TEST

Nitroblue tetrazolium blood test

A blood test that checks whether neutrophils can produce an oxygen burst used to kill certain germs.

WHAT IT MEASURES
Neutrophils' ability to make an infection-killing oxygen burst

SAMPLE
Blood

FASTING NEEDED?
No

HOW LONG IT TAKES
About 5 to 10 minutes

RESULTS IN
Several days, based on specialist laboratory schedules

USED FOR
Screening and diagnosis of chronic granulomatous disease

MAIN LIMITATION
Qualitative and operator-dependent; partial defects may be missed

Overview

The nitroblue tetrazolium blood test, often shortened to the NBT test, checks how well certain white blood cells called neutrophils work. Neutrophils help protect you from bacterial and fungal infections.

When activated, healthy neutrophils produce reactive oxygen chemicals. This is called an oxidative, or respiratory, burst. It helps the cells kill germs they have taken in.

In the laboratory, neutrophils are exposed to a stimulant and a pale yellow chemical called nitroblue tetrazolium. Cells with a working oxidative burst change the chemical into dark blue deposits that can be seen under a microscope.

The NBT test was widely used to investigate chronic granulomatous disease, or CGD. CGD is an inherited immune system condition in which phagocytes, including neutrophils, cannot kill certain germs normally. Many laboratories now use a dihydrorhodamine, or DHR, flow cytometry test instead because it gives more detailed and quantitative information.

Why the test is ordered

The main purpose of an NBT test is to look for a problem with the neutrophil oxidative burst. It may be ordered when CGD is suspected.

Reasons for considering this type of testing include:

The test does not identify the exact genetic change causing CGD. It also does not assess every part of the immune system.

- Repeated or unusually severe bacterial or fungal infections
- Infections caused by organisms that rarely affect healthy people
- Granulomas, which are clusters of immune cells causing inflammation
- A close relative with CGD or a known CGD-related genetic change
- An earlier immune test suggesting abnormal neutrophil function

Who should have the test

You may be offered the test if your medical and family history suggests a possible neutrophil function disorder. Children are tested more often because CGD commonly becomes apparent in childhood, but testing can also be appropriate for adults with milder or later-presenting forms.

Relatives of someone with CGD may need testing. However, DHR flow cytometry and genetic testing are usually more informative for identifying people who carry an X-linked CGD-related genetic change.

NBT is not a routine blood test for common infections. Its use is generally limited to specialist immune or blood cell laboratories.

How to prepare

You do not usually need to fast. You can generally eat, drink water, and take your usual medicines unless you have been given different instructions because other blood tests are being collected at the same time.

Tell the testing team about recent infections, immune treatments, stem cell transplantation, gene therapy, or granulocyte transfusions. Granulocyte transfusions provide donor white blood cells and can affect the result.

Do not stop antibiotics like amoxicillin or doxycycline , steroids like prednisone or hydrocortisone , or other prescribed medicines on your own. If a medicine could complicate interpretation, the laboratory or specialist team will decide whether the timing of the test should change.

The sample needs careful handling and may have to reach a specialist laboratory quickly. Your appointment may therefore be arranged for a particular day or time.

What happens during the test

A healthcare professional takes blood from a vein, usually in your arm. A band may be placed around your upper arm, and the skin is cleaned before a small needle is inserted.

Collecting the sample usually takes less than 10 minutes. The blood is then sent promptly to the laboratory because neutrophils need to remain alive and able to respond.

In the laboratory, the cells are usually tested after exposure to a substance that activates neutrophils. A laboratory professional examines the cells under a microscope and assesses how many contain the dark blue reaction product. Control samples help show whether the testing conditions worked properly.

Understanding your results

Results may report the proportion of neutrophils that changed the dye, the strength of the reaction, or simply whether the response was normal, reduced, or absent.

A strong blue reaction means the tested neutrophils produced an oxidative burst. Little or no colour change can suggest a problem in the enzyme system responsible for that burst.

There is no single universal reference range for every NBT method. Laboratories use different stimulants like methylphenidate or lisdexamfetamine , processing methods, control samples, and reporting systems. Interpretation can also be influenced by age, sex, pregnancy status, medicines, recent treatment, and the clinical context.

An abnormal result does not establish the exact cause by itself. It is usually checked with DHR flow cytometry and, when appropriate, genetic testing. A normal NBT result reduces the likelihood of classic CGD but may not exclude a milder or partial defect.

What affects the results

NBT testing relies on living neutrophils. Several factors can make the result misleading even when the laboratory equipment is working correctly:

Ordinary red blood cell transfusions do not supply neutrophils, but you should still report any recent transfusion so the type can be confirmed.

Routine meals and normal daily activity are not major influences. Because intense exercise can temporarily change white blood cell activity, follow any laboratory-specific instructions about strenuous exercise before sampling.

- Delayed transport: Neutrophils can lose function as a blood sample ages
- Incorrect temperature or handling: Poor storage can damage cells and produce an abnormally weak response
- Very low neutrophil count: There may not be enough cells for a reliable assessment
- Granulocyte transfusion: Normal donor neutrophils can temporarily mask the recipient's abnormal cells
- Stem cell transplant or gene therapy: Mixed groups of functioning and non-functioning cells can complicate interpretation
- Current illness or inflammation: Already activated neutrophils may change the unstimulated background reaction
- Partial genetic variants: Some neutrophils may produce enough activity to appear normal on a manual test
- Carrier patterns: A person carrying an X-linked change may have two cell populations that are easier to see with DHR testing

Accuracy and limitations

NBT testing can identify many people with a severe or complete failure of the neutrophil oxidative burst. However, it is a mainly visual, manual test. The result can be influenced by the observer, the stimulation method, and sample quality.

There is no single reliable percentage for how often NBT misses CGD or gives an abnormal result in someone without it. Methods differ between laboratories, and the test has largely been replaced by more precise techniques.

NBT is more likely to miss partial defects than DHR flow cytometry. It can also be falsely reassuring after a granulocyte transfusion or when functioning donor cells are present after transplantation. A damaged or delayed sample may look abnormal even when the person's neutrophils work normally.

DHR flow cytometry measures the response of thousands of individual cells. It can show the amount of oxidative activity and identify mixed cell populations more clearly. Genetic testing is the better test for confirming the responsible gene change, but it is usually interpreted alongside a functional test.

A normal NBT result does not rule out other immune deficiencies or other neutrophil disorders.

Risks and complications

The main risks are a small bruise or soreness where the needle entered. Some people feel faint during or shortly after a blood draw.

What happens next

A normal result may be reviewed alongside your infection history, examination, blood counts, and other immune tests. If concern remains, DHR flow cytometry or another specialist test may still be arranged.

A reduced or absent response usually needs confirmation with DHR testing. Genetic testing may then help identify the specific type of CGD and guide testing for relatives. Follow-up is normally arranged through an immunology, infectious diseases, haematology, or genetics service.

The test result itself is not an emergency. However, do not wait for the result if you become unwell. If you have a known or suspected immune deficiency, follow your existing care plan for fever or infection. Seek emergency help for severe breathing difficulty, confusion, collapse, or rapidly worsening illness.

Common questions

Can I drink coffee before the test?

Usually, yes. The NBT test does not require fasting. If other fasting blood tests are being collected, follow the instructions for those tests.

Does the time of day matter?

The neutrophil response is not usually interpreted using a particular time-of-day range. Collection may be scheduled early so the fresh sample can reach a specialist laboratory promptly.

Can I exercise beforehand?

Normal activity is generally acceptable. Ask about strenuous exercise because intense activity can temporarily affect white blood cell numbers and activation.

Will antibiotics affect the result?

Antibiotics do not correct the inherited oxidative burst defect being tested. Do not stop them unless the clinician who prescribed them tells you to do so.

Is the NBT test the same as the DHR test?

No. Both examine the neutrophil oxidative burst, but DHR uses flow cytometry and provides more detailed information. DHR is now preferred in many laboratories.

Can a normal result completely rule out CGD?

No. A manual NBT test can miss partial defects or mixed cell populations. If the medical history remains concerning, DHR flow cytometry and genetic testing may be more appropriate.

Sources

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