



Revised 17 Feb. 2008 (Vers. 3.0)

For Veterinary Use Only

For research use only. Not intended for any animal or human therapeutic or diagnostic use.

**Contents and Storage** 

**Storage** Store at 2 to 8°C.

#### **Contents**

Reagents Provided	96 Test Kit	192 Test Kit
Sw IL-8 Standard, lyophilized, recombinant Sw IL-8. Refer to vial label for quantity and reconstitution volume.	2 vials	4 vials
Standard Diluent Buffer. Contains 15 mM sodium azide; 25 mL per bottle.	1 bottle	2 bottles
Incubation Buffer. Contains 8 mM sodium azide; 12 ml per bottle.	1 bottle	1 bottle
Sw IL-8 Antibody-Coated Wells, 96 wells per plate.	1 plate	2 plates
Sw IL-8 Biotin Conjugate, (Biotin-labeled anti-IL-8). Contains 15 mM sodium azide; 11 mL per bottle.	1 bottle	2 bottles
Streptavidin-Peroxidase (HRP), (100x) concentrate. Contains 3.3 mM thymol; 0.125 mL per vial.	1 vial	2 vials
Streptavidin-Peroxidase (HRP) Diluent. Contains 3.3 mM thymol; 25 mL per bottle.	1 bottle	1 bottle
Wash Buffer Concentrate (25x); 100 mL per bottle.	1 bottle	1 bottle
Stabilized Chromogen, Tetramethylbenzidine TMB); 25 mL per bottle.	1 bottle	1 bottle
Stop Solution; 25 mL per bottle.	1 bottle	1 bottle
Plate Covers, adhesive strips.	3	6

**Disposal Note:** This kit contains materials with small quantities of sodium azide. Sodium azide reacts with lead and copper plumbing to form explosive metal azides. Upon disposal, flush drains with a large volume of water to prevent azide accumulation. Avoid ingestion and contact with eyes, skin and mucous membranes. In case of contact, rinse affected area with plenty of water. Observe all federal, state and local regulations for disposal.

#### Safety

All blood components and biological materials should be handled as potentially hazardous. Follow universal precautions as established by the Centers for Disease Control and Prevention and by the Occupational Safety and Health Administration when handling and disposing of infectious agents.





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#### Introduction

#### Intended Use

The DRG Swine Interleukin-8 (Sw IL-8) ELISA is to be used for the in vitro quantitative determination of Sw IL-8 in serum, buffered solution, or cell culture medium.

The assay will recognize both natural and recombinant Sw IL-8.

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#### Principle of the Method

The DRG Sw IL-8 kit is a solid phase sandwich <u>Enzyme Linked-Immuno-Sorbent Assay</u> (ELISA). An antibody specific for Sw IL-8 has been coated onto the wells of the microtiter strips provided. Samples, including standards of known Sw IL-8 content, control specimens, and unknowns, are pipetted into these wells.

During the first incubation, the Sw IL-8 antigen binds to the immobilized (capture) antibody on one site. After washing, a biotinylated antibody specific for Sw IL-8 is added. During the second incubation, this antibody binds to the immobilized Sw IL-8 captured during the first incubation.

After removal of excess second antibody, Streptavidin-Peroxidase (enzyme) is added. This binds to the biotinylated antibody to complete the four-member sandwich. After a third incubation and washing to remove all the unbound enzyme, a substrate solution is added, which is acted upon by the bound enzyme to produce color.

The intensity of this colored product is directly proportional to the concentration of Sw IL-8 present in the original specimen.

### **Background Information**

Interleukin-8/Neutrophil-Activating Peptide-1 selectively stimulates the ability of neutrophils and T-lymphocytes to invade injured or inflamed tissue (1,9,11,13). Exogenous stimuli like LPS (10), but also IL-1, TNF- $\alpha$  and TNF- $\alpha$  induce the secretion of IL-8 (8) in a variety of different cell types including monocytes, endothelial and epithelial cells, peripheral blood mononuclear cells, dermal fibroblasts (8), keratinocytes (8), neutrophils (3), hepatocytes, synovial cells (4), and T-lymphocytes (11). When IL-8 was subcutaneously injected into rats, both lymphocytes and neutrophils migrated to the site of injection within 3 hours. At lower dosages, only lymphocytes migrated towards the site of injection, while at higher dosages primarily neutrophils were attracted. It was found that

T-lymphocytes are 10 times more sensitive to IL-8 than neutrophils. IL-8 exerts its effects via specific cell membrane receptors (chemotactic agonist receptor-family) with homogeneous high-affinity activity and two binding sites for its ligand (2,7). The receptor density is determined by the cell type and ranges from 300 on T-lymphocytes up to 20,000 on neutrophils (11). After binding of IL-8, the receptor expression is downregulated >90% within 10 minutes at 37°C, together with the internalization of the ligand (11,12). IL-8 is proteolytically degraded in the cytoplasm and released into the culture medium as soluble fragments (11).

The IL-8 receptors are probably recycled (12). Besides its chemotactic influence, IL-8 exerts other distinct characteristics. In neutrophils it triggers the secretion of superoxide anions and lysosomal enzymes, thereby indirectly augmenting the permeability of blood vessels (14), and IL-8 enhances the fungicidal activity against *Candida albicans*. Neutrophils are more readily liberated from the bone marrow reservoir under the influence of this cytokine (11). *In vitro*, IL-8 stimulates a rapid Mac-1 as well as CR 1, p150,95 and LFA-1 expression on neutrophils which enables the adherence to activated vascular endothelial cells expressing e.g. ICAM-1 (5). This may account for the accumulation of neutrophils at IL-8 injection sites. Other findings suggest that endothelial-derived IL-8 may function to attenuate inflammatory events at the interface between vessel wall and blood, via inhibiting neutrophil adhesion to cytokine-activated endothelial monolayers. Therefore these cells seem to be protected against neutrophil-mediated damage (6). In basophils, besides its chemotactic effects, IL-8 stimulates the histamine liberation.





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The property of IL-8 to stimulate movement of neutrophils across endothelial monolayers *in vitro* supports the concept of a central role for this molecule in the accumulation of neutrophils at inflammatory lesions *in vivo*.

#### Methods

#### Materials Needed But Not Provided

- Microtiter plate reader (at or near 450 nm) with software
- Calibrated adjustable precision pipettes
- Distilled or deionized water
- Plate washer: automated or manual (squirt bottle, manifold dispenser, etc.)
- Glass or plastic tubes for diluting solutions
- Absorbent paper towels
- Calibrated beakers and graduated cylinders

#### **Procedural Notes**

- 1. When not in use, kit components should be refrigerated. All reagents should be warmed to room temperature before use.
- 2. Microtiter plates should be allowed to come to room temperature before opening the foil bags. Once the desired number of strips has been removed, immediately reseal the bag and store at 2 to 8°C to maintain plate integrity.
- 3. Samples should be collected in pyrogen/endotoxin-free tubes.
- 4. Samples should be frozen if not analyzed shortly after collection. Avoid multiple freeze-thaw cycles of frozen samples. Thaw completely and mix well prior to analysis.
- 5. When possible, avoid use of badly hemolyzed or lipemic sera. If large amounts of particulate matter are present, centrifuge or filter prior to analysis.
- 6. It is recommended that all standards, controls and samples be run in duplicate.
- 7. When pipetting reagents, maintain a consistent order of addition from well-to-well. This ensures equal incubation times for all wells.
- 8. Do not mix or interchange different reagent lots from various kit lots.
- 9. Do not use reagents after the kit expiration date.
- 10. Absorbances should be read immediately, but can be read up to 2 hours of assay completion. For best results, keep plate covered in the dark.
- 11. In-house controls should be run with every assay. If control values fall outside pre-established ranges, the accuracy of the assay is suspect.
- 12. All residual wash liquid must be drained from the wells by efficient aspiration or by decantation followed by tapping the plate forcefully on absorbent paper. *Never* insert absorbent paper directly into the wells.
- 13. Because Stabilized *Chromogen* is light sensitive, avoid prolonged exposure to light. Avoid contact between chromogen and metal, or color may develop.





