

# Application of Luminex Multiplex System in IO Research



WuXi AppTec Research Service Division, Oncology & Immunology Unit



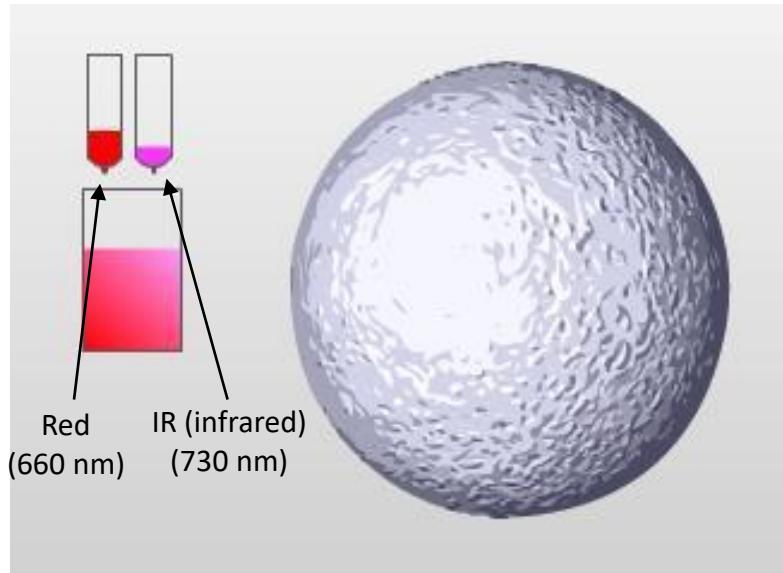
2019.11

# Multiplex immunoassays: Luminex Multiplex System

## Detection principle

### 1. Microsphere color coding

- Red and infrared fluorescent dyes are coated with microspheres
- Each fluorescence has 10 concentrations
- $10 \times 10 = 100$  different proportions of fluorescence

