DR SPUR'S MYSTERY CASE

A case of an early bird catching the worm

Welcome to Dr Spur's Immunology Clinic
Referral letter:

Dr N Vanmali
Gynaecologist

421 Ceader Road
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Dear Dr Spur

I have a primigravida patient who is 32 weeks pregnant. On sonar it appears that she is carrying a boy.

She asked me at her most recent prenatal visit about newborn screening specifically for primary immunodeficiencies. The reason for her asking is because she has a maternal cousin who had a boy who passed away at the age of nine months with severe, overwhelming sepsis. A diagnosis was never confirmed, but the paediatrician said that all future newborn babies in the family should be screened for a primary immunodeficiency.

Is such a test available? How reliable is the test, and when should it be performed?

Thank you as always for your sound advice.

Kind regards

Dr N Vanmali
Gynaecologist

Dear Dr Vanmali

Thank you for asking this very important question.

During the past few years, neonatal screening assays have been developed to detect diseases hallmarked by the absence of T- or B-lymphocytes, classically seen in severe combined immunodeficiency (SCID) and severe agammaglobulinaemias, including X-linked agammaglobulinaemia (XLA) or Bruton's disease.

Babies with SCID appear healthy at birth, but without early treatment most of them die before the age of one year. A bone-marrow transplant before the age of three months gives the best chance of saving these babies' lives and curing this condition. It is crucial to make this diagnosis as early as possible, before the patients receive live vaccines or present with infections. This is because live vaccines and infections can be fatal, and also greatly diminish the success rate of a bone-marrow transplant.

Patients with agammaglobulinaemia, including XLA, usually present later in life, due to protective maternal antibodies. But they are at great risk for live vaccines, such as oral polio, which can lead to fatal infections. These patients are also at risk for severe infections, especially pulmonary infections. Pulmonary complications are the most common cause of mortality and morbidity in these patients. Early identification can prevent the administration of live vaccines and help with the early initiation of immunoglobulin replacement therapy. This can prevent pulmonary and other infectious complications.

T-cell receptor excision circles (TRECs) and kappa-deleting recombination excision circles (KRECs) can be used to detect T- or B-cell lymphopaenia. TRECs and KRECs are measured simultaneously using a quantitative PCR-based method on DNA extracted from dried blood spots (DBS) or peripheral blood from a newborn (not from cord blood, because this will include maternal blood and could give a false positive result).

TRECs are formed as circular excision products during T-cell receptor gene rearrangement in developing T-lymphocytes in the thymus. Therefore, TRECs are a marker for recently formed T-lymphocytes; absent or strongly reduced levels of

TRECs indicate a low level of newly formed T-lymphocytes. In a systematic review by Van der Spek et al, the sensitivity of TREC-based newborn screening for typical SCID was found to be 100%.

Similarly, KRECs develop during the maturation and somatic recombination of the B-cell receptor locus and their copy number correlates with the number of freshly formed naïve B-cells.

In resource-poor countries, TREC or KREC assays by PCR may not be available, but I will elaborate on ways to detect a primary immunodeficiency early in a healthcare setting where PCR-based methods are not available.

If you have a baby with a high risk of having a severe PID where PCR-based technology for screening is not available, a lymphocyte count and a CD4 or a CD8 count can be performed.

The majority of SCID cases can be identified by a full blood count and differential to determine the absolute number of lymphocytes. T-cells are approximately 70% of lymphocytes in healthy infants, and an absence of T-cells causes the total lymphocyte count of most infants with SCID to be low. However, some forms of SCID are associated with the presence of B-lymphocytes, and maternal T-cells are also sometimes found in the blood of infants with SCID. Therefore, lymphocyte counts would not capture all SCID cases and therefore in these patients the lymphocyte subsets by flow cytometry should be requested. Fortunately, because of mass HIV testing and monitoring, CD4 or CD8 assays should be readily available.

If agammaglobulinaemia is present in the family or this condition is clinically suspected, one should also ask for CD19+B-cells on flow cytometry. It is important to note that normal immunoglobulins do not exclude a diagnosis of PID owing to the presence of maternal antibodies.

Who should be screened with a TREC or a KREC PCR?

- Preferably all newborns, before the administration of live vaccines, dependent on the resources available.
- All babies with a family history of severe primary immunodeficiency affecting B- or T-cells.
- Neonates or older babies or children where the diagnosis of a severe primary immunodeficiency affecting B- or T-cells is suspected.

What specimen is required?

- EDTA blood from the baby and not cord blood (please ensure that there is no dilution of the specimen if it is collected from an intravenous line).
- A dried bloodspot on a Guthrie card obtained after a heel prick.

What does an absent or low TREC or KREC mean?

