# Blood Culture: Study and Statistics

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**ABSTRACT-** Blood culture test is done in the laboratory in which blood is taken from the patient which is then inoculated into bottles containing culture media to find whether infection causing micro- organisms (bacteria or fungi) have conquered the patient's bloodstream. The complete process of blood culture consists of- collecting the sample correctly, detecting and identifying micro-organisms causing bloodstream infections and providing an antibiotic susceptibility test result for the clinician. Sepsis is considered as one of the challenges in critical care, and early diagnosis of the same is one of the most decisive factors in determining outcome of the patient. Blood culture is the most recurrent diagnostic tool available for the detection of bacteremia and fungemia a. It is a very important inspection with major inference for the diagnosis and treatment of patients with infections of bloodstream and sepsis. A blood culture which is positive establishes or confirms that there is an infectious etiology for the patient's sickness. It will also give the etiologic agent for antimicrobial susceptibility testing enabling optimization of antibiotic therapy. Beginning early and effective antibiotics can have a notable impact on the outcome of the disease. For the blood culture process to be effective and proper, however, it is very crucial that samples are collected suitably. Acquiescence with blood culture collection can improve the quality and clinical value of blood culture investigations and surely decrease the incidence of sample contamination and "false-positive" readings. Blood cultures should always be requested when an infection in the bloodstream or sepsis is suspected. Symptoms in a patient are which may lead to a suspicion of a bloodstream infection are undetermined fever, shocks, chills, rigors, severe local infection, abnormally raised heart rate, low blood pressure and raised respiratory rate. Blood cultures should be collected as early as possible after the onset the symptoms. Ideally, they should be obtained prior to the execution of any kind of anti-microbial therapy. If the patient is already on an anti-microbial therapy, blood cultures should be taken immediately right before administering the next dose. The main objective of the training was to collect the blood samples of around thousand patients and determine the adequate blood volume which is 1-4 ml in children and 8-10 ml in adults. The suspected organisms causing bacteremia were found to be 59% for gram negative, 38% for gram positive and 3% for fungus. A histogram was prepared showing adequate blood volume for adults and children.



## 1.INTRODUCTION

Blood cultures are used to detect the presence of bacteria or fungi in the blood, to identify the type present, and to guide treatment. Testing is used to identify a blood infection (septicemia) that can lead to sepsis, a serious and life-threatening complication. Individuals with a suspected blood infection are often treated in intensive care units, so testing is often done in a hospital setting.

Although blood samples may be used to detect viruses, this article focuses on the use of blood cultures to detect and identify bacteria and fungi.

Other related tests that may be performed include:

- Gram stain—a relatively quick test used to detect and identify the general type of bacteria
- Susceptibility testing—determines the drug (antimicrobial) that may be most effective in treating the infection

Often, a complete blood count (CBC) is ordered along with or prior to the blood culture to determine whether the person has an increased number of white blood cells, indicating a potential infection. Sometimes other testing is also performed, such as a chemistry panel to evaluate the health status of a person's organs, or a urine, sputum, or cerebrospinal fluid (CSF) culture to help identify the source of the original infection. This is especially true when a person has symptoms associated with a urinary tract infection, pneumonia, or meningitis. A health practitioner may order blood cultures when a person has signs and symptoms of sepsis, which indicates that bacteria, fungi, or their toxic by-products are causing harm in the body. A person with sepsis may have:

- Chills, fever
- Nausea
- Rapid breathing, rapid heartbeat
- Confusion

As the infection progresses, more severe symptoms may develop, such as:

- Inflammation throughout the body
- The formation of many tiny blood clots in the smallest blood vessels

- A dangerous drop in blood pressure
- The failure of one or more organs

When a person has had a recent infection, surgical procedure, prosthetic heart valve replacement, or immunosuppressive therapy, the person is at a higher risk of a systemic infection and drawing blood cultures would be appropriate when symptoms are present. Blood cultures are drawn more frequently in newborns and young children, who may have an infection but may not have the typical signs and symptoms of sepsis.

Two or more blood cultures that are positive for the same bacteria or fungi means that the person tested likely has a blood infection with that microorganism. The results typically identify the specific bacteria or fungi causing the infection.

Blood infections are serious and need to be treated immediately, usually in a hospital. Sepsis is a complication that can be life-threatening, especially in people with weakened immune systems. Health practitioners who suspect sepsis may begin patients on intravenous broad spectrum antibiotics that are effective against a wide range of bacteria while waiting for the blood culture or susceptibility testing results. When results become available, the treatment may be changed to an antimicrobial agent that is more specific for the bacteria or fungi detected in the blood cultures.

If one blood culture set is positive and one set is negative, it may mean that an infection or skin contaminant is present. The health practitioner will consider the person's clinical status and the type of bacteria or fungi found before making a diagnosis. Also, additional testing may be warranted in this case.

Blood culture sets that are negative after several days (often reported as "no growth") mean that the probability that a person has a blood infection caused by bacteria or fungi is low. If symptoms persist, however, such as a fever that does not go away, additional testing may be required. A few reasons that symptoms may not resolve even though blood culture results are negative may include:

- Some microorganisms are more difficult to grow in culture, and additional blood cultures using special nutrient media may be done to try to grow and identify the pathogen.
- Viruses cannot be detected using blood culture bottles designed to grow bacteria. If the health practitioner suspects that a viral infection may be the cause of the person's symptoms, then other laboratory tests would need to be performed. The tests that would be ordered depend upon the person's clinical signs and the type of virus the health practitioner suspects is causing the infection.

Results from other tests that may be done in conjunction with blood cultures may indicate sepsis even though blood cultures may be negative. Some of these may include:

- Complete blood count (CBC). An increased white blood cell (WBC) count may indicate infection.
- Complement. Levels of C3 may be increased.
- A urine or sputum culture may be positive, indicating a possible source of infection that may have spread to the blood.
- A CSF analysis may reveal a possible source of infection.

Sepsis means that the bacteria or fungi have spread throughout the body, an affected person may experience many different symptoms of illness. As the immune system works to fight the infection, it produces many factors to kill the bacteria or fungi that can also make a person feel sick. Septicemia can cause a fall in blood pressure (shock), a rapid heart rate, and a decrease in blood flow to the brain, heart, and kidneys. It can also affect blood clotting factors, leading to disseminated intravascular coagulation (DIC), which can cause generalized bleeding. Bacteria in the blood may also spread to the joints and cause septic arthritis.

Rapid tests are available that can detect several different types of bacteria that are commonly known to cause infections of the blood. These tests are used in follow up to positive blood cultures to quickly identify the bacteria that are present. They can identify types such as methicillin-resistant *Staphylcoccus aureus* (MRSA), which is typically difficult to treat, and gram negative rods such as *E. coli* that live in the gastrointestinal

tract. Rapid Identification can facilitate treatment with appropriate antibiotics.

## 2.NORMAL AND ABNORMAL RESULTS

A normal value means that no bacteria or other germs were seen in your blood sample. An abnormal (positive) result means that germs were identified in your blood. The medical term for this is bacteremia. This can be the result of sepsis. Sepsis is a medical emergency and you will be admitted to a hospital for treatment. Other types of germs, such as a fungus or a virus, may also be found in a blood culture.

Sometimes, an abnormal result can be due to contamination. This means bacteria may be found, but it came from your skin or from the lab equipment, instead of your blood. This is called a false-positive result. It means you do not have a true infection. Veins and arteries vary in size from one patient to another, and from one side of the body to the other. Obtaining a blood sample from some people may be more difficult than from others.