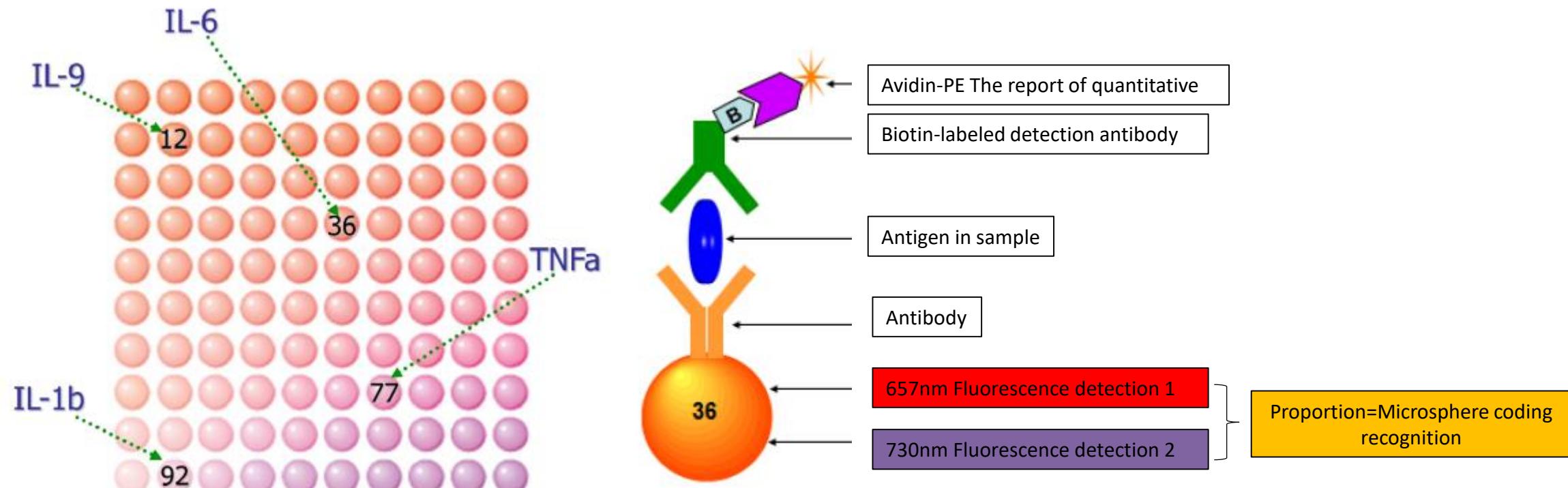


# Multiplex immunoassays: Luminex Multiplex System

## Detection principle

### 2. Antibodies are conjugated to microspheres

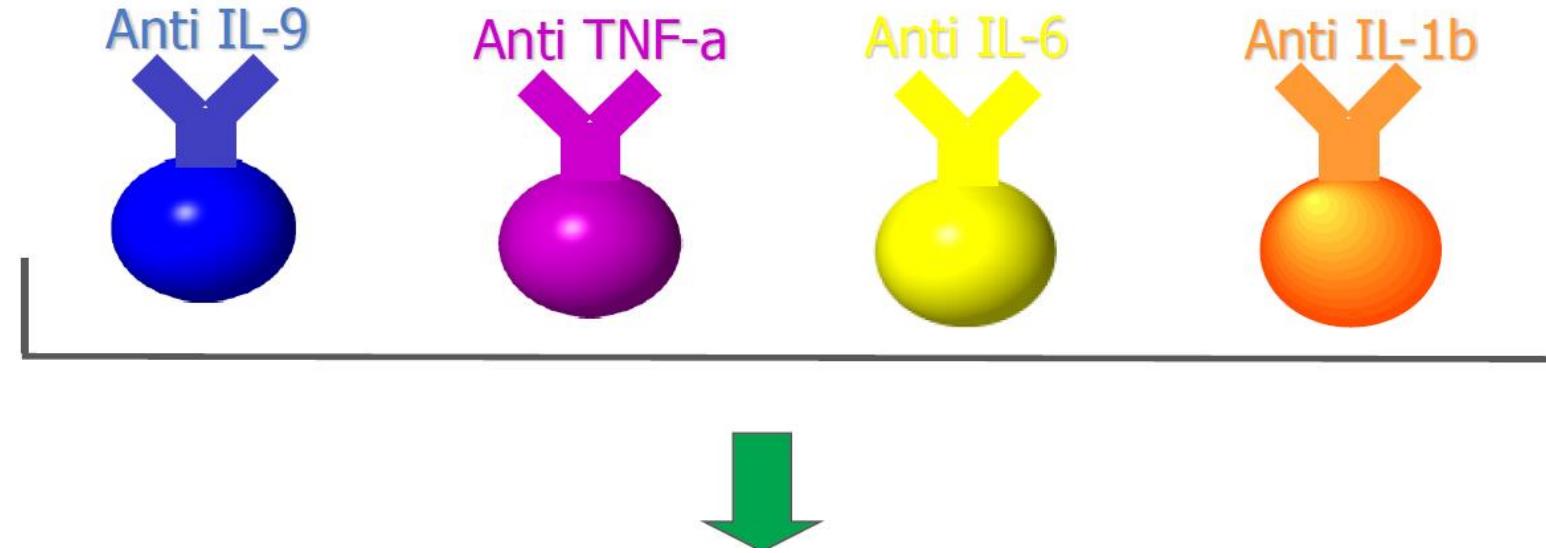
- Different specific antibodies were conjugated to different colored microspheres
- The spheres of each color point to a specific analyte
- The coupled microspheres are used for multiple detection after mixing



