever	Others (Specify)	
Name of Patient: _____								
Date of Birth (DOB): ____/____/____ (Day/Month/Year)			Age (If DOB is unknown)		Year	Month (if <2)		
Sex:	Write M for Male and F for Female							
Patient's Address:		Kebele:		House number:				
Woreda:		Zone:		Region:				
Locating information:		Location when symptom started		Current Location				
If applicable or if the patient is neonate or child, please write full name of mother and father								
Date seen at Health Facility ____/____/____			Date Health Facility notified Wored /Zone ____/____/____		Date of Onset: ____/____/____			
Number of Vaccine/TT dose received:			For cases of NNT*, Measles, Yellow Fever & Meningitis (For NNT, Measles, Yellow Fever – refer immunization card & for Meningitis ask history). *For NNT case please complete the additional case investigation form					
Date of last vaccination:			____/____/____ (NNT, Measles, Yellow Fever & Meningitis only)					
Associated with Epidemics			1. YES	2. NO				
In/Out Patient			1. In patient	2. Outpatient				
Treatment given			1. YES	2. NO				
Outcome of the patient at the time of report			1. Alive	2. Dead	3. Unknown			
Fill only if specimen is collected and sent to the lab								
Date of specimen collection: ____/____/____			Date of specimen sent to lab: ____/____/____					
Type of Specimen: (Put Click mark ✓)		Stool	Blood	Serum	CSF	Throat Swab	Other (Specify)	
Date form sent to Woreda: ____/____/____ (Day/Month/Year-EC)								
Name and Signature of the person completing the form _____ Tel: _____								
For official Use only								
ID Number		Date form received at National Level ____/____/____ (DD/MM/YY-EC)						
Final Classification of Case		1. Confirmed		2. Probable		3. Discarded	4. Suspect	
Final Classification of Measles		1. Laboratory confirmed	2. Confirmed by Epidemiological link		3. Clinical compatible		4. Discarded	
Name and Signature of the Official: _____		Date(EC) _____						

**For Health Facility: If lab specimen is collected, complete the following information. And send a copy of the form to the lab with the specimen.**

Date of specimen collection: \_\_\_\_/\_\_\_\_/\_\_\_\_ Specimen source: Stool Blood CSF

Date Specimen sent to lab: \_\_\_\_/\_\_\_\_/\_\_\_\_ Other

For the Lab: Complete this section and return the form to woreda team and clinician

Date lab specimen: \_\_\_\_\_ Specimen Condition: Adequate Not adequate

Disease/Condition	Type of test	Results (P= pending)	Disease /Condition	Type of test	Results	Virus Detection	
Cholera	Culture	+ - P	Yellow Fever	IgM	+ - P		
	Direct Exam	+ - P	Measles	IgM	+ - P		
				Method used for Direct Exam	Rubella	IgM	+ - P
<b>Meningitis</b>							
N. meningitidis	Culture	+ - P	RVF	IgM	+ - P	+ - P	
S. pneumonia	Culture	+ - P	Ebola	IgM	+ - P	+ - P	
H. influenza	Culture	+ - P	CCHF	IgM	+ - P	+ - P	
N. meningitidis	Latex	+ - P	Lassa	IgM	+ - P	+ - P	
S. pneumonia	Latex	+ - P	Marburg	IgM	+ - P	+ - P	
H. influenza	Latex	+ - P					
Shigella Dysenteriae	Culture	SD type 1 Other shig No shig					
Plague	Culture	+ - P					
	IFA > 1: 64	+ - P					

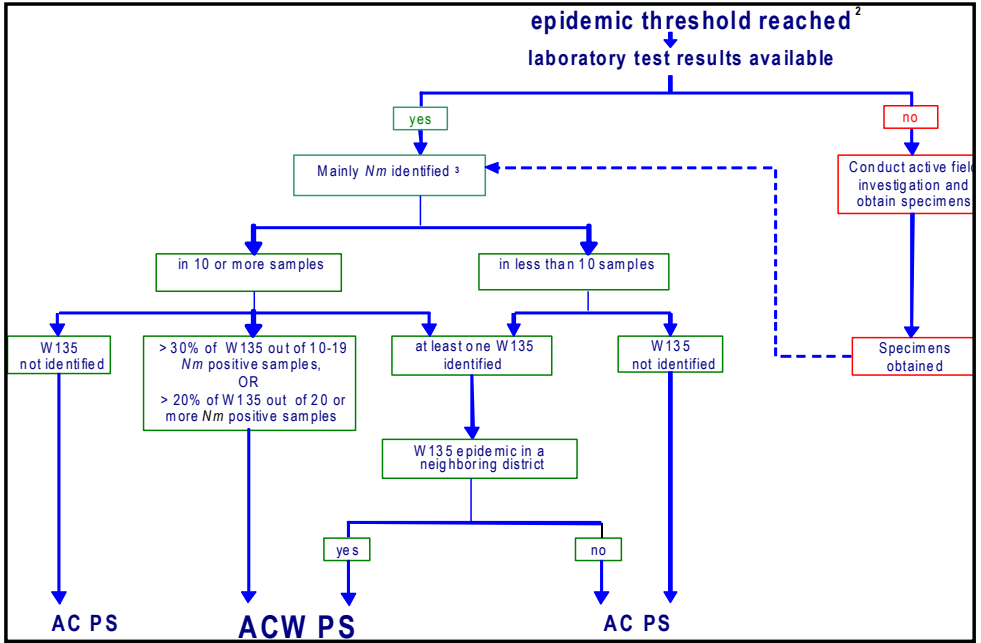
Date lab sent results to woreda: \_\_\_\_\_ Other lab results: \_\_\_\_\_

