GE Healthcare

# Amersham Interleukin-10 [(m)IL-10], Mouse Biotrak ELISA System

## Product booklet

Code: RPN2722 (96 wells)

# Page finder

# 1. Legal



http://www.gehealthcare.com/lifesciences

GE Healthcare UK Limited.
Amersham Place, Little Chalfont,
Buckinghamshire, HP7 9NA
UK

# 2. Handling

## 2.1. Safety warnings and precautions

**Warning: *For research use only.***

Not recommended or intended for diagnosis of disease in humans or animals. Do not use internally or externally in humans or animals.

All chemicals should be considered as potentially hazardous. We therefore recommend that this product and components are handled only by those persons who have been trained in laboratory techniques and that it is used in accordance with the principles of good laboratory practice. Wear suitable protective clothing, such as laboratory overalls, safety glasses and gloves. Care should be taken to avoid contact with skin or eyes. In the case of contact with skin or eyes, wash immediately with water. See material safety data sheet(s) and/or safety statement(s) for specific advice.

## 2.2. Storage

Store at 2-8ºC

## 2.3. Expiry

The expiry date is stated on the package and will normally be at least 4 weeks from the date of despatch.

# 3. Description

The Biotrak™ mouse interleukin-10 ELISA system from GE Healthcare provides a simple, specific and precise quantitative determination of (m)IL-10 in cell culture supernatants and serum.

The assay is based on a solid phase ELISA, which utilizes an antibody for (m)IL-10 bound to the wells of a microplate together with a biotinylated antibody to (m)IL-10 and streptavidin conjugated to horseradish peroxidase (figure 1). Although GE Healthcare IL-10 immunoassay contains recombinant IL-10 and antibodies raised against recombinant (m)IL-10 it has been shown to quantitate accurately both natural (m)IL-10 and recombinant (m)IL-10.

(m)IL-10 can be measured in the range 37-3000 pg/ml (1.85-150 pg/well) in less than 6 hours using the protocol provided with the kit. Each pack contains sufficient material for 96 wells. If one standard curve is constructed, 42 unknowns can be measured in duplicate.

- High sensitivity <12 pg/ml (0.6 pg/well)
- Same day protocol
- Pre-coated plate
- Specific for (m)IL-10

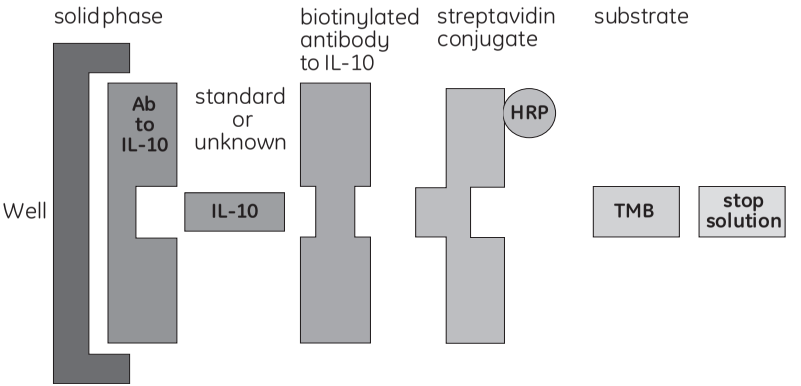



**Figure 1.** Mouse interleukin-10 ELISA assay design

# 4. Critical parameters

**1**) Allow samples and all reagents to reach room temperature prior to performing the assays. Do not use water baths to thaw samples or reagents.

**2**) Mix samples and all reagents thoroughly before use.

**3**) Avoid excessive foaming of reagents. Also avoid exposure of reagents to excessive heat or light during storage and incubation.

**4**) Avoid handling the tops of the wells both before and after filling.

**5**) Standards and samples should be assayed in duplicate.

**6**) Run a separate standard curve for each assay.

**7**) The total dispensing time for each plate should not exceed 20 minutes.

**8**) Use only coated wells from the same reagent batch for each assay. Also do not mix reagents from different kit lots.