# Multiplex immunoassays: Luminex Multiplex System

Detection principle

## 3. Perform multiple reactions

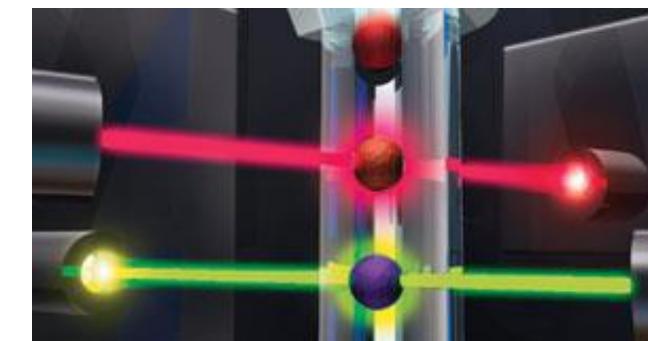
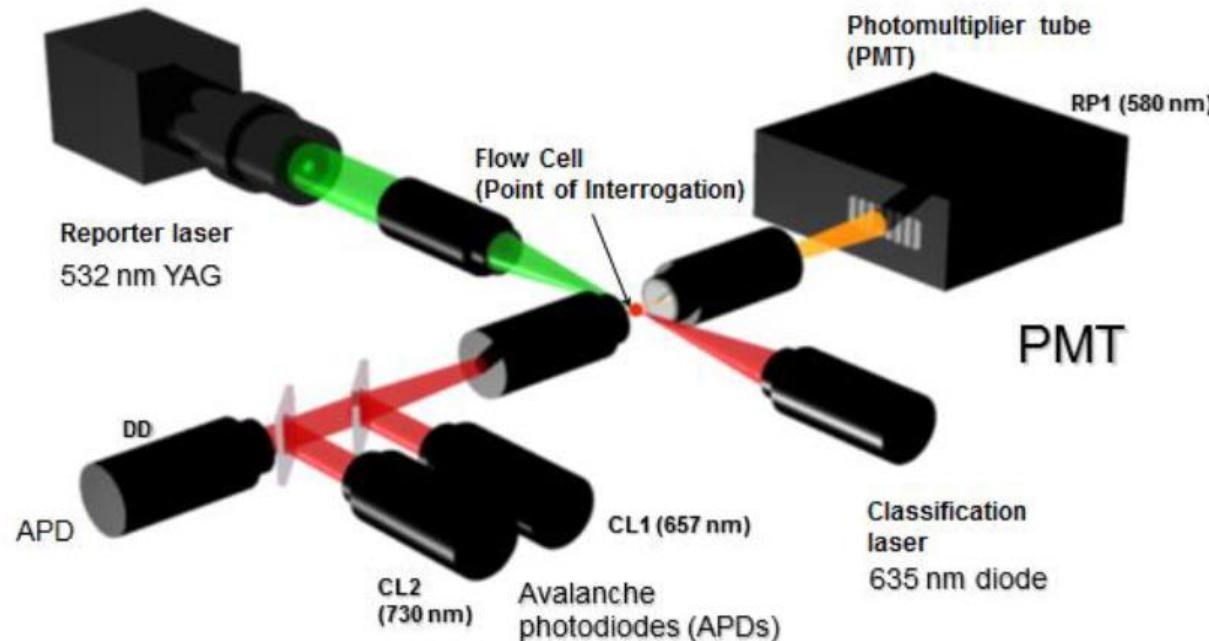


- Different tests were mixed simultaneously in the same reaction hole
- The instrument classified and quantified microspheres for each test
- Also report all test results

# Multiplex immunoassays: Luminex Multiplex System

## Detection principle

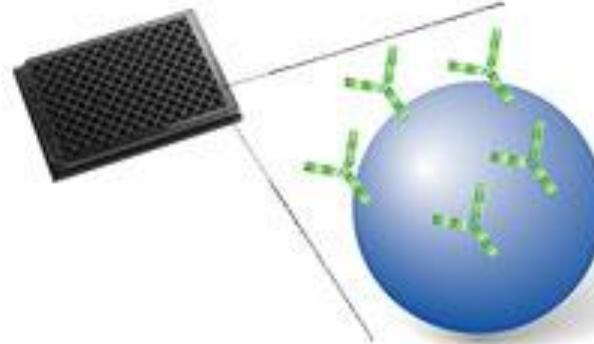
- The stained microspheres pass through the liquid flow path step by step
- The red light source (635nm classification laser) in the instrument measures the dye in the microsphere, identifies the microsphere coding and analyzes the species class
- The green light (532nm report laser) measured the microsphere report molecule, that is, the analyte concentration



