T-Cell *Xtend*®

Technical information

References:

1. Oxford Immunotec. Data on File, 2008.
2. Schmielau J, Finn OJ. Activated granulocytes and granulocyte-derived H_20_2 are the underlying mechanism of suppression of T-cell function in advanced cancer patients. Cancer Research 2001; **61** 4756-4760.
3. Malmberg KJ, Arulampalam V, Ishihara F, *et al*. Inhibition of activated/memory (CD45RO+) T cells by oxidative stress associated with block of NF-κB activation. J Immunol. 2001;**167** 2595-2601.
4. Wang S, Stew S, Cyktor J, Carruthers B, Turner J, Restrepo B. Validation of increased blood storage times with the T-SPOT.*TB* assay with T-Cell *Xtend* reagent in individuals with different tuberculosis risk factors. J. Clin. Microbiol. 2012; doi:**10** 1128.
5. Wang S, Powell D, Nagaraja H, Morris J, Schlesinger L & Turner J. Evaluation of a modified interferon-gamma release assay for the diagnosis of latent tuberculosis infection in adult and paediatric populations that enables delayed processing. Scandinavian Journal of Infectious Diseases. Dec 2010; **42(11-12)** 845-850.
6. Bouwman J, Thijsen S, Bossink. A Improving the timeframe between blood collection and interferon gamma release assay using T-Cell *Xtend*. J Infect 2012 Feb; **64(2)** 197-203.

Contact **Oxford Immunotec for further information**.

Oxford Immunotec Limited, 94C Innovation Drive, Milton Park, Abingdon, Oxfordshire, OX14 4RZ, UK
Tel: +44 (0)1235 442780 Fax: +44 (0)1235 442781 Email: info@oxfordimmunotec.com
www.oxfordimmunotec.com

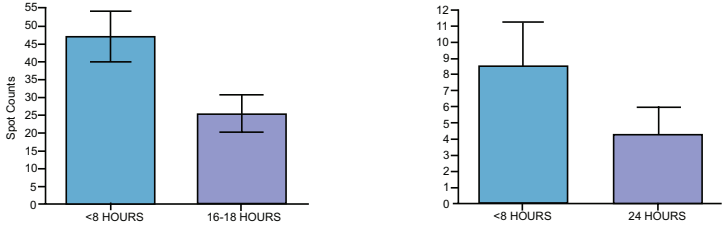
Oxford Immunotec
Harnessing the power of T cell measurement

TI-TTK.610-UK-V3

The T-SPOT®.*TB* assay functions optimally with whole blood stored for up to 8 hours, but what happens if blood is stored for longer?

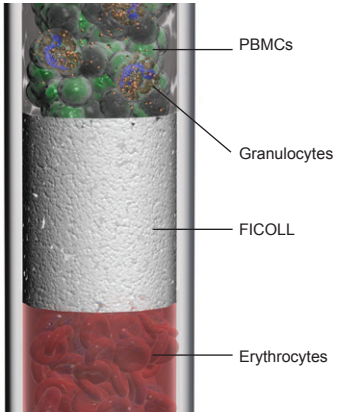
- Performing the T-SPOT.*TB* assay on blood stored for more than 8 hours may result in a decrease in spot counts and an increase in non-specific background in assay wells
- When peripheral blood mononuclear cells (PBMCs) are separated using FICOLL-PAQUE*, the majority of the red blood cells and granulocytes are pelleted on the basis of density
- When blood is stored for longer than 8 hours, granulocytes can become activated causing a decrease in their density[1]. As a result, the granulocytes can contaminate the enriched PBMC layer