**9**) For sample, biotinylated antibody and conjugate incubations a humidified incubator may be used to prevent evaporation loss due to incomplete plate sealing.

**10**) It is important that the wells are washed thoroughly and uniformly. If using an automatic washer check operation of head before starting. If washing by hand ensure that all wells are completely filled at each wash.

**11**) A small amount of precipitate may be present in some vials. It will not affect assay performance and should be ignored.

**12**) Use a new adhesive plate cover for each incubation step in the ELISA.

**13**) New pipette tips should be used for each standard and sample. GE Healthcare recommends the use of multiple tips or multi-shot dispensing pipettes for the addition of other reagents to the wells.

# 5. Additional materials and equipment required

The following materials and equipment are required:

- Pipettes or pipetting equipment with disposable tips (50 µl, 100 µl, 200 µl, 400 µl and 1.0 ml)*
- Equipment capable of pipetting 30 µl
- 15 ml plastic tube
- Equipment capable of pipetting 8 ml
- Disposable polypropylene test tubes - do not use polystyrene, polycarbonate or glass
- Measuring cylinder 2 l
- Distilled or deionized water
- Plate reader capable of reading at 450 nm
- A centrifuge for preparing streptavidin-HRP solution
- Wash bottle or automatic plate washer

**Optional equipment**
Assays may be performed with commercially available microplate washers to aid convenience and assay throughput.

* GE Healthcare supplies a range of pipettes and disposable tips (see related products).

# 6. Specimen collection and sample preparation

**Cell culture supernatants**
Centrifuge to remove any particulate material.

**Serum**
Serum samples should be allowed to clot at room temperature. Immediately after clotting, spin down. Specimens should be clear and non-haemolyzed whenever possible.

**Storage of test samples**
Serum and culture supernatant samples that are to be assayed within 24 hours should be stored at 2-8°C. Specimens to be stored for longer periods of time should be frozen at -70°C to avoid loss of biologically active cytokine. Avoid freezing and thawing samples more than once. Test samples should be assayed in duplicate each time the ELISA is performed, 50 µl of sample per well is required in this assay.

**Dilution of test samples**
If it is suspected that the (m)IL-10 concentration of a sample exceeds the highest point of the standard curve, prepare one or more five-fold dilutions of the test sample. Mix thoroughly between dilutions and before assaying.

# 7. ELISA procedure

## 7.1. Reagent preparation

**Wash buffer concentrate**

Any precipitate formed during storage will redissolve upon dilution. Dilute 30-fold with distilled or deionized water to prepare 1500 ml of wash buffer. Store at 2-8°C until the expiry date of the kit. Do not use wash buffer if it becomes visibly contaminated on storage.

**Streptavidin-HRP solution**

Prepare the exact amount of streptavidin-HRP solution no more than 15 minutes prior to use. Do not prepare more conjugate than required. Do not store diluted conjugate.

The streptavidin-HRP concentrate may require spinning down to force the contents to the bottom of the vial. Add 30 µl of streptavidin-HRP concentrate per 12 ml of streptavidin-HRP dilution buffer in a plastic 15 ml tube and mix gently. If running partial plates use 2.5 µl of streptavidin-HRP concentrate and 1 ml of streptavidin-HRP buffer per strip being run.

## 7.2. Preparation of working standards

**1**) Two vials of lyophilized standard are provided with this kit Reconstitute and use one vial per partial plate.

**2**) Prepare standards shortly before use. Use within one hour of reconstitution. **Do not store reconstituted standards.**

**3**) When **running culture supernatant samples,** reconstitute standard in distilled or deionized water. Reconstitution volume is stated on the standard vial label. The standard will take approximately 1 minute to dissolve. Mix by gently inverting the vial. Use your culture medium to prepare the dilutions of the standard curve, go to step 5) below for further instructions. If running a partial plate, refer to step 1) above.

**4**) If **running serum samples**, reconstitute standard with distilled or deionized water. Reconstitution volume is stated on the standard vial label. The standard will take approximately 1 minute to dissolve. Mix by gently inverting the vial. Use the assay buffer provided to prepare the dilutions of the standard curve. If running a partial plate, refer to step 1) above.