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### **Directions for Washing**

- o **Incomplete washing will adversely affect the test outcome**. All washing must be performed with *Wash Buffer* provided.
- o Washing can be performed manually as follows: completely aspirate the liquid from all wells by gently lowering an aspiration tip into the bottom of each well. Take care not to scratch the inside of the well. After aspiration, fill the wells with at least 0.4 mL of diluted *Wash Buffer*. Let soak for 15 to 30 seconds, then aspirate the liquid. Repeat as directed under Assay Procedure. After the washing procedure, the plate is inverted and tapped dry on absorbent tissue.
- O Alternatively, the *Wash Buffer* may be put into a squirt bottle. If a squirt bottle is used, flood the plate with *Wash Buffer*, completely filling all wells. After the washing procedure, the plate is inverted and tapped dry on absorbent tissue.
- o If using an automated washer, follow the washing instructions carefully.

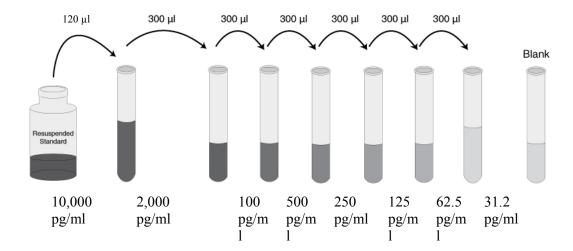
### Preparation of Reagents

#### **Dilution of Standard**

- 1. Reconstitute standard to 10,000 pg/mL with *Standard Diluent Buffer*. Refer to standard vial label for instructions. Swirl or mix gently and allow to sit for 10 minutes to ensure complete reconstitution. Use standard within 1 hour of reconstitution.
- 2. Add 0.120 mL of the reconstituted standard to a tube containing 0.480 mL *Standard Diluent Buffer*. Label as 2000 pg/mL Sw IL-8. Mix.
- 3. Add 0.300 mL of Standard Diluent Buffer to each of 6 tubes 1,000, 500, 250, 125, 62.5 and 31.2 pg/ml Sw IL 8
- 4. Make serial dilutions of the standard as described in the following dilution figure. Mix thoroughly between steps.

#### **Important:**

Remaining reconstituted standard should be discarded. Return the Standard Diluent Buffer to the refrigerator.







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### **Preparing Streptavidin-HRP**

*Note:* **Prepare within 15 minutes of usage**. The *Streptavidin-HRP* 100x concentrate is in 50% glycerol. This solution is viscous. To ensure accurate dilution, allow *Streptavidin-HRP* concentrate to reach room temperature. Gently mix. Pipette *Streptavidin-HRP* concentrate slowly. Remove excess concentrate solution from pipette tip by gently wiping with clean absorbent paper.

- 1. Dilute 10 μL of this 100x concentrated solution with 1 mL of *Streptavidin-HRP Diluent* for each 8-well strip used in the assay. Label as Streptavidin-HRP Working Solution.
- 2. Return the unused *Streptavidin-HRP* concentrate to the refrigerator.

# of 8-Well Strips	Volume of Streptavidin-HRP Concentrate	<b>Volume of Diluent</b>
2	20 μL solution	2 mL
4	40 μL solution	4 mL
6	60 μL solution	6 mL
8	80 μL solution	8 mL
10	100 μL solution	10 mL
12	120 μL solution	12 mL

#### **Dilution of Wash Buffer**

- Allow the 25x concentrate to reach room temperature and mix to ensure that any precipitated salts have redissolved.
  Dilute 1 volume of the 25x wash buffer concentrate with 24 volumes of deionized water
  (e.g., 50 mL may be diluted up to 1.25 liters, 100 mL may be diluted up to 2.5 liters).
  Label as Working Wash Buffer.
- 2. Store both the concentrate and the Working Wash Buffer in the refrigerator. The diluted buffer should be used within 14 days.

#### Assay Procedure

### Be sure to read the *Procedural Notes* section before carrying out the assay.

Allow all reagents to reach room temperature before use. Gently mix all liquid reagents prior to use.

