



Thermo Scientific MSIA Insulin D.A.R.T.'S

THERMO SCIENTIFIC™ MSIA™ INSULIN D.A.R.T.'S AND MSIA™ INSULIN WORKFLOW

Products Information:

MSIA™ Insulin D.A.R.T.'S		
Compatible with the Versette™ Automated Liquid Platform (part # 650-MSIA) and FinnpiPette® Novus i Multichannel Electronic Pipettes (for immuno-precipitation) (part # 991SP12)		
<i>Cat. No.</i>	<i>Description</i>	<i>Packaging</i>
991001096	MSIA™ Insulin D.A.R.T.'S	Pack of 96 tips
991001024	MSIA™ Insulin D.A.R.T.'S	Pack of 24 tips

Storage

Upon receipt, store at 4°C. Product shipped with an ice pack.

Disclaimer

These products are supplied for life science research use only. They are not intended for medicinal, diagnostic or therapeutic use.

Warnings/Precautions

- MSIA Insulin D.A.R.T.'S are for Research Use Only. Not to be used in diagnostic procedures.
- Do not use reagents beyond the expiration date.
- Do not use MSIA Insulin D.A.R.T.'S by mouth.
- Do not smoke or eat in areas in which MSIA Insulin D.A.R.T.'S or specimens are handled.
- Avoid contact of skin and mucous membranes with specimens.
- Handle all biological samples and materials in contact with the biological sample in accordance with CLSI guidelines for preventing the transmission of blood-borne pathogens during laboratory procedures.
- Contamination of MSIA Insulin D.A.R.T.'S may result in erroneous results.
- Do not use glass tubes for biological samples.

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Introduction

The Mass Spectrometric Immunoassay (MSIA™) is a fast, convenient and highly reproducible method for the enrichment and mass spectrometric detection of targeted analytes. The use of immuno-enrichment provides increased assay sensitivity, while mass spectrometric detection provides greater specificity with the ability to simultaneously target and differentiate protein variations within the targeted analyte.

Integration of Thermo Scientific™ MSIA™ D.A.R.T.'S, Thermo Scientific™ automated/semi-automated liquid handlers, and Thermo Scientific™ LC/MS platforms together provides a convenient and effective system to perform MSIA.

Thermo Scientific™ MSIA™ Insulin D.A.R.T.'S provide efficient enrichment of insulin from biological samples, facilitating the simultaneous and sensitive detection of insulin and insulin analogues by Thermo Scientific LC/MS platforms. The MSIA Insulin D.A.R.T.'S anti-insulin antibody targets a conserved region within the insulin beta-chain enabling various insulin analogues, including endogenous human insulin, therapeutic analogues, as well as non-human animal analogues, to be enriched for mass spectrometric detection. The convenient pipette tip format of the MSIA Insulin D.A.R.T.'S allow for insulin to be enriched in parallel from multiple samples with the use of Thermo Scientific liquid handlers and enable a wide range of sample volumes from as little as few micro-liters to as large as milliliter volumes to be addressed. Immuno-affinity capture and enrichment of insulin analogues from prepared biological samples using the MSIA Insulin D.A.R.T.'S is performed through a simple repetitive pipetting motion. After insulin enrichment, the tips are rinsed (in the same fashion) and the bound insulin analogues are then dissociated from the tips using an elution buffer selected specifically for the Insulin MSIA.

Important Product Information

- Store the Thermo Scientific™ MSIA™ Insulin D.A.R.T.'S at 4°C, do not freeze.
- This product is intended for single use only.
- Thermo Scientific™ MSIA™ Insulin D.A.R.T.'S are not intended for the transfer or measurement of liquids. This product is intended for micro-scale analyte purification of insulin prior to mass spectrometric detection.
- For more information, see www.thermoscientific.com/msia.

Procedure for MSIA Insulin

Note: The following information provided in this procedure describes the use of the Thermo Scientific™ MSIA™ Insulin D.A.R.T.S® in Insulin MSIA™ using the semi-automated Thermo Scientific™ Novus i multi-channel bench top/mobile platform. The steps described can easily be translated to a fully automated Versette robotic platform (please contact MSIA™ support for more details). These procedures are intended for **general guidance only** and may require further optimization by the end-user.