Other risks associated with having blood drawn are slight, but may include:

- Excessive bleeding
- Fainting or feeling lightheaded
- Hematoma (blood accumulating under the skin)
- Infection (a slight risk any time the skin is broken)

A blood culture is done to:

- Find a bacterial infection that has spread into the blood, such as meningitis, osteomyelitis, pneumonia, a kidney infection, or sepsis. A culture can also show what type of bacteria is causing the infection.
- Find a fungal infection, such as yeast, in the blood.
- Check for endocarditis, which is an infection of the valves of the heart.
- Find the best antibiotics to kill the bacteria or fungi. This is called sensitivity testing.
- Find the cause of an unexplained fever or shock or a person becoming extremely ill.

**Blood Culture**

| Blood culture | |
|---|---|
| **Normal:** | No bacteria or fungus is found. Normal culture results are called negative. |
| **Abnormal:** | Bacteria or fungus grows in the culture. Abnormal culture results are called positive. |

If bacteria are found in the culture, another test is often done to find the best antibiotic that will kill the bacteria. This is called sensitivity or susceptibility testing. Sensitivity testing is important so the blood infection is treated correctly. This also helps prevent bacteria from becoming resistant to antibiotics.



## 3.METHOD

A minimum of 10 ml of blood is taken through venipuncture and injected into two or more "blood bottles" with specific media for aerobic and anaerobic organisms. A common medium used for anaerobes is thioglycollate broth.

The blood is collected using aseptic technique. This requires that both the tops of the culture bottles and the venipuncture site of the patient are cleaned prior to collection by swabbing with 70% isopropyl alcohol (povidone and left to dry before venipuncture).

To maximise the diagnostic yield of blood cultures, multiple sets of cultures (each set consisting of aerobic and anaerobic vials filled with 3–10 mL) may be ordered by medical staff. A common protocol used in US hospitals includes the following:

- Set 1 = left antecubital fossa at 0 minutes
- Set 2 = right antecubital fossa at 30 minutes
- Set 3 = left or right antecubital fossa at 90 minutes

Ordering multiple sets of cultures increases the probability of discovering a pathogenic organism in the blood and reduces the probability of having a positive culture due to skin contaminants.

After inoculating the culture vials, advisably with new needles and not the ones used for venipuncture, the vials are sent to the clinical pathology microbiology department. Here the bottles are entered into a blood culture machine, which incubates the specimens at body temperature. The blood culture instrument reports positive blood cultures (cultures with bacteria present, thus indicating the patient is "bacteremia"). Most cultures are monitored for five days, after which negative vials are removed.

If a vial is positive, a microbiologist will perform a Gram stain on the blood for a rapid, general identification of the bacteria, which the microbiologist will report to the attending physician of the bacteremic patient. The blood is also subcultured or "subbed" onto agar plates to isolate the pathogenic organism for culture and susceptibility testing, which takes up to three days. This culture and sensitivity (C&S) process identifies the species of bacteria. Antibiotic sensitivities are then assessed on the bacterial isolate to inform clinicians with respect to appropriate antibiotics for treatment.

Some guidelines for infective endocarditis recommend taking up to six sets of blood for culture (around 60 ml).



## 4.LABORATORY INVESTIGATION OF BLOOD CULTURE SAMPLE

Bacteremia is the presence of bacteria in the blood stream. It may be transient, intermittent or continuous. Septicemia is the presence of bacteremia with the symptoms of systemic infection. Typical symptoms include pyrexia, chills, rigors, hypotension, shock and severe cases of multiple organ failure.

### 4.1 BLOOD CULTURE METHODS

A blood culture involves taking a sample of blood from the patient and inoculating blood into blood culture bottle, the bottle is then transported to the laboratory for incubation. The procedure has 3 steps:

- Sample taking
- Transport
- Processing

Once the culture bottles have been inoculated, they should be clearly labeled with the full patient details accompanying report. Forms should also include patient's symptoms and current or recent antibiotic therapy & sent to the laboratory as soon as possible. It is important that full clinical details are provided as this gives information to the laboratory staff on organisms likely to be encountered. Further, tests that may need to be carried out on positive bottles, and whether bottles require additional procedures such as extended incubation/terminal subculture. Blood culture bottle is inoculated on to culture media at the end of incubation period. Once the samples are taken and labeled, they must be transported to the laboratory as quickly as possible. They should be placed in a clearly labeled incubator provided and maintained by the microbiology laboratory. On no account should blood culture bottles be refrigerated. It may adversely affect some organisms, delay organism growth and lengthen time for the bottles to become positive. If no incubator is available, then they should be kept at ambient temperatures. Once received in the laboratory, samples should be loaded on to any automated system as soon as possible to allow positive samples to be identified.