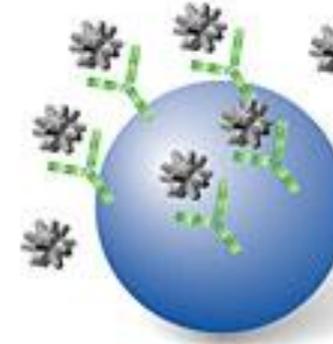
# Multiplex immunoassays: Luminex Multiplex System

## Bio-Plex Cytokine Assay Workflow

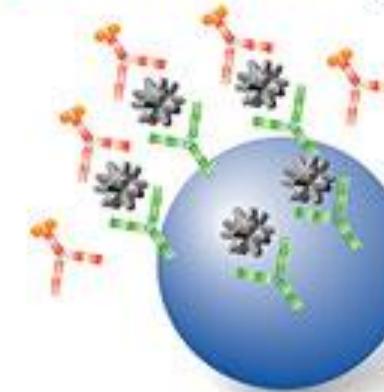
Step One  
Dispense capture beads



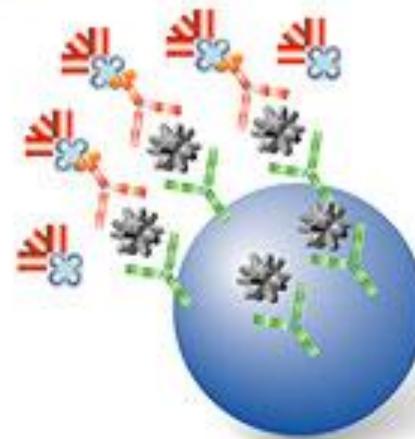
Step Two  
Add samples



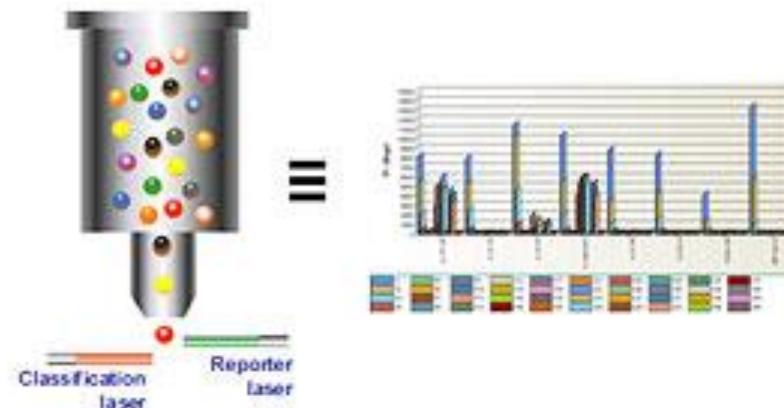
Step Three  
Add detection antibody



Step Four  
Add reporter dye

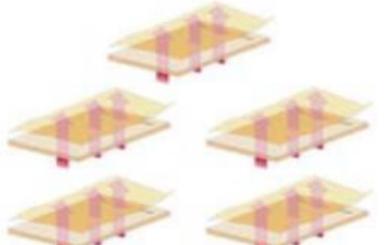
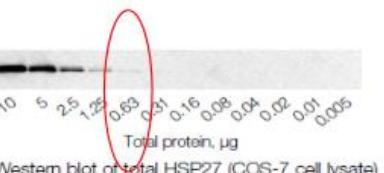
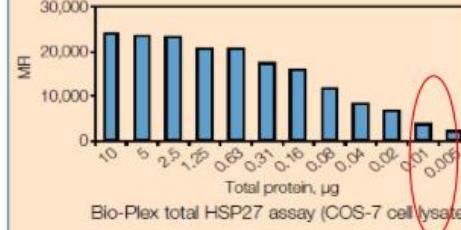