A. Additional Materials Required

- Thermo Scientific™ Finnpipette Novus i Electronic Multichannel Pipette and adjustable pipette stand (See Appendix A, MSIA™ related Thermo Scientific™ Products)
- Assortment of Finnpipette*F1 Adjustable-volume Pipette.
- Assortment of Finntip*Flex* Pipet Tips
- 96-well polypropylene microplates, 0.5mL, 1mL, or 2mL (Thermo Scientific™ Product No. 267334, 260251, 278743). Please note it is highly recommend using low-bind plastic ware to ensure efficient performance of the assay and prevent loss of analyte due to adsorption to the plastics.
- 96-well Robotic PCR Plate (Thermo Scientific™ Product No. AB-1300)
- Insulin, human recombinant (Life Technologies™ Product No. 12585-014)
- 4[D10] Leu Insulin, human (Peptides International, Product No. PLP-3404-v)
- ACTH 1-24 (Cell Science Product No. CRA108)
- N-Octyl-β-D-glucoside (NOG, Thermo Scientific™ Product No. 28310)
- Ultra-pure water, LC-MS grade preferred (Fisher Scientific Product No. W6-4).
- Wash Buffer: Phosphate-buffered saline, pH 7.2 (PBS, Thermo Scientific™ Product No. 28372)
- Dilution Buffer: 150 mM NOG in Phosphate-buffered saline, pH 7.2
- Elution solvent: 15mg/L ACTH 1-24 in 33% acetonitrile in aqueous 0.4% trifluoroacetic acid.

B. Alternate Equipment

- Thermo Scientific™ Versette Liquid Handling Platform (See Appendix A, MSIA™ related Thermo Scientific™ Products)

C. Considerations

- A single MSIA™ Insulin D.A.R.T.S® is required for each sample.
- Dedicate a row or column of the microtiter plate for assay development, thus allowing up to 8 (per column) or 12 (per row) samples to be addressed in parallel with the Finnpipette Novus i Electronic 12-Channel Pipette. For larger numbers of sample the Thermo Scientific™ Versette Liquid Handling Platform is available, allowing for up to 96 samples to be processed in parallel.
- An internal reference is recommended to be added to all samples to enable normalization of all events from sample preparation to detection by LC-MS. A suggested internal reference for Insulin MSIA is 4[D10] Leu Insulin, due to its cross-reactivity with the MSIA™ Insulin D.A.R.T.S and ability to be differentiated from other insulin analogues by LC-MS.
- If the samples were frozen or refrigerated, they must be thawed and/or warmed to at least room temperature before preparation (~25 °C). A 37°C water bath may be employed to expedite this process and ensure consistent sample temperature. It is highly recommended that freeze/thaw cycles be avoided for biological samples.

Quick Tip: Centrifuge raw samples prior to aliquoting to ensure the removal of any particulates or debris that may clog the MSIA™ Insulin D.A.R.T.S®.

- Dilution and Wash Buffer can be prepared in bulk and stored at 4°C until use. Buffers must be warmed to room temperature (~25 °C) prior to use for efficient performance.
- Store insulin standards following manufacture's recommendations, preferably at $\geq 1\text{g/L}$ and aliquoted to avoid repeated freeze/thaw cycles.

Quick Tip: To reduce loss of the eluted analyte to absorption to plastic or glassware, a blocking agent (e. g., 15 mg/L adrenocorticotrophic hormone (ACTH) in the elution solvent) may be used. Care should be taken in selecting a blocking agent that does not negatively affect downstream processes or detection of the targeted analyte.

- Transfer the eluent directly from the MSIA Insulin D.A.R.T.'S to the container that will be used in further processing or analysis to avoid loss of the eluent.

D. Internal Reference

1. Bring Dilution buffer to room temperature (~25°C).
2. Prepare 4[D10] Leu Insulin at 100pM in dilution buffer. For each sample and insulin standard, 250 μL of 100pM 4[D10] Leu Insulin is required. It is recommended to make >10% excess to ensure sufficient amount for entire study
3. The internal reference should be used soon after its preparation (< 1-2 hours) to reduce chances of loss due adsorption to its container.

E. Sample Preparation

1. Allow biological samples to warm to room temperature (~25°C).
2. Centrifuge samples at a low speed to pellet precipitate and debris, then carefully collect sample without disrupting to ensure precipitates and debris are not transferred to sample preparations where they could clog MSIA™ Insulin D.A.R.T.'S.
3. For each sample, deposit 500 μL of the sample into an individual well of a 1mL microplate. It is recommended that the samples be added to the rows (A-H) of the 1mL microplate to enable up to 12 samples to be addressed simultaneously with the 12-channel Novus i.
4. Dilute each sample with 250 μL internal reference prepared in section D.
5. Soon after preparing samples (< 1 hour) proceed to section F to begin extraction of insulin with MSIA™ Insulin D.A.R.T.'S.

F. Insulin Extraction

1. For each sample, two wells of Wash Buffer (200 μL) and two wells of ultra-pure water are required. Deposit 200 μL of wash buffer into the wells of 96-well microplate corresponding to the wells of rows A and B. Deposit 200 μL of ultra-pure water into the wells of the same 96-well microplate corresponding to the wells of rows C and D.

Quick Tip: Dedicate a row or column for each step for washing with Wash Buffer or water in the microtiter plate.

2. If Finnpiptette Novus i Electronic 12-Channel Pipette and Adjustable Pipette Stand (sold separately) is used, snap on multichannel pipette bracket to Finnpiptette Novus i Electronic 12-Channel Pipette.
3. Securely affix MSIA™ Insulin D.A.R.T.'S onto the Finnpiptette Novus i Electronic 12-Channel Pipette nose cone. Mount the Finnpiptette Novus i Electronic 12-Channel Pipette onto its stand, raise pipette up from the deck. Insert microtiter plate onto the stand deck.

4. The workflow for insulin extraction is presented in **Table 1**. The pipetting cycle iterations are performed by methodically aspirating and dispensing (standard pipette mixing action) using the listed cycle volumes provided.

Table 1. Insulin Extraction Workflow for Finnpiquette Novus i Electronic 12-Channel Pipette:

Steps	Description	Microtiter plate Volume (μ L)	Speed Setting	Cycle Volume (μ L)	No. of Cycle Iterations	Approx. Allotted time per step
1	Wash Buffer	200	1	150	20	2 min 50 sec
2	Analytical Sample	750	1	250	500	1hr 11 min
3	Wash Buffer	200	1	150	20	2 min 50 sec
4	Water	200	1	150	20	2 min 50 sec
5	Water	200	1	150	20	2 min 50 sec
6	Elution Solvent	75	1	50	100	

Quick Note: To maintain proper sample and wash flow through the Thermo Scientific MSIA™ Insulin D.A.R.T.'S, ensure that the open end remains completely immersed in the volume of liquid during each step of the workflow. Failure to do so will result in aspiration of air and inconsistency in tip treatment. Also, do not press the MSIA™ Insulin D.A.R.T.'S into the bottom of the liquid holding vesicle as this will restrict flow and prevent consistent performance.

5. Turn on Finnpiquette Novus i Electronic 12-Channel Pipette by pressing the trigger button once.
6. Press Menu (left selection key) and scroll the function list with scroll key and select MSIA function with OK (right selection key).
7. Position the microplate with Wash Buffer under the pipette. Lower the MSIA Insulin D.A.R.T.'S into the Wash Buffer reservoirs using the slide vertical control knob on the stand. Ensure that the open end remains completely immersed in the volume of liquid during each step of the workflow to maintain proper sample and wash flow through the MSIA Insulin D.A.R.T.'S. Failure to do so will result in aspiration of air and inconsistency in tip treatment. Also, do not press the MSIA Insulin D.A.R.T.'S into the bottom of the liquid holding vesicle as this will restrict flow.
8. Pre-rinse MSIA Insulin D.A.R.T.'S with Wash Buffer prior to addressing samples. Select the WASH function on the Finnpiquette Novus i Electronic 12-Channel Pipette. The WASH submenu appears with preferred default values for the three variables of SPEED, VOLUME and number of CYCLES. Please adjust the speed, volume and mixing cycles to match those described in **Table 1** by pressing the edit button.

Quick Tip: Nine PROGRAM submenus under the MSIA menu can be setup with user defined values and saved with preferred SPEED, VOLUME and CYCLE numbers using the pipette scroll key and selection keys.

Quick Tip: Any program may be interrupted by selecting CANCEL (left selection key), which will stop the run.

9. Press the trigger button **once**, located on the back of the pipette handle, to rinse the tips with Wash Buffer. Immediately, the Finnpiquette Novus i Electronic 12-Channel Pipette will begin mixing and the cycle numbers will start counting down. When finished the Finnpiquette Novus i Electronic 12-

Channel Pipette will indicate completion of the WASH step by requiring the user to depress the trigger to perform the “BLOWOUT.”

10. To BLOWOUT, raise the Finnpiptette Novus i Electronic 12-Channel Pipette so that the distal ends of the MSIA Streptavidin D.A.R.T.'S above the liquid level, yet still remaining within the wells of the microtiter plate. Depress the trigger once to allow the Finnpiptette Novus i Electronic 12-Channel Pipette to dispense any remaining liquid from the tips.
11. To extract insulin from samples using the MSIA Insulin D.A.R.T.'S, scroll the function list and select CAPTURE with OK (right selection key). The CAPTURE submenu appears with preferred default values for the three variables of SPEED, VOLUME and number of CYCLES. Please adjust the speed, volume and mixing cycles to the values in **Table 1** by pressing the edit button.
12. Position the microplate with samples under the pipette. Lower the MSIA Insulin D.A.R.T.'S into the samples using the slide vertical control knob on the stand. Ensure that the open end remains completely immersed in the samples during each step of the workflow to maintain proper sample flow through the MSIA Insulin D.A.R.T.'S. In addition, ensure the distal ends of the tips are not pressed against the bottom of the microtiter wells, restricting the flow during pipetting.
13. Press trigger button; cycling starts and the cycle number counts down.
14. After completing CAPTURE step, lift MSIA Insulin D.A.R.T.'S above the samples and press trigger to BLOWOUT the remaining liquid from the tips.
15. Once the MSIA Insulin D.A.R.T.'S are loaded with insulin and internal reference, they are ready for subsequent washes and elution.
16. Position the microplate with Wash Buffer under the pipette. Lower the MSIA Insulin D.A.R.T.'S into the Wash Buffer reservoirs using the slide vertical control knob on the stand. Ensure that the open end remains completely immersed in the volume of liquid during each step of the workflow to maintain proper wash flow through the MSIA Insulin D.A.R.T.'S. Also, do not press the MSIA Insulin D.A.R.T.'S into the bottom of the liquid holding vesicle as this will restrict flow.
17. Select the WASH function with OK. Please adjust the speed, volume and number mixing cycles to match the values in **Table 1**. Again, press trigger button **once** to start each wash step.
18. After completing WASH step, lift MSIA Insulin D.A.R.T.'S above the Wash Buffer and press trigger to BLOWOUT the remaining liquid from the tips.
19. Repeat Steps 16 - 18 for each of the water washes to wash away salts and detergents that are harmful to the LC-MS process. Once completed, captured insulin may be eluted from the MSIA Insulin D.A.R.T.'S by the remaining steps.
20. Elute in the container that will be used in further processing or analysis to avoid loss of the eluent.

Quick Tip: For LC-MS systems equipped with an autosampler capable of loading from microtiter plates, we recommend eluting into an AB1300 PCR plate for these purposes.

21. Fill appropriate wells of an AB1300 plate, or alternative vial, with 75 uL Elution Solvent. The Elution Solvent chosen must sufficiently disrupt the antibody/antigen complex. Adrenocorticotrophic hormone (ACTH) reduces the loss of the eluted analyte to absorption to plastic or glassware.
22. Position the MSIA Insulin D.A.R.T.'S low enough into the wells containing the Elution Solvent to pipet without flow restriction of the solvent through the pipette tips. Select the ELUTE function with OK. Please adjust the speed, volume and number mixing cycles to match the values in **Table 1**. Again, press trigger button **once** to start each wash step.

23. Press trigger button to start elution step. After completing ELUTE step, lift MSIA Insulin D.A.R.T.'S above the samples and press trigger to BLOWOUT function.
24. Dilute eluents with 25 μ L water (LC/MS grade) to reduce organic to < 25% to enable proper sample loading onto LC column.

Quick Tip: The percentage of organic present when loading eluents onto LC column must be less than the percentage of organic when the insulin is eluted from the column during the LC run.

25. Load entire eluent into LC-MS for analyses.

G. MSIA Insulin Quick Start Protocol

Detailed instructions are in Sections C through F.

Quick Tip: Dedicate row(s) or column(s) of Wash Buffer(s) and Water in the microtiter plate for each wash step the assay.

Quick Tip: When positioning the tips, ensure the distal ends of the tips are placed just above the bottom of microtiter well and below the liquid's level. Please note pressing against the bottom of the microtiter wells will restrict the flow.

1. Load a well of Wash Buffer (200 μ L) for each MSIA Insulin D.A.R.T.'S that will be used.
2. WASH- Pre-rinse the tips with Wash Buffer prior to insulin extraction (**Table 1, Step 1**).

Table 1. MSIA Insulin Workflow for Finnpiquette Novus i Electronic 12-Channel Pipette:

Steps	Description	Microtiter plate Volume (μ L)	Speed Setting	Cycle Volume (μ L)	No. of Cycle Iterations	Approx. Allotted time per step
1	Wash Buffer	200	1	150	20	2 min 50 sec
2	Analytical Sample	750	1	250	500	1hr 11 min
3	Wash Buffer	200	1	150	20	2 min 50 sec
4	Water	200	1	150	20	2 min 50 sec
5	Water	200	1	150	20	2 min 50 sec
6	Elution Solvent	75	1	50	100	

3. BLOWOUT- Raise the Finnpiquette Novus i Electronic 12-Channel Pipette so that the ends of the MSIA Insulin D.A.R.T.'S are above the liquid level, yet still remaining within the wells of the microtiter plate. Depress the trigger once to dispense any remaining liquid.
4. Dispense 500 μ L sample into an individual well of 1 mL microplate. Dilute sample with 250 μ L internal reference standard in dilution buffer.
5. CAPTURE- Load the target analyte from the analytical sample, followed by BLOWOUT (**Table 1, Step 2**).
6. WASH- Wash the MSIA Insulin D.A.R.T.'S with Wash Buffer to remove non-specific binding, followed by BLOWOUT (**Table 1, Step 3**).
7. WASH- Rinse with water to remove salts and detergents, followed by BLOWOUT (**Table 1, Steps 4-5**).
8. Elute- Submerge the MSIA Insulin D.A.R.T.'S in 75 μ L Elution Solvent in a low-bind AB-1300 PCR plate. Elute captured insulin and internal standard, followed by BLOWOUT (**Table 1, Steps 6**).

9. Dilute eluent 25 μ L water (LC/MS grade) to reduce percentage of organic to ~25% for efficient loading of insulin onto LC column.
10. Load entire eluent onto LC and analyze by LC-MS (see **Sections H** and **I** for details).

H. Equipment and Accessories for LC-MS

- Ultimate 3000-XRS (Thermo Scientific Product No. 5200.04) with column thermostat and autosampler
- Q Exactive System with Ion Max Source (Thermo Scientific Product No. 726020)
- Pre-column filter (Thermo Scientific Product No. A-138)
- Sample loop 200 μ L (Thermo Scientific Product No. CH-952411)
- Upchurch® Scientific High-Pressure PEEK Tubing Model #1535L (Fisher Scientific Product No. 05-701-18)
- Proswift™ RP-4H Protein Columns 250mm x 1mm (Thermo Scientific Product No. 066640)

I. LC-MS of Insulin

1. Perform LC on a 250mm x 1mm Proswift™ RP-4H Protein Column maintained at 50 °C.
2. Reduce the percentage of organic to 25% by adding 28 μ L of LC-MS grade water to the eluent. An increased organic composition prevents the LC column from retaining the eluent.
3. Elute using a linear gradient comprised of (A) 0.1 % (v/v) formic acid in water and (B) 0.1 % (v/v) formic acid in acetonitrile (10-50 % eluent B over 10 minutes).

Quick Tip: LC-MS grade solvent and additives should be used to achieve greatest sensitivity.

4. Perform MS data acquisition using the Q Exactive™ mass spectrometer with the following source conditions: heated electrospray ionization (HESI-II) source probe temperature of 350 °C, capillary temperature of 325 °C, sheath 40 AU, auxiliary 12 AU, and sweep gas 2 AU.

Note: Source conditions are a recommended starting point, and should be optimized by the end user to ensure spray stability (reduced TIC variation).

5. Perform full scan MS data acquisitions with a resolution setting of 70,000 (at m/z 200) over an m/z range of 700-2000.
6. Use product ion data (triggered data dependant MS/MS scans) for sequence verification.
7. Process all data using Pinpoint™ software (version 1.3). For each of the insulin variants, extract the six most abundant isotopes for each of the three most abundant precursor charge states (each with a mass tolerance ± 7 ppm) from the full scan MS data (i.e. the cumulative intensity of 18 extracted ions). Normalize the cumulative peak intensities for each insulin analogue to the cumulative peak intensities for the insulin internal standard (4[D10] Leu Insulin).

Additional Information

Visit the www.thermoscientific.com/msia for additional information and application notes relating to this product.

Product stable for 12 months from date of manufacture when handled and stored according to Manufacturer instructions, see details under **Warranty**.

The most current versions of all MSIA product instructions are available at www.thermoscientific.com/msia.

Ordering Information

MSIA D.A.R.T'S for Immunoaffinity Capture		
Compatible with the Thermo Scientific Versette Automated Liquid Handler and Thermo Scientific Finnpiptette Novus i 12-Channel Electronic Pipette.		
Cat. No.	Description	Packaging
991STR11	MSIA Streptavidin D.A.R.T.'S	Pack of 96 tips
991STR12	MSIA Streptavidin D.A.R.T.'S	Pack of 24 tips
991PRT11	300 µl MSIA D.A.R.T.'S, Protein A	Pack of 96 tips
991PRT12	300 µl MSIA D.A.R.T.'S, Protein A	Pack of 24 tips
991PRT13	300 µl MSIA D.A.R.T.'S, Protein G	Pack of 96 tips
991PRT14	300 µl MSIA D.A.R.T.'S, Protein G	Pack of 24 tips
991PRT15	300 µl MSIA D.A.R.T.'S, Protein A/G	Pack of 96 tips
991PRT16	300 µl MSIA D.A.R.T.'S, Protein A/G	Pack of 24 tips
991CUS02	300 µl MSIA D.A.R.T.'S, Custom*	Pack of 96 tips
991R	300 µl MSIA D.A.R.T.'S, Reloadable Rack	1 reloadable rack, tips are not included
Automated Liquid Handling Platform		
Cat. No.	Description	
650-01-BS	Versette Automated Liquid Handler*	
Multichannel Pipettes and Pipette Stand		
Cat. No.	Description	Quantity
991SP12	Finnpiptette Novus i Electronic 12-Channel Pipette and Adjustable Pipette Stand	1 pipette and 1 pipette stand

*Base unit only. Additional parts required.

Warranty: Sellers warrants that the Products will operate or perform substantially in conformance with Seller's published specifications and be free from defects in material and workmanship, when subjected to normal, proper and intended usage by properly trained personnel, for the period of time set forth in the product documentation, published specifications or page insert. If a period of time is not specified in Seller's product documentation, published specifications or package inserts, the warranty period shall be one (1) year from the date of shipment to Buyer for equipment and ninety (90) days for all other products (the "Warranty Period"). Seller agrees the Warranty Period, to repair or replace at Seller's option, defective Products so as to cause the same to operate in substantial conformance with said published specifications, provided that Buyer shall (a) promptly notify Seller in writing upon the discovery of any defect, which notice shall include the product model and serial number (if applicable) and details of the warranty claim, and (b) after Seller's review. Seller will provide Buyer with service data and/or a Return Material Authorization ("RMA"), which may include biohazard decontamination procedures and other product-specific handling instructions, then, if applicable, Buyer may return the effective Products to Seller with all costs prepaid by Buyer. Replacement parts may be new or refurbished at the election of Seller. All replaced parts shall become the property of Seller. Shipment to Buyer repaired or replacement Products shall be made in accordance with the Delivery provisions of the Seller's Term and Conditions of Sale. Consumables are expressly excluded this warranty. Notwithstanding the foregoing, Products supplied by Seller that are obtained by Seller from an original manufacturer or third party supplier are not warranted by Seller, but Seller agrees to assign to Buyer any warranty rights in such Product that Seller may have from the original manufacturer or third party supplier, to the extent such assignment is allowed by such original manufacturer or third party supplier. In no event shall Seller have any obligation to make repairs, replacements or corrections required, in whole or in part, as the result as (i)normal wear and tear, (ii)accident, disaster or event of force majeure, (iii)misuse, fault or negligence of or by Buyer, (iv)use of the Products in a manner for which they were not designed, (v)causes external to the Products such as, but not limited to, power failure or electrical power surges, (vi)improper storage and handling of the Products or (vii) use of the Products in combination with equipment or software not supplied by Seller. If Seller determines that Products for which Buyer has requested warranty services are not covered by the warranty hereunder. Buyer shall pay or reimburse Seller for all costs of investigating and responding to such request at Seller's then prevailing time and materials rates. If Seller provides repair services or replacement parts that are not covered by this warranty, Buyer shall pay Seller therefore at Seller's then prevailing time and materials rates. Any installation, maintenance, repair, service, relocations or alteration to or of, or other tampering with, the products preformed by any person or entity other than seller without sellers' prior written approval, or any use of replacement parts not supplied by seller, shall immediately void and cancel all warranties with respect to the affected products. The obligations created by this warranty statement to repair or replace a defective product shall be the sole remedy of buyer in the event of defective product except as expressly provided in this warranty statement, seller disclaims all other warranties, whether express or implied, oral or written, with respect to the products, including without limitations all implied warranties of merchantability or fitness for any particular purpose. Seller does not warrant that the products are error-free or will accomplish any particular result.

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