Name of lab sending results: \_\_\_\_\_ Other pending tests: \_\_\_\_\_





## Annex 7. Decisional Tree for the Use of ACW Trivalent Polysaccharide (PS) Vaccine



## **Annex 8: Instructions on Using Trans-Isolate (TI) Bottles**

How to use the Trans-Isolate (T-I) system for isolation and transport of meningococci and other agents causing bacterial meningitis from CSF

### **1. Procedure for inoculating T-I medium for transporting meningococci and other agents causing bacterial meningitis from CSF:**

- Remove a bottle of Trans-Isolate (TI) medium from refrigerator at least 30 minutes before inoculating it with the specimen. Allow the bottle to warm to room temperature which is more favorable for growth of the organism.
- Before inoculating the bottle, check to see if there is any visible growth or turbidity. If there is visible growth or turbidity, discard the bottle, because it may be contaminated.
- Lift up the small lid in the middle of the metal cap on top of the TI bottle.
- Disinfect the top of the TI bottle with 70% alcohol or iodine. Allow to dry (usually 30 to 60 seconds).
- Use a sterile syringe and sterile needle preferably 21G, 0.8 mm. To aspirate 500 microliters (one-half of an ml) of cerebrospinal fluid (CSF) from the tube containing CSF.
- Inject the CSF into the TI bottle through the disinfected dry stopper on the top of the TI bottle.

### **2. Transport and incubation of TI bottles, and inoculation of the culture media**

The procedures to follow depend upon how promptly the TI bottles can reach the laboratory of reference that will perform culture and isolation.

If TI bottles cannot reach the laboratory of reference within 24 hours:

- Label the TI bottle with the date, name of the patient, and any other necessary identifiers.



- Ventilate the TI bottle with a sterile cotton plugged needle. The Needle should not dip into the culture media (broth).
- Store the ventilated TI bottle in an upright position at room temperature. Make sure it is away from excessive heat, direct sunlight, and dust.
- Before transporting the bottle, remove the ventilating needle from the top of the TI bottle. This will prevent leakage and contamination during shipment.
- Transport the TI bottle in a sealed plastic bag to minimize the risks of contamination and attach the case report form.

If TI bottles can reach the laboratory of reference within 24 hours:

- Label the TI bottle with the date, name of the patient, and any other necessary identifiers.
- Ship the TI bottles without ventilation.
- Transport the TI in a sealed plastic bag to minimize the risk of contamination and attach the case report form.

### ***3. Additional recommendations about the proper use of TI bottles and ventilating the inoculated TI bottles:***

- The TI bottles can be used for at least 1 year after the date of production provided that they are stored in the refrigerator.
- Freezing TI bottles destroys the TI medium.
- Non-inoculated TI bottles should be packed in cold packs for shipment to the laboratory of reference.
- Contamination is the single most problematic point with the system. Aseptic measures and understanding the risks are necessary to achieve good recovery of the isolates.

**The table below shows the general principles to use for the estimation assuming an attack rate of 150/100,000 and cases seen being 56 for the population indicated.**

<b>Formula</b>	<b>Example</b>
Total population in the woreda affected	295,484
Likely cumulative attack rate for season (based on past outbreak)	150/100,000
Estimated number of cases during season (population x cumulative AR)	$295,484 \times 150/100,000 = 443$
Minus number of actual cases reported.	443 minus 56 = 387
Plus additional 25% contingency stock (387 x 25%)	387 plus 97 = 484
<b>Antibiotics needed:</b>	
Ceftriaxone (4 vials per treatment) plus needles and syringes	$484 \times 4 = 1,936$ vials
Oily chloramphenicol (6 vials per treatment) plus water for injection, needles and syringes. Note that oily chloramphenicol is an alternative drug to Ceftriaxone.	$484 \times 6 = 2904$ vials
Lumbar Puncture kits (30 LP set +25% wastage) per woreda	38
Diagnostic kits (Pastorex®) of 25 test	2 kits per woreda
Trans-isolate media	38