**5**) Label 6 tubes, one for each standard curve point: 3000 pg/ml, 1000 pg/ml, 333 pg/ml, 111 pg/ml, 37.6 pg/ml and 0 pg/ml. Then prepare 1:3 serial dilutions for the standard curve as follows:

**6**) Pipette 400 µl of appropriate diluent (see steps 3) and 4) above) into each tube.

**7**) Pipette 200 µl of the reconstituted standard into the first tube, 3000 pg/ml and mix.

**8**) Pipette 200 µl of this dilution into the second tube labelled 1000 pg/ml and mix.

**9**) Repeat serial dilutions three more times. These concentrations, 3000 pg/ml, 1000 pg/ml, 333 pg/ml, 111 pg/ml, 37 pg/ml and 0 pg/ml are your standard curve.

## 7.3. Assay protocol

This ELISA provides the flexibility to run two partial plates on separate occasions. Decide the number of strips you wish to run, leaving the strips to be used in the frame. Remove the unnecessary strips and store them in the foil pouch with the desiccant provided at 2-8°C, making sure the foil pouch is sealed tightly. After running the assay, retain the plate frame for the second partial plate.

When adding the TMB substrate reagent, pour out from the bottle only the amount needed to run the first half plate. Do not combine left over substrate with that reserved for the second half of the plate.

Care must be taken to ensure that the remaining TMB substrate reagent is not contaminated. **If the substrate reagent is bright blue prior to use, it has been contaminated. DO NOT USE.**

**1**) Prepare assay reagents and working standards as described in the previous sections.

**2**) Set up the microplate with sufficient wells to enable the running of all standards and samples as required (see figure 2).

**3**) Remove excess microplate strips from the frame and store in the resealable foil bag.

**4**) Add 50 µl of assay buffer reagent to each well that is to be used.

**5**) Add 50 µl of standard or sample per well. Cover with adhesive strip provided and incubate for 3 hours at room temperature (20-25°C).

**6**) Remove adhesive strip and aspirate or decant each well and wash, repeating the process twice for a total of three washes. Wash vigorously by filling each well with wash buffer using a wash bottle, pipette or manifold dispenser. Complete removal of liquid at each step is essential for good performance. After the last wash, remove any remaining wash buffer by inverting the plate and blotting it against clean paper towelling.

**7**) Add 50 µl of pre-mixed biotinylated antibody conjugate. Cover with a new adhesive strip and incubate for 1 hour at room temperature (20-25°C).

**8**) Repeat the aspiration/wash step as in step 6.

**9**) Add 100 µl of the pre-diluted streptavidin-HRP conjugate. Cover with a new adhesive strip and incubate for 30 minutes at room temperature (20-25°C).

**10**) Repeat the aspiration/wash step as in step 6.

**11**) Add 100 µl of TMB substrate solution into each well and incubate for 30 minutes at room temperature (20-25°C). If the substrate reagent is bright blue prior to use, do not use.
**THE PLATE SHOULD BE DEVELOPED IN THE DARK.**
Do not cover the plate with aluminium foil or an adhesive strip.