**Note**: A standard curve must be run with each assay.

- 1. Determine the number of 8-well strips needed for the assay. Insert these in the frame(s) for current use. (Re-bag extra strips and frame. Store these in the refrigerator for future use.)
- 2. Add 50 µl of the *Incubation Buffer* to all wells. Well(s) reserved for chromogen blank should be left empty.
- 3. Add 100 µl of the *Standard Diluent Buffer* to the zero standard wells. Well(s) reserved for chromogen blank should be left empty.
- 4. For the standard curve, add 100  $\mu$ l of standards to the appropriate microtiter wells.
  - See PREPARATION OF REAGENTS.
  - For buffered solutions or cell culture samples, add  $100 \mu l$  of sample to each well. For serum samples and controls, add  $50 \mu l$  of *Standard Diluent Buffer* to each well followed by  $50 \mu l$  of sample.
- 5. Cover plate with *plate cover* and incubate for **2 hours at room temperature**.
- 6. Thoroughly aspirate or decant solution from wells and discard the liquid. Wash wells 4 times. See DIRECTIONS FOR WASHING.





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- 7. Pipette 100 μl of biotinylated anti-IL-8 (*Biotin Conjugate*) solution into each well except the chromogen blank(s). Tap gently on the side of the plate to mix.
- 8. Cover plate with *plate cover* and incubate for **1 hour at room temperature**.
- 9. Thoroughly aspirate or decant solution from wells and discard the liquid. Wash wells 4 times. See DIRECTIONS FOR WASHING.
- 10. Add 100 μl Streptavidin-HRP Working Solution to each well except the chromogen blank(s). See PREPARATION OF REAGENTS.
- 11. Cover plate with the *plate cover* and incubate for **30 minutes at room temperature**.
- 12. Thoroughly aspirate or decant solution from wells and discard the liquid. Wash wells 4 times. See DIRECTIONS FOR WASHING.
- 13. Add 100 µl of Stabilized Chromogen to each well. The liquid in the wells will begin to turn blue.
- 14. Incubate for **30** minutes at room temperature and in the dark. *Note*: Do not cover the plate with aluminum foil or metalized mylar. The incubation time for chromogen substrate is often determined by the microtiter plate reader used. Many plate readers have the capacity to record a maximum optical density (O.D.) of 2.0. The O.D. values should be monitored and the substrate reaction stopped before the O.D. of the positive wells exceed the limits of the instrument. The O.D. values at 450 nm can only be read after the *Stop Solution* has been added to each well. If using a reader that records only to 2.0 O.D., stopping the assay after 20 to 25 minutes is suggested.
- 15. Add 100 μl of *Stop Solution* to each well. Tap side of plate gently to mix. The solution in the wells should change from blue to yellow.
- 16. Read the absorbance of each well at 450 nm having blanked the plate reader against a chromogen blank composed of 100 µl each of *Stabilized Chromogen* and *Stop Solution*. Read the plate within 2 hours after adding the *Stop Solution*.
- 17. Use a curve fitting software to generate the standard curve. A four parameter algorithm provides the best standard curve fit.
- 18. Read the concentrations for unknown samples and controls from the standard curve. **Multiply value(s) obtained for serum sample(s) by 2 to correct for the 1:2 dilution.** (Samples producing signals greater than that of the highest standard should be diluted in *Standard Diluent Buffer* and reanalyzed, multiplying the concentration found by the appropriate dilution factor.)





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#### Typical Data (Example)

The following data were obtained for the various standards over the range of 0 to 2000 pg/mL Sw IL-8.

Standard Sw IL-1β (pg/mL)	Optical Density (450 nm)
2000	2.45
1000	1.58
500	0.86
250	0.50
125	0.34
62.5	0.25
31.2	0.20
0	0.13

#### **Performance Characteristics**

### Sensitivity

The minimum detectable dose of Sw IL-8 is < 10 pg/mL. This was determined by adding two standard deviations to the mean O.D. obtained when the zero standard was assayed 30 times.

### Precision

### 1. <u>Intra-Assay Precision</u>

Samples of known Sw IL-8 concentration were assayed in replicates of 16 to determine precision within an assay.