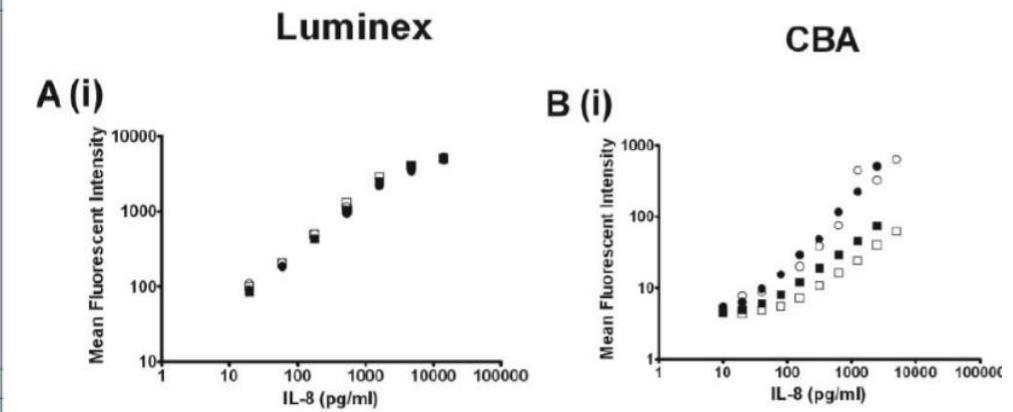
Step Five  
Fluorescent sorting and data reduction



# Multiplex immunoassays: Luminex Multiplex System

## Advantages:

|                                  | Western Blot  | xMAP Technology   |
|----------------------------------|---|---|
| Number of phosphoproteins        | 5   | 5   |
| Number of samples                | 12  | 12  |
| Total data points                | 60  | 60  |
| Number of gels or 96-well plates | 5   | 1   |
|                                  |    |    |
| Data points per gel or plate     | 12  | 60  |
| Total time required              | ~8 hr + one overnight incubation  | 3 hr + one overnight incubation   |
| Sample volume                    | 125 µl  | 25 µl   |
| Dynamic range                    | Limited   | 50-900 µg/ml total protein  |
| Sensitivity*                     | <br>Western blot of total HSP27 (COS-7 cell lysate) | <br>Bio-Plex total HSP27 assay (COS-7 cell lysate) |



Luminex kits were found to be highly reproducible and reliable; CBA kits were not due to aberrant standards.

# Multiplex immunoassays: Luminex Multiplex System

## Sample types include:

- Serum
- Plasma
- Cell/Tissue Lysate
- Cerebrospinal Fluid
- Synovial Fluid
- Blister Fluid
- Tissue Culture Supernatants
- Fat Interstitial Fluid
- Bronchoalveolar Lavage Fluid
- Nasal Lavage Fluid
- Peritoneal Fluid



# Multiplex immunoassays: Luminex Multiplex System

## Applications include:

### Human

- [Acute Phase Assays](#)
- [Apolipoprotein Assays](#)
- [Apoptosis Assays](#)
- [Chemokine Assays](#)
- [Cytokine Chemokine and Growth Factors](#)
- [Diabetes Assays](#)
- [Inflammation Assays](#)
- [Isotyping Assays](#)
- [Kidney Toxicity Assays](#)
- [Th17 Cytokine Assays](#)

### Mouse

- [Chemokine Assay](#)
- [Cytokine, Chemokine, and Growth Factor Assays](#)
- [Diabetes Assay](#)

### Rat

- [Cytokine, Chemokine, and growth factor Assays](#)
- [Diabetes Assay](#)
- [Kidney Toxicity Assay](#)