### 4.2 BLOOD CULTURE COLLECTION

- Sterile blood sampling that is hand hygiene, sterile dressing pack, sterile gloves and clean skin with chlorhexidine.
- Take from fresh venipuncture/alternatively newly inserted peripheral cannulae and central lines.
- If line sepsis is suspected, culture should be taken from central line and peripherally.
- Blood placed into aerobic and anaerobic culture medium.
- Remove dust cap from culture bottles and top swabbed with alcohol before blood is injected.
- Maximum yield if tube is neither under-filled or over-filled.

### 4.4 NUMBER OF CULTURES

- 2 sets of cultures from separate sites before starting antibiotics is ideal.
- 3 cultures have a 96% sensitivity in detecting bacteremia.
- More than 4 cultures generally offer little additional benefit.
- Additional cultures may be beneficial if suspected endocarditis or pre-existing anti-microbial agents.
- Antibiotics should not be withheld pending in the critically ill septic patients.

**INCUBATION & CULTURE**

- Incubation at room temperature initially to avoid killing temperature sensitive bacteria.
- Turbid appearance may indicate microbial growth are placed in 2 holders with light passed through an alarm which sounds when significant microbial growth interrupts the passage of light through the bottles.
- All samples are sub-cultured on to agar plates to test and allow organism identification for antibiotic sensitivity testing.

**CONTAMINATION**

- Suspect if single random culture of bacillus species, coagulase negative staphylococcus or diptheroids.
- Less likely if grown in more than 1 bottle (another reason for taking multiple cultures from various sites).

**4.3 BLOOD CULTURE BOTTLES AND THEIR ADVANTAGES**

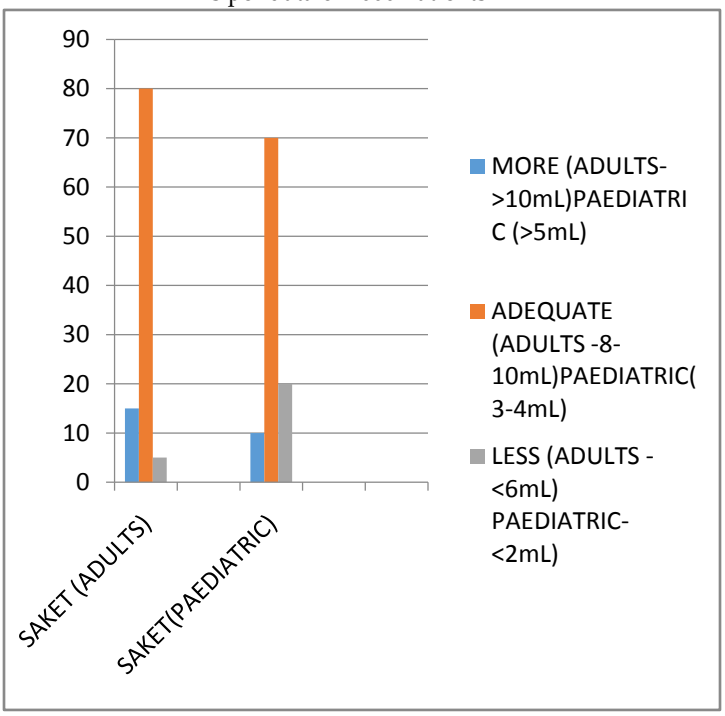
- These bottles reduce the potential for biohazard exposure.
- Lighter than glass.
- Cost effective storage, shipping and disposal.
- Multi-layer (gas impermeable design maintains aerobic and anaerobic environment)

Adapt to a variety of protocols and enables transport without a special container.