	Sample 1	Sample 2	Sample 3	
Mean (pg/mL)	251.3	572.4	1072.9	
SD	18.8	35.1	82.4	
%CV	7.5	6.1	7.7	
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SD = Standard Deviation

CV = Coefficient of Variation





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#### 2. Inter-Assay Precision

Samples were assayed 48 times in multiple assays to determine precision between assays.

	Sample 1	Sample 2	Sample 3	
Mean (pg/mL)	252.8	609.3	1067.8	
SD	20.0	51.7	99.2	
%CV	7.9	8.5	9.3	

SD = Standard Deviation

CV = Coefficient of Variation

#### Linearity of Dilution

Swine serum containing 1985 pg/mL of measured Sw IL 8 was serially diluted in Standard Diluent Buffer over the range of the assay. Linear regression analysis of samples versus the expected concentration yielded a correlation coefficient of 0.99.

#### Recovery

The recovery of Sw IL 8 added to pooled swine serum averaged 93%.

The recovery of Sw IL 8 added to tissue culture medium containing 1% fetal bovine serum averaged 107%, while the recovery of Sw IL 8 added to tissue culture medium containing 10% fetal bovine serum averaged 103%.

Sera from Yorkshire and Chester White pigs have been validated for use in this assay.

Other strains of swine have not been tested and consequently their use has not been validated.

#### **Specificity**

Buffered solutions of a panel of substances at 10,000 pg/mLl were assayed with the Sw IL 8 kit. The following substances were tested and found to have no cross reactivity:

human IL 1 $\beta$ , IL 2, IL 3, IL 4, IL 7, IL 10, IL 13, IFN  $\gamma$ , TNF  $\alpha$ , SCF;

mouse IL 1β, IL 2, IL 4, IL 6, IL 10, IFN γ, TNF α rat IFN γ, MCP 1, TNF α.

Significant cross reactivity was observed to human IL 8.

#### Limitations of the Procedure

Do not extrapolate the standard curve beyond the top standard point; the dose-response is non-linear in this region and accuracy is difficult to obtain. Dilute all samples above the top standard point with *Standard Diluent Buffer*; reanalyze these and multiply results by the appropriate dilution factor.

The influence of various drugs, aberrant sera (hemolyzed, hyperlipidemic, jaundiced, etc.) and the use of biological fluids in place of serum samples have not been thoroughly investigated. The rate of degradation of native Sw IL-8 in various matrices has not been investigated. The immunoassay literature contains frequent references to aberrant signals seen with some sera, attributed to heterophilic antibodies. Though such samples have not been seen to date, the possibility of this occurrence cannot be excluded.





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## **Appendix**

## Troubleshooting Guide

Elevated background	Cause: Insufficient washing and/or draining of wells after washing. Solution containing either biotin or Streptavidin-HRP can elevate the background if residual is left in the well. Solution: Wash according to the protocol. Verify the function of automated plate washer. At the end of each washing step, invert plate on absorbent tissue on countertop and allow to completely drain and tap forcefully if necessary to remove residual fluid. Cause: Contamination of substrate solution with metal ions or oxidizing reagents. Solution: Use distilled/deionized water for dilution of wash buffer and use plastic equipment. DO NOT COVER plate with foil. Cause: Contamination of pipette, dispensing reservoir or substrate solution with Streptavidin-HRP conjugate. Solution: Do not use chromogen that appears blue prior to dispensing onto the plate. Obtain new vial of chromogen. Cause: Incubation time is too long or incubation temperature is too high. Solution: Reduce incubation time and/or temperature.
Elevated sample/ standard ODs	Cause: Incorrect dilution of standard stock solution; intermediary dilutions not followed correctly.  Solution: Follow the protocol instructions regarding the dilution of the standard.  Cause: Incorrect dilution of the Streptavidin-HRP conjugate.  Solution: Warm solution of Streptavidin-HRP concentrate to room temperature, draw up slowly and wipe tip with kim-wipe to remove excess. Dilute ONLY in Streptavidin diluent provided.  Cause: Incubation times extended.  Solution: Follow incubation times outlined in protocol.  Cause: Incubations carried out at 37°C when RT is dictated.  Solution: Perform incubations at RT (= 25 ± 2°C) when instructed in the protocol.
Poor standard curve	Cause: Improper preparation of standard stock solution.  Solution: Dilute lyophilized standard as directed by the vial label only with the standard diluent buffer or in a diluent that most closely matches the matrix of your sample.  Cause: Reagents (lyophilized standard, standard diluent buffer, etc.) from different kits, either different cytokine or different lot number, were substituted.  Solution: NEVER substitute any components from another kit.  Cause: Errors in pipetting the standard or subsequent steps.  Solution: Always dispense into wells quickly and in the same order. Do not touch the pipette tip on the individual microwells when dispensing. Use calibrated pipettes and the appropriate tips for that device.