### Other

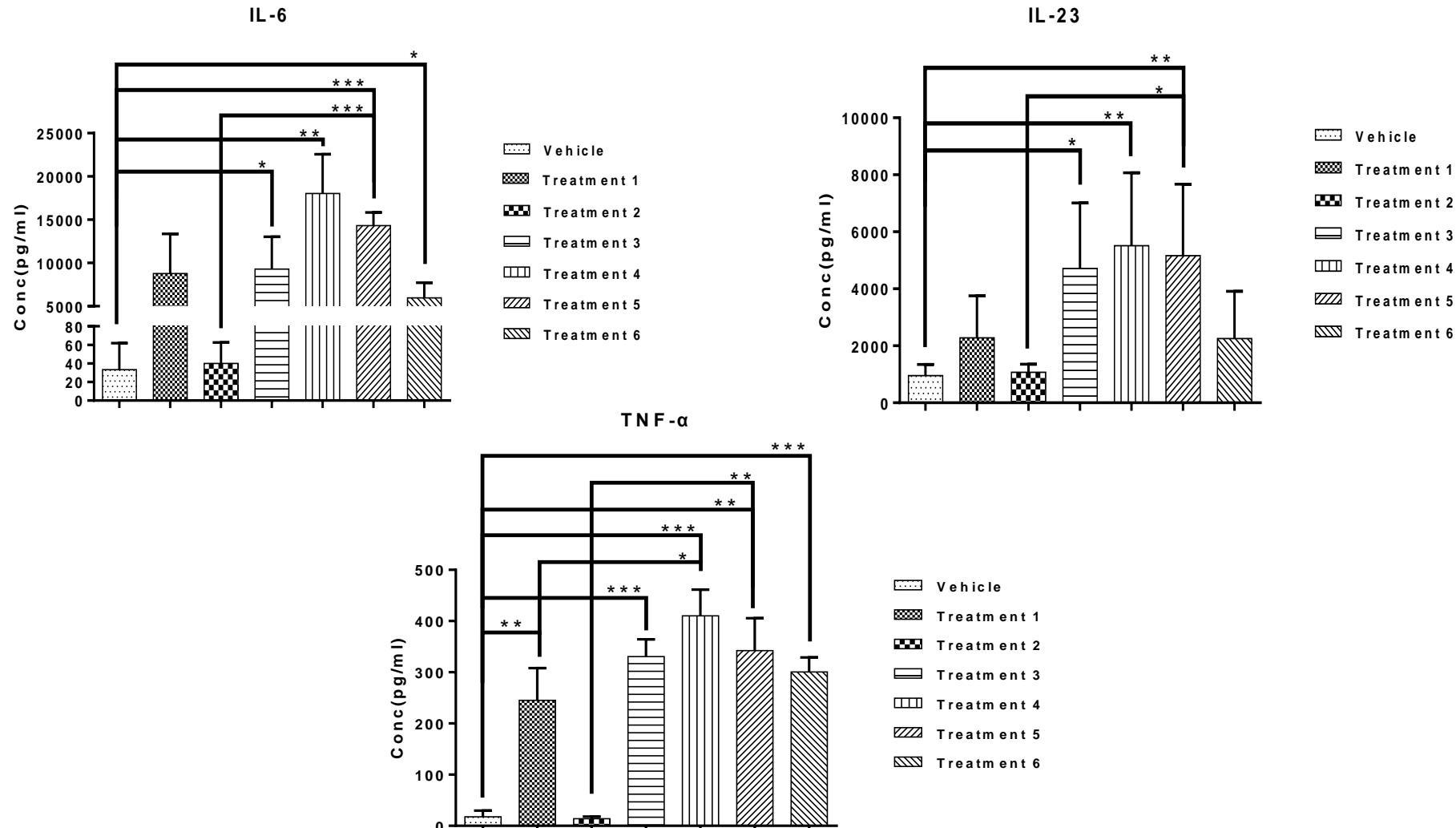
- [Canine Kidney Toxicity Assay](#)
- [Multi species TGF Beta Assays](#)
- [Multi species Cell Signaling Assays](#)

**Bio-Plex Multiplex immunoassays** use **Luminex magnetic beads** for the quantification of over 450 biologically relevant targets: **assays** for inflammation, disease, cancer, cell signaling and growth, apoptosis, toxicity, and more.

# Multiplex immunoassays: Luminex Multiplex System

## Showcase Data

Using Luminex to examine the expression levels of cytokines in C57BL/6 plasma after different treatments.





# OUR COMMITMENT

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For questions and requests, please email to [info\\_onco@wuxiapptec.com](mailto:info_onco@wuxiapptec.com)



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