**12**) Add 100 µl of stop solution to each well.

**13**) Determine the optical density of each well within 30 minutes, using a spectrophotometer set to 450 nm.

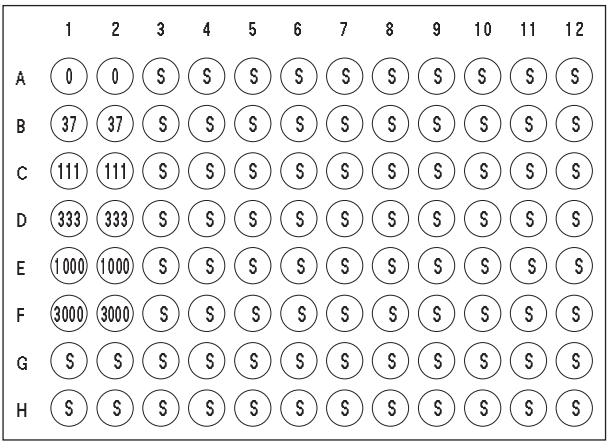
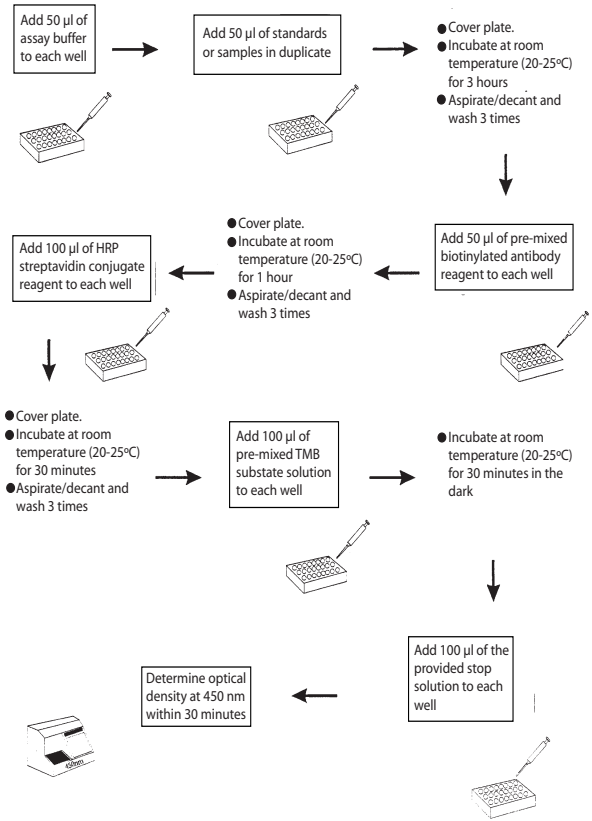



**Figure 2.** Recommended positioning of standards (0-3000 pg/ml) and samples (S)

**Table 1.** Assay protocol (all volumes are in microlitres)

| | Zero standard | Standards | Samples |
|---|---|---|---|
| Assay buffer | 50 | 50 | 50 |
| Standard | – | 50 | – |
| Standard diluent or cell culture media | 50 | – | – |
| Sample | – | – | 50 |
| Cover plate, incubate at room temperature for 3 hours. | | | |
| Aspirate/decant and vigorously wash all wells three times with wash buffer. | | | |
| Pre-mixed biotinylated antibody | 50 | 50 | 50 |
| Cover plate, incubate at room temperature for 1 hour. | | | |
| Aspirate/decant and vigorously wash all wells three times with wash buffer. | | | |
| Streptavidin-HRP conjugate | 100 | 100 | 100 |
| Cover plate, incubate for 30 minutes at room temperature | | | |
| Aspirate/decant and vigorously wash all wells three times with wash buffer | | | |
| Substrate | 100 | 100 | 100 |
| Incubate at room temperature for 30 minutes in the dark | | | |
| Stop solution | 100 | 100 | 100 |
| Determine optical density at 450 nm within 30 minutes. | | | |

## Summary of assay protocol



# 8. Data processing

## 8.1. Calculation of results

Typical assay data is shown in table 2.

Average the duplicate readings for each standard, control and sample and subtract the zero standard optical density.

Plot the optical density for the standards versus the concentration of the standards and draw the best curve. The data can be linearized using a log/log plot and regression analysis can be applied to the log transformation.

Figure 3 shows such a plot of the data from table 2. The standard curve is provided for illustration only. A standard curve should be generated for each set of samples to be assayed. This allows for the measurement of 42 unknowns in duplicate.

## 8.2. Typical assay data

The following data (table 2) were obtained for a standard curve using the protocol provided.