- SCID
- Severe agammaglobulinaemia
- Syndromes with variably affected cellular immunity that may be severe, including:
  - Complete di George syndrome or partial di George syndrome with low T-cells
  - Ataxia telangiectasia
  - CHARGE syndrome
  - Jacobsen syndrome
  - Trisomy 21
  - RAC2 dominant interfering mutation
  - DOCK8 deficient hyper-IgE syndrome
  - Cartilage hair hypoplasia
- Low T-cells (or B-cells) as a consequence of other conditions, including:
  - Neonatal cardiac surgery
  - Neonatal leukaemia
  - Gastrointestinal malformations
  - Extreme prematurity (resolves to normal with time)
  - Intrauterine growth retardation
  - Maternal immunosuppressive medication during pregnancy, including azathioprine, tacrolimus, mercaptopurine and Rituximab.

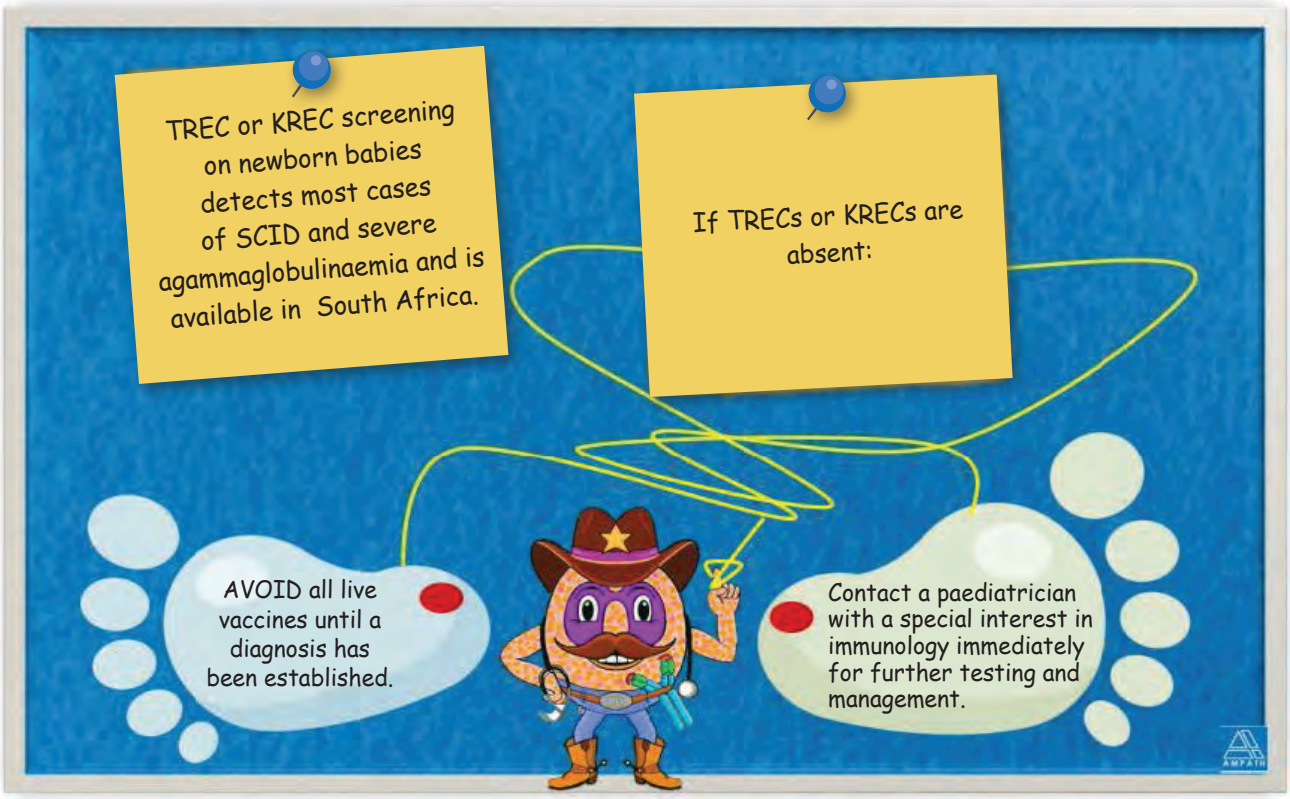
After a positive screening, diagnostic follow-up is required in **all** patients (1) to determine whether the patient suffers from typical SCID, another form of T-cell lymphopaenia, severe agammaglobulinaemia or one of the above-mentioned conditions and (2) to establish a specific genetic diagnosis. Infants with abnormally low numbers of T-cells and/or B-cells should ideally be seen promptly by a paediatrician with a special interest in immunology to determine whether the infant has SCID or severe agammaglobulinaemia.

Once the diagnosis is confirmed, immediate measures can be put in place for all patients, including immunoglobulin replacement therapy and prophylactic antibiotics, the avoidance of live vaccines and protection from exposure to infections – all this while the best type of definitive treatment is planned.

What are the limitations of this test?

- The screening test may be false positive in premature babies under 37 weeks. If the initial screening shows decreased TREC or KREC in a premature baby, it is suggested that the test be repeated once 37 weeks corrected gestation is reached.
- TREC or KREC screening may detect only abnormalities in the cellular and humoral immune system and will not detect abnormalities in the phagocyte, complement, innate or other parts of the immune system.

Dr Spur's take-home message:



Dr Spur's mystery SOLVED:

'An early diagnosis of PID, saves lives'

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