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Weak/no color develops	Cause: Reagents not at RT (25 ± 2°C) at start of assay.  Solution: Allow ALL reagents to warm to RT prior to commencing assay.  Cause: Incorrect storage of components, e.g., not stored at 2 to 8°C.  Solution: Store all components exactly as directed in protocol and on labels.  Cause: Working Streptavidin-HRP solution made up longer than 15 minutes before use in assay.  Solution: Use the diluted Streptavidin-HRP within 15 minutes of dilution.  Cause: TMB solution lost activity.  Solution 1: The TMB solution should be clear before it is dispensed into the wells of the microtiter plate. An intense aqua blue color indicates that the product is contaminated. Please contact Technical Support if this problem is noted. To avoid contamination, we recommend that the quantity required for an assay be dispensed into a disposable trough for pipetting. Any TMB solution left in the trough should be discarded.  Solution 2: Avoid contact of the TMB solution with items containing metal ions.  Cause: Attempt to measure analyte in a matrix for which the ELISA assay has not been optimized.  Solution: Please contact Technical Support for advice when using nonvalidated sample types.  Cause: Wells have been scratched with pipette tip or washing tips.  Solution: Use caution when dispensing and aspirating into and out of microwells.
Poor Precision	Cause: Errors in pipetting the standards, samples or subsequent steps.  Solution: Always dispense into wells quickly and in the same order. Do not touch the pipette tip on the individual microwells when dispensing. Use calibrated pipettes and the appropriate tips for that device. Check for any leaks in the pipette tip.  Cause: Repetitive use of tips for several samples or different reagents.  Solution: Use fresh tips for each sample or reagent transfer.  Cause: Wells have been scratched with pipette tip or washing tips.  Solution: Use caution when dispensing and aspirating into and out of microwells.

#### References

- 1. Baggiolini, M., et al. (1989) J. Clin. Invest. 84:1045-1049.
- 2. Baldwin, E.T., et al. (1991) Proc. Natl. Acad. Sci. USA (Biochemistry) 88:502-506.
- 3. Bazzoni, F., et al. (1991) J. Exp. Med. 173:771-774.
- 4. DeMarco, D., et al. (1991) Biochem. Biophys. Res. Comm. 174:411-416.
- 5. Detmers, P.A., et al. (1990) J. Exp. Med. 171:1155-1162.
- 6. Gimbrone, M.A., et al. (1989) *Science* 246:1601-1603.
- 7. Grob, P.M., et al. (1990) J. Biol. Chem. 265:8311-8316.
- 8. Larsen, C.G., et al. (1989) *Immunology* 68:31-36.
- 9. Leonard, E.J. (1990) NAP-1 (IL-8). Immunol. Today 11:223-224.
- 10. Martich, G.D., et al. (1991) J. Exp. Med. 173:1021-1024.
- 11. Matsushima, K. and J.J. Oppenheim (1989) Cytokine 1:2-13.





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- 12. Samanta, A.K., et al. (1990) J. Biol. Chem. 265:183-189.
- 13. Smith, W.B., et al. (1991) Immunology 72:65-72.
- 14. Sticherling, M., et al. (1991) J. Invest. Dermatol. 96:26-30.

## **Limited Warranty**

DRG is committed to providing our customers with high-quality goods and services. Our goal is to ensure that every customer is 100% satisfied with our products and our service. If you should have any questions or concerns about a DRG product or service, please contact our Technical Support Representatives. DRG warrants that all of its products will perform according to the specifications stated on the Certificate of Analysis. The company will replace, free of charge, any product that does not meet those specifications. This warranty limits DRG Corporation's liability only to the cost of the product. No warranty is granted for products beyond their listed expiration date. No warranty is applicable unless all product components are stored in accordance with instructions. DRG reserves the right to select the method(s) used to analyze a product unless DRG agrees to a specified method in writing prior to acceptance of the order. DRG makes every effort to ensure the accuracy of its publications, but realizes that the occasional typographical or other error is inevitable. Therefore DRG makes no warranty of any kind regarding the contents of any publications or documentation. If you discover an error in any of our publications, please report it to our Technical Support Representatives. DRG assumes no responsibility or liability for any special, incidental, indirect or consequential loss or damage whatsoever. The above limited warranty is sole and exclusive. No other warranty is made, whether expressed or implied, including any warranty of merchantability or fitness for a particular purpose.





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## Symbols used with DRG Assays

Symbol	English	Deutsch	Français	Español	Italiano
[]i	Consult instructions for use	Gebrauchsanweisung beachten	Consulter les instructions d'utilisation	Consulte las instrucciones de uso	Consultare le istruzioni per l'uso
C€	European Conformity	CE-Konfirmitäts- kennzeichnung	Conformité aux normes européennes	Conformidad europea	Conformità europea
IVD	In vitro diagnostic device	In-vitro-Diagnostikum	Usage Diagnostic in vitro	Para uso Diagnóstico in vitro	Per uso Diagnostica in vitro
RUO	For research use only	Nur für Forschungszwecke	Seulement dans le cadre de recherches	Sólo para uso en investigación	Solo a scopo di ricerca
REF	Catalogue number	Katalog-Nr.	Numéro de catalogue	Número de catálogo	Numero di Catalogo
LOT	Lot. No. / Batch code	Chargen-Nr.	Numéro de lot	Número de lote	Numero di lotto
$\sum$	Contains sufficient for <n> tests/</n>	Ausreichend für "n" Ansätze	Contenu suffisant pour "n" tests	Contenido suficiente para <n> ensayos</n>	Contenuto sufficiente per "n" saggi
1	Storage Temperature	Lagerungstemperatur	Température de conservation	Temperatura de conservación	Temperatura di conservazione
$\square$	Expiration Date	Mindesthaltbarkeits-datum	Date limite d'utilisation	Fecha de caducidad	Data di scadenza
***	Legal Manufacturer	Hersteller	Fabricant	Fabricante	Fabbricante
Distributed by	Distributor	Vertreiber	Distributeur	Distribuidor	Distributore
Content	Content	Inhalt	Conditionnement	Contenido	Contenuto
Volume/No.	Volume / No.	Volumen/Anzahl	Volume/Quantité	Volumen/Número	Volume/Quantità

Symbol	Portugues	Dansk	Svenska	Ελληνικά
(i	Consulte as instruções de utilização	Se brugsanvisning	Se bruksanvisningen	Εγχειρίδιο χρήστη
(€	Conformidade com as normas europeias	Europaeisk overensstemmelse	Europeisk överensstämmelse	Ευρωπαϊκή Συμμόρφωση
IVD	Diagnóstico in vitro	In vitro diagnostik	Diagnostik in vitro	in vitro διαγνωστικό
RUO				
REF	Catálogo n.º	Katalognummer	Katalog nummer	Αριθμός καταλόγου
LOT	No do lote	Lot nummer	Batch-nummer	Αριθμός Παρτίδος
$\sum$		Indeholder tilsttrækkeligt til "n" test	Innehåller tillräckligt till "n" tester	Περιεχόμενο επαρκές για «n» εξετάσεις
1	Temperatura de conservação	Opbevarings-temperatur	Förvaringstempratur	Θερμοκρασία αποθήκευσης
	Prazo de validade	Udløbsdato	Bäst före datum	Ημερομηνία λήξης
***	Fabricante	Producent	Tillverkare	Κατασκευαστής
Distributed by				
Content	Conteúdo	Indhold	Innehåll	Περιεχόμενο
Volume/No.	Volume/Número	Volumen/antal	Volym/antal	Όγκος/αριθ