**Table 2.** Typical assay data

| Tube | Optical density | Zero standard subtracted |
|---|---|---|
| Zero standard | 0.065 | – |
| 37 pg/ml standard | 0.100 | 0.035 |
| 111 pg/ml standard | 0.142 | 0.077 |
| 333 pg/ml standard | 0.274 | 0.209 |
| 1000 pg/ml standard | 0.633 | 0.568 |
| 3000 pg/ml standard | 2.011 | 0.946 |

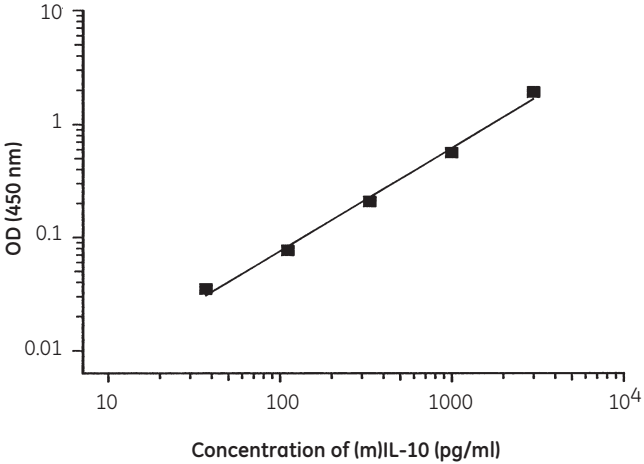

10
1
0.1
0.01
OD (450 nm)
10
100
1000
10^4
Concentration of (m)IL-10 (pg/ml)

**Figure 3.**

# 9. Additional Information

## 9.1. Specificity

This ELISA is specific for the measurement of natural and recombinant mouse IL-10. It does not crossreact with mouse; IL-1α, IL-1β, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IFNγ, TNFα, GM-CSF or human IL-10.

## 9.2. Reproducibility

**Within-assay precision**

The within-assay coefficient of variation of the ELISA has been determined to be <10%.

**Between-assay precision**

The between-assay coefficient of variation of the ELISA has been determined to be <10%.

## 9.3. Sensitivity

The minimum detectable dose of (m)IL-10 was determined to be <12 pg/ml (0.6 pg/well), by adding two standard deviations to the optical density of zero and calculating the corresponding concentration from the standard curve.

## 9.4. Normal levels

The average level of mouse IL-10 in 8 serum samples from apparently healthy Balb/c mice was 18 pg/ml with a range of 0-53 pg/ml.

## 9.5. LPS-stimulated (m)IL-10 production

To induce mouse IL-10 production Balb/c mice were injected with LPS, 50 µg/mouse. Serum samples were harvested by heart puncture over a 3 hour time course and were then run in the mouse IL-10 ELISA.

Additionally, splenocytes harvested from Balb/c mice were stimulated with 20 µg/ml LPS over a three day time course. Supernatants were collected at various time points and run in the mouse IL-10 ELISA. The results of these two experiments are represented in the table below:

| ***In vivo* LPs-stimulated serum Samples** | | **LPs-stimulated splenocyte supernatants** | |
|---|---|---|---|
| **Time, post-injection** | **Mouse IL-10** | **Time, after stimulation** | **Mouse IL-10** |
| 0 minutes | 30 pg/ml | Day 1 | 79 pg/ml |
| 60 minutes | 1170 pg/ml | Day 2 | 277 pg/ml |
| 90 minutes | 4225 pg/ml | Day 3 | 852 pg/ml |
| 120 minutes | 1623 pg/ml | | |
| 150 minutes | 1064 pg/ml | Unstimulated control | |
| 180 minutes | 497 pg/ml | Day 3 | 0 pg/ml |

**Note:** Strain to strain differences in the base levels of mouse IL-10 and non-specific activity (up to 45 pg/ml) was observed when testing commercially available normal serum samples in this assay. We recommend running appropriate control mouse samples in every experiment.

## 9.6. Linearity of dilution

Linearity of dilution was determined by serially diluting ten different positive samples. Dilutions were run in the ELISA and 'found' doses are plotted against 'expected' doses. The slope of the regression line was close to 1 indicating that the samples dilute linearly.