**5. RESULT**

**Volume of Blood Found**

As per data of 1000 Patients

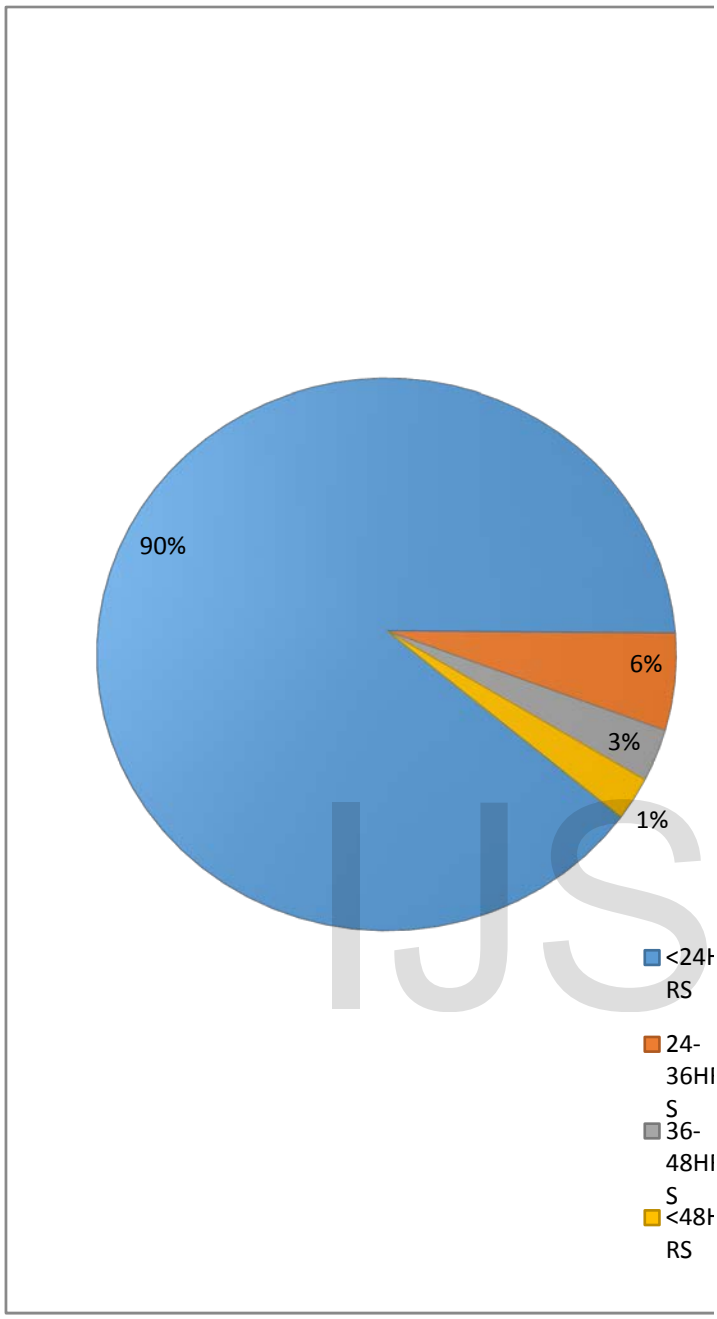



For the patients aged above 12 years, the blood volume was found to be around 15% which is more than 10 ml. The adequate blood volume is 8-10 ml which constitutes around 80%. Less than 6 ml is put into category of less volume and constitutes about 5%.