Effect of blood storage on T-SPOT.*TB* performance



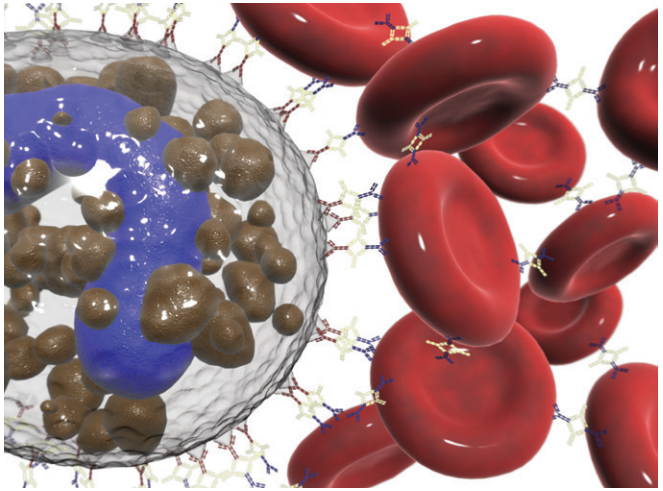
- Granulocytes are known to become activated in some disease states (e.g. cancer)[2,3]

- Activated granulocytes can release pre-stored granules[2], which may cause oxidative stress to lymphocytes in the PBMC layer. This may reduce cell viability and, more specifically, the ability of cells to release interferon-gamma
- The presence of granulocytes may also result in non-specific background, affecting results with stored blood

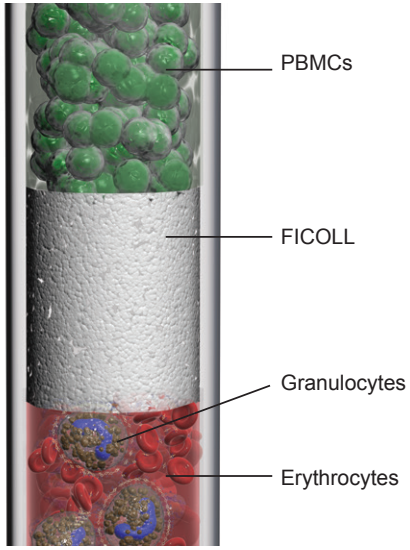


With stored blood granulocytes can appear in the PBMC layer

The T-Cell *Xtend* reagent allows the T-SPOT.*TB* assay to be performed on blood samples stored for up to 32 hours without compromising accuracy



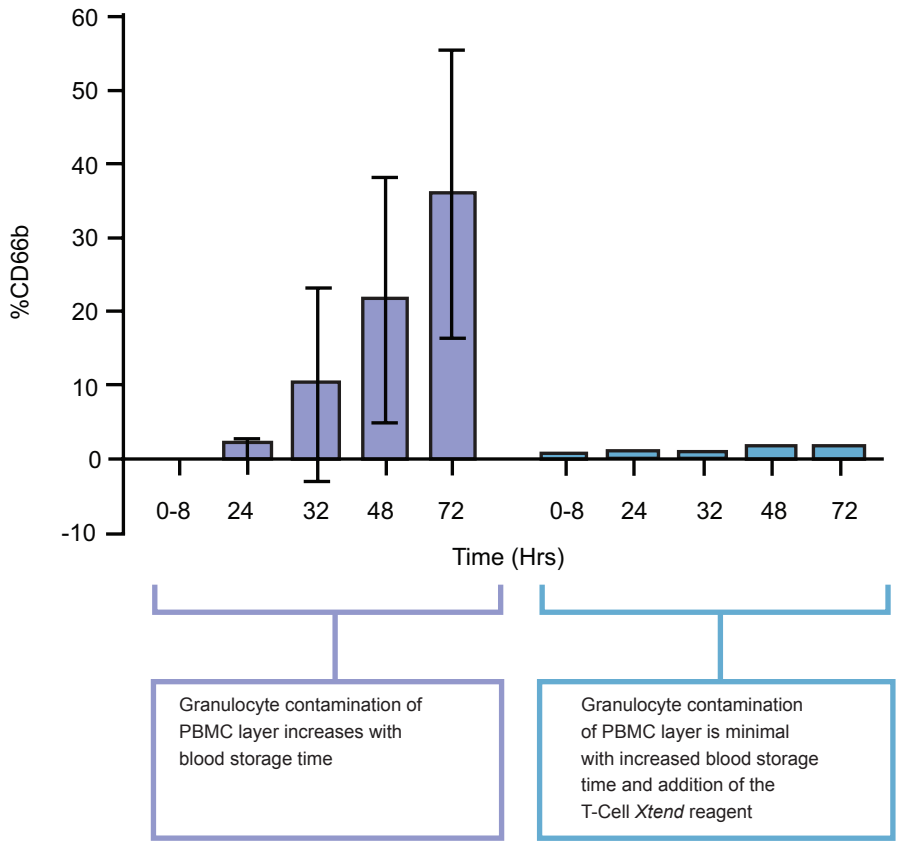
The T-Cell *Xtend* reagent is an antibody complex which recognises CD66b, a specific cell surface marker of granulocytes, and cross-links the granulocytes to red blood cells