## 9.7. Recovery

Recovery in this ELISA was determined by spiking three different levels of recombinant mouse IL-10 into five serum samples from healthy Balb/c mice and three serum pools from various mouse strains. Recoveries were as follows:

| Spike level | Mean recovery |
|---|---|
| 100 pg/ml | 116% |
| 500 pg/ml | 102% |
| 1500 pg/ml | 101% |

# 10. Troubleshooting guide

**1. Low optical densities**

**1.1**) Check the colour of the substrate in the wells. If it is blue add acid to terminate the reaction and develop the correct yellow colour prior to reading.

**1.2**) Check reader wavelength.

**1.3**) Ensure all reagents have been equilibrated to room temperature before use.

**1.4**) Check reagents have been correctly reconstituted.

**1.5**) Check reagents have been stored under the recommended conditions.

**1.6**) Check incubation times and temperatures.

**1.7**) Ensure that the plate is read within 30 minutes of adding the stop reagent.

**2. High optical densities**

**2.1**) Check point 1.4.

**2.2**) Check point 1.6.

**3. Poor replication**

**3.1**) Ensure automatic washers are working correctly, or that each well is completely filled and emptied at every wash step when hand washing.

**3.2**) Check pipette calibration.

**3.3**) Ensure troughs used with multichannel pipettes are dedicated to individual components.

**3.4**) If splashing occurs when using multishot pipettes, lubricate the pipette barrel.

**3.5**) Ensure that no cross contamination occurs by using a fresh adhesive plate cover at each incubation stage.

**3.6**) Ensure that plates have been carefully placed into the plate reader to avoid splashing.

4. **Standard replication is good, but the standards do not fit the curve**
   Check standard dilution procedure.

5. **High non-specific binding**
   Check point 3.1.

6. **Brown 'precipitate' in wells**
   Under conditions where there are very high levels of enzyme activity in the wells, a brown precipitate will appear on addition of acid. This will initially yield a high optical density but this will decay rapidly. This is indicative of a dilution error with the streptavidin-HRP conjugate.

# 11. Background

Mouse interleukin-10 ((m)IL-10) is a 160 amino acid (18 kDa) glycoprotein that inhibits cytokine synthesis by the TH1 subpopulation of T cells, the effectors of cell-mediated immunity. IL-10 is expressed by activated TH2 cells, mast cells, macrophages and monocytes and exerts profound inhibitory effects on the monocyte/macrophage lineage, including downregulation of class II MHC and suppression of IL-1$\alpha$ and $\beta$, IL-6, IL-8, GM-CSF and TNF$\alpha$ production. The IL-10 receptor belongs to the class II CKR family and is expressed in B cells, thymocytes, mast cells and macrophages.

IL-10 synergizes with IL-2 and IL-4 to promote the proliferation of thymocytes and acts in conjunction with IL-3 and IL-4 to enhance mast cell survival. IL-10 is homologous (70%) to BCRF1, a protein encoded by an open reading frame in the EBV genome. The BCRF1 protein inhibits cytokine synthesis, but does not display other biological effects characteristic of IL-10.

Exploration of IL-10's role *in vivo* has revealed its importance in regulating inflammatory responses. L-10 'knockout' mice spontaneously develop enterocolitis, or bowel inflammation. In a skin irritant model, these mice exhibit profound tissue damage, which can be blocked by anti-TNF$\alpha$ antibodies, confirming IL-10's anti-inflammatory properties *in vivo.* Spleen cells from IL-10 knockout mice produce large amounts of IFN$\gamma$ and IL-6 following antigenic stimulation; these mice also exhibit increased contact hypersensitivity responses, a TH1-mediated function. The suppressive effects of IL-10 suggest that this molecule may have utility as a general suppressor of immune function. Application areas include infectious disease, transplantation, induction of tolerance and possibly cancer;

# 12. References

**1.** FIORENTINO, D.F. *et al., J. Immunology,* **141**(11), p.3815, 1991.

**2.** MOSMANN, T.R. and MOORE, K.W., *Immuno. Today,* **12**(3), p.A49, 1991.

**3.** ZLOTNIK, A. and MOORE, K.W., *Cytokine*, **3**(5), p.366, 1991.

**4.** MOORE, K.W. *et al., Science,* **248**, p.1230, 1990.

**5.** VAN SNICK, J. *et al., Proc. Natl. Acad. Sci.*, **83**, pp.9679-9683, 1986.

**6.** DE VRIES, J.E., *Ann. Med.,* **27**(5), pp.537-541, 1995.

**7.** HO, A.S. and MOORE, K.W., *Ther. Immunol.,* **1**(3), pp.173-85, 1994.

# 13. Related products

**Biotrak range of human cytokine ELISA systems**

| | |
|---|---|
| Interleukin-1α [(h)IL-1α] | RPN 2750 |
| Interleukin-1β [(h)IL-1β] | RPN 2751 |
| Interleukin-2 [(h)IL-2] | RPN 2752 |
| Soluble interleukin-2 receptor [(h)sIL-2R] | RPN 2767 |
| Interleukin-4 [(h)IL-4] | RPN 2753 |
| Interleukin-5 [(h)IL-5] | RPN 2761 |
| Interleukin-6 [(h)IL-6] | RPN 2754 |
| Interleukin-8 [(h)IL-8] | RPN 2764 |
| Interleukin-10 [(h)IL-10] | RPN 2755 |
| Interleukin-12 [(h)IL-12], (p40 and p70) | RPN 2765 |
| Interleukin-13 [(h)IL-13] | RPN 2766 |
| Granulocyte-macrophage colony stimulating factor [(h)GM-CSF] | RPN 2756 |
| Interferon-gamma [(h)IFNγ] | RPN 2757 |
| Tumour necrosis factor-alpha [(h)TNFα] | RPN 2758 |
| Interferon-alpha [(h)]IFNα] | RPN 2759 |
| Granulocyte colony stimulating factor [(h)G-CSF] | RPN 2762 |
| Transforming growth factor-$\beta_1$ [(h)TGF$\beta_1$ ] | RPN 2763 |

**Biotrak range of high sensitivity human cytokine ELISA systems**

| | |
|---|---|
| Interleukin-1α [(h)IL-1α] | RPN 2780 |
| Interleukin-1β [(h)IL-1β] | RPN 2781 |
| Interleukin-4 [(h)IL-4] | RPN 2783 |

| | |
|---|---|
| Interleukin-6 [(h)IL-6] | RPN 2784 |
| Interleukin-10 [(h)IL-10] | RPN 2785 |
| Granulocyte-macrophage colony stimulating factor [(h)GM-CSF] | RPN 2786 |
| Interferon-gamma [(h)IFNγ] | RPN 2787 |
| Tumour necrosis factor-alpha [(h)TNFα] | RPN 2788 |
| Interferon-alpha [(h)IFNα] | RPN 2789 |

**Biotrak range of mouse cytokine ELISA systems**

| | |
|---|---|
| Interleukin-1α [(m)IL-1α] | RPN 2719 |
| Interleukin-1β [(m)IL-1β] | RPN 2720 |
| Interleukin-2 [(m)IL-2] | RPN 2710 |
| Interleukin-3 [(m)IL-3] | RPN 2711 |
| Interleukin-4 [(m)IL-4] | RPN 2712 |
| Interleukin-5 [(m)IL-5] | RPN 2713 |
| Interleukin-6 [(m)IL-6] | RPN 2714 |
| Granulocyte-macrophage colony stimulating factor [(m)GM-CSF] | RPN 2716 |
| Interferon-gamma [(m)IFNγ] | RPN 2717 |
| Tumour necrosis factor-alpha [(m)TNFα] | RPN 2718 |

**Biotrak range of cell adhesion molecule assays**

| | |
|---|---|
| h-sICAM-1 ELISA | RPN 247 |
| h-sELAM-1 ELISA | RPN 248 |
| h-sVCAM-1 ELISA | RPN 249 |
| Mouse sICAM-1 ELISA | RPN 2721 |

**Range of unlabelled and radiolabelled growth factors and cytokines**

**Cell proliferation assay system and reagents**

| | |
|---|---|
| Cell proliferation assay system - version 2 | RPN 250 |
| Cell proliferation kit (for immunocytochemical/ immunohistochemical measurement) | RPN 200 |
| Monoclonal anti-bromodeoxyuridine | RPN 202 |
| Cell proliferation labelling reagent | RPN 201 |

