



Product Guide for
Ludger Permethylation Kit™
without Methyl Iodide (MeI)

Product # LT-PERMET-VP96

Ludger Document # LT-PERMET-VP96-guide-v1.0

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Specifications for LT-PERMET-VP96

Application	This kit is used for the permethylation of glycans. Permethylation stabilizes glycan sialic acids for MALDI mass spectrometry analysis, and aids linkage analysis studies. The kit can be used with glycans released from glycoproteins.
Description	This kit contains reagents for the permethylation of glycans. <i>Note: This kit does not include methyl iodide / Iodomethane (see shipping section for details)</i>
Number of Samples	Sufficient for up to 96 samples.
Amount of Sample	Up to 1 µg of released glycans.
Suitable Samples	Any unlabelled purified glycans, released from glycoprotein by either PNGaseF, PNGase A, beta-elimination or hydrazinolysis.
Storage	Store at 2-8°C.
Shipping	The product should be shipped between 2-8°C. <i>Note: Due to shipping restrictions of methyl iodide/iodomethane, we are unable to provide this component with the kit. Therefore, we recommend purchasing iodomethane with purity ≥99.0% (GC) from your local chemical provider. Iodomethane Synonym: methyl iodide CAS Number: 74-88-4</i>
Safety	For research use only. Not for human or drug use. Please read the Safety Data Sheet (SDS's) for all chemicals used. All processes involved permethylation reagents should be performed using appropriate personal safety protection – eyeglasses, chemically resistant gloves and where appropriate in a laboratory fume cupboard.

Kit Contents



The kit contains the following items:

Catalogue Id.	Item	Quantity
LT-PERMET-PLATE-96	96 well Permethylation Plate	1 Plate
LT-PERMET-DMSO-96	dimethyl sulfoxide (DMSO)	40 mL
LT-PERMET-DCM-96	dichloromethane (DCM)	60 mL
LT-PERMET-LID-96	96 well Plate Lid	1 Lid / Mat
LT-BALANCE-PLATE	96 well Balance Plate	1 Plate
N/A	EMPTY-BOTTLE-FOR-MEI	1 bottle

Note: The kit can be used to process 1-96 samples. The plate is scalable and can be used for any number of samples between 1 and 96. Store the unused kit reagents and the unused plate wells in the recommended storage conditions to enable their use for further sample preparation at a later date. Please use within the expiry date.

Additional Reagents and Equipment Required

- methyl iodide (MeI) with purity $\geq 99.0\%$ (GC) (synonym: iodomethane).
- Pure water: resistivity above 18 M Ω -cm, particle free ($>0.22 \mu\text{m}$), TOC <10 ppb.
- Plate shaker
- Reaction Eppendorf vials (1.5 or 2.0 ml) for final transfer of extracted sample (vials need to be chloroform or dichloromethane resistant otherwise use glass vials).
- pH indicator strips
- Centrifugal evaporator (e.g. Savant, HETO or similar).
- