

## Monocyte Activation Test for pyrogen testing of biopharmaceutical products

**Commonly used acronym:** MAT

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### Contact person

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### Organisation

**Name of the organisation** Janssen Pharma of JNJ

**Department** Analytical Development

**Country** Belgium

**Geographical Area** Flemish Region

### Partners and collaborations

Janssen Pharma of JNJ

## SCOPE OF THE METHOD

<b>The Method relates to</b>	Human health
<b>The Method is situated in</b>	Regulatory use - Routine production: Regulatory use - GMP process validation
<b>Type of method</b>	In vitro - Ex vivo
<b>Species from which cells/tissues/organs are derived</b>	Human blood
<b>Type of cells/tissues/organs</b>	Peripheral Blood Mononuclear Cells (PBMC)
<b>Specify the type of cells/tissues/organs</b>	Peripheral Blood Mononuclear Cells (PBMC)

## DESCRIPTION

### Method keywords

PBMC

ELISA

endotoxins and non-endotoxin pyrogens

alternative to rabbit pyrogen test

IL-6

European pharmacopoeia

### Scientific area keywords

pyrogenicity  
Biopharmaceuticals

### Method description

Pharmaceutical products intended for parenteral use must be free from pyrogenic (fever-inducing) contamination. Pyrogens comprise endotoxin from Gram-negative bacteria and non-endotoxin pyrogens (NEP) from Gram-positive bacteria, viruses, and fungi. The longstanding compendial test for pyrogens is the Rabbit Pyrogen Test (RPT) but in 2010 the Monocyte Activation Test (MAT) for pyrogenic and pro-inflammatory contaminants was introduced into the European Pharmacopoeia (Ph. Eur.) as a 'non-animal' replacement for the RPT. The developed MAT method was fully validated for GMP purposes according to Ph. Eur. MAT, Quantitative test, Method A to test for pyrogenic and pro-inflammatory substances in therapeutic monoclonal antibodies (mAb). The MAT uses cryo-preserved PBMC with an interleukin-6 (IL-6)-based ELISA readout. The method has been successfully approved by EMA in scope of commercial licensing applications (MAA) for several mAb-based drug products.

### Lab equipment

- CO2 incubator;
- Washer;
- ELISA plate reader;
- SoftMax Pro.

### Method status

Internally validated  
Published in peer reviewed journal

## PROS, CONS & FUTURE POTENTIAL

### Advantages

- *In vitro* test which replaces rabbit-based testing;
- (Semi-) Quantitative.

### Challenges

- Extensive validation required;
- Requires well-characterized PBMCs.

### Modifications

In case the drug product would interfere with an IL-6-based readout, other cytokines such as IL-1 beta may need to be explored and validated as alternative.

## REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

### Associated documents

[Daniels et al - ALTEX 2022 - MAT for therapeutic monoclonal antibodies.pdf](#)  
[Daniels et al - Curr Res Tox 2024 - MAT Fit for purpose testing.pdf](#)

### Links

[Validation of the monocyte activation test with three therapeutic monoclonal an...](#)  
[Fit for purpose testing and independent GMP validation of the monocyte activati...](#)

### Other remarks

Partners for this method: Sanquin (m.molenaar@sanquin.nl) & Janssen Pharma of JNJ (rdanie22@its.jnj.com)

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