This cross-linking increases the density of granulocytes so they pellet when applied to a density gradient

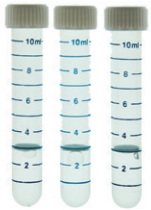
The T-Cell *Xtend* reagent removes unwanted granulocytes from the sample

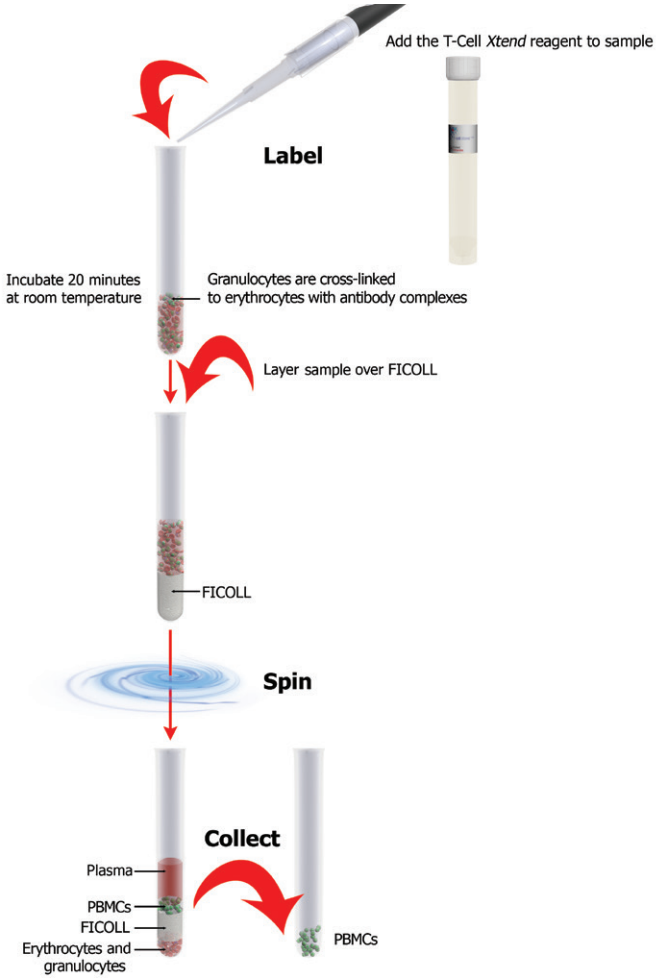
Use of the T-Cell *Xtend* reagent has been demonstrated to reduce the presence of granulocytes in the PBMC layer, thus allowing prolonged storage of blood samples for up to 32 hours before performing the T-SPOT.*TB* assay



The T-Cell *Xtend* reagent is added to blood samples in the laboratory, immediately before commencing the T-SPOT.*TB* assay

T-Cell *Xtend* may also be used with Leucosep tubes to simplify the FICOLL separation procedure





Proceed to ELISPOT Assay

Benefits of using the T-Cell *Xtend* reagent

Equivalent accuracy to standard T-SPOT.*TB* results but with added flexibility[4-6]

- Samples can be collected at a distance from the testing laboratory
- Samples can be collected throughout the day or stored overnight
- Laboratory can perform T-SPOT.*TB* on blood samples received on 2 consecutive days at the same time
- Enables the use of cost-effective, overnight delivery service
- Removal of activated granulocytes may improve performance of the T-SPOT.*TB* assay in certain disease states (e.g. cancer)