For the children below 12 years, the blood volume was found to be around 10% which is more than 5ml. The adequate blood volume of kids is 3-4 ml which constitutes around 70%. Less than 2 ml is put into category of less blood volume and accounts for around 20%

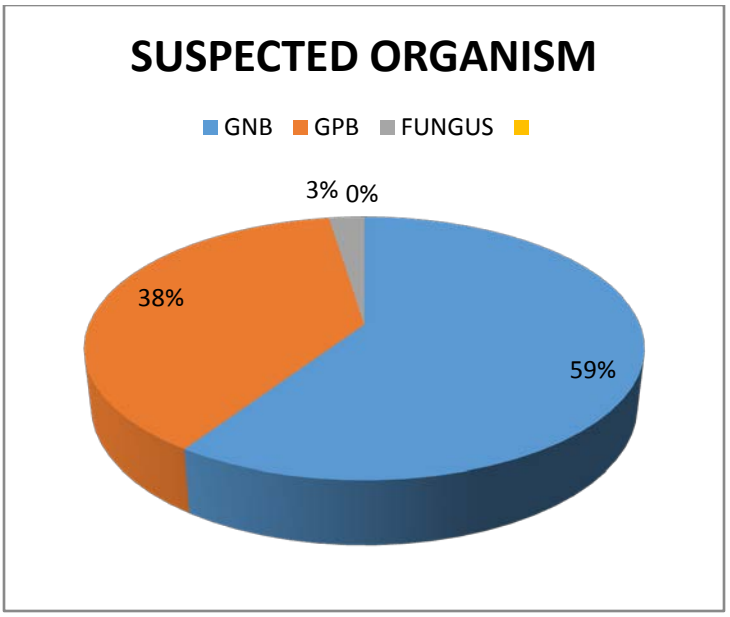
**Time taken for Positive Blood Culture**

As per data of 1000 Patients




The pie chart shows the timing in which the sample of blood culture showed positive result. More than 90% organisms showed positive result in more than 24 hours. About 6% showed positive result in 24-36 hours. 3% were detected in 36-48 hours and about 1% in more than 48 hours.

**Percentage of Cause for Bacteremia**




After collecting the data of thousand patients, 59% constituted the gram negative bacteria, 38% accounted for gram positive bacteria and only 3% were fungi.

## 6.DISCUSSION

It is totally automated test system for incubating and monitoring culture bottles for growth of microbes. The system consists of incubators, data management computers, growth media bottles along with reagents. In this, bacteria metabolises carbohydrate, produces energy and synthesizes low weight metabolites .Glycerol & oleic acid are often selected as carbon source, once ingested they are converted to acetyl CoA and are oxidized through the Krebs's cycle. Carbon dioxide and free electrons are the major byproducts of oxidation. The system utilizes a colorimetric sensor & reflected light to monitor the amount of carbon dioxide dissolved in the culture medium .There is liquid emulsion sensor (les) which is impermeable to most ions including H* ions and to components of media and whole as degraded blood. It is freely permeable to CO2. As CO2 diffuses across the membrane & dissolve in the water contained in the sensor, free hydrogen ions are generated. Free H* ions interacts and indicates in the sensor. As CO2 is produced, the concentration of H* ions increases & the p h decreases causing the sensor to change to light green or yellow. A led projects light into the sensor. The light reflected by the sensor is measured by a photodetector & as more 2 is generated, more red light is reflected.(lighter colors more reflect light than darker colors).the photodiode converts the reflected light into an electron signal that is measured every 10 minutes & plotted every hour as a point on a graph.

Mathematical algorithm analyse the readings & slope of the curve over time to determine positive & negative. For the patients aged above 12 years, the blood volume was found to be around 15% which is more than 10 ml. The adequate blood volume is 8-10 ml which constitutes around 80%. Less than 6 ml is put into category of less volume and constitutes about 5%.For the children below 12 years, the blood volume was found to be around 10% which is more than 5ml. The adequate blood volume of kids is 3-4 ml which constitutes around 70%. Less than 2 ml is put into category of less blood volume and accounts for around 20%.More than 90% organisms showed positive result in more than 24 hours. About 6% showed positive result in 24-36 hours. 3% were detected in 36-48 hours and about 1% in more than 48 hours. After collecting the data of thousand patients, 59% constituted the gram negative bacteria, 38% accounted for gram positive bacteria and only 3% were fungi. Importance of blood volume includes increased sensitivity, high volume precision, high percentage of detection and decrease the turn around time.