**Pipettes and pipette tips**

| Single channel, variable volume pipettes<br>Volme range | |
|---|---|
| 0.5–10 µl | RPN 2340 |
| 5–50 µl | RPN 2341 |
| 50–200 µl | RPN 2342 |
| 200–1000 µl | RPN 2343 |
| 1–5 ml | RPN 2344 |

| Multi-channel, variable volume pipettes | |
|---|---|
| 8 channel, 5–50 µl | RPN 2372 |
| 8 channel, 50–250 µl | RPN 2373 |

GE Healthcare range also includes fixed volume, 4 and 12 multi-channel variable volume pipettes, a range of pipette tips and related accessories.

**GE Healthcare offices:**

GE Healthcare Bio-Sciences AB
Björkgatan 30 751 84
Uppsala
Sweden

GE Healthcare Europe GmbH
Munzinger Strasse 5 D-79111
Freiburg
Germany

GE Healthcare UK Limited
Amersham Place
Little Chalfont
Buckinghamshire
HP7 9NA
UK

GE Healthcare Bio-Sciences Corp
800 Centennial Avenue
P.O. Box 1327
Piscataway
NJ 08855-1327
USA

GE Healthcare Bio-Sciences KK
Sanken Bldg. 3-25-1
Hyakunincho Shinjuku-ku
Tokyo 169-0073
Japan

**GE Healthcare regional office contact numbers:**

**Asia Pacific**
Tel: +85 65 62751830
Fax: +85 65 62751829

**Australasia**
Tel: + 61 2 8820 8299
Fax: +61 2 8820 8200

**Austria**
Tel: 01/57606-1613
Fax: 01/57606-1614

**Belgium**
Tel: 0800 73 890
Fax: 02 416 8206

**Canada**
Tel: 1 800 463 5800
Fax: 1 800 567 1008

**Central, East, & South East Europe**
Tel: +43 1 972 720
Fax: +43 1 972 722 750

**Denmark**
Tel: 45 70 25 24 50
Fax: 45 45 16 2424

**Eire**
Tel: 1 800 709992
Fax: +44 1494 542010

**Finland & Baltics**
Tel: +358 9 512 3940
Fax: +358 9 512 39439

**France**
Tel: 01 69 35 67 00
Fax: 01 69 41 98 77

**Germany**
Tel: 0800 9080 711
Fax: 0800 9080 712

**Greater China**
Tel: +852 2100 6300
Fax: +852 2100 6338

**Italy**
Tel: 02 26001 320
Fax: 02 26001 399

**Japan**
Tel: +81 3 5331 9336
Fax: +81 3 5331 9370

**Korea**
Tel: 82 2 6201 3700
Fax: 82 2 6201 3803

**Latin America**
Tel: +55 11 3933 7300
Fax: + 55 11 3933 7304

**Middle East & Africa**
Tel: +30 210 96 00 687
Fax: +30 210 96 00 693

**Netherlands**
Tel: 0800-82 82 82 1
Fax: 0800-82 82 82 4

**Norway**
Tel: +47 815 65 777
Fax: +47 815 65 666

**Portugal**
Tel: 21 417 7035
Fax: 21 417 3184

**Russia, C.I.S. & N.I.S**
Tel: +7 495 956 5177
Fax: +7 495 956 5176

**Spain**
Tel: 902 11 72 65
Fax: 935 94 49 65

**Sweden**
Tel: 018 612 1900
Fax: 018 612 1910

**Switzerland**
Tel: 0848 8028 10
Fax: 0848 8028 11

**UK**
Tel: 0800 515 313
Fax: 0800 616 927

**USA**
Tel: +1 800 526 3593
Fax: +1 877 295 8102

http://www.gehealthcare.com/lifesciences
GE Healthcare UK Limited
Amersham Place, Little Chalfont, Buckinghamshire, HP7 9NA
UK

imagination at work

RPN2722PL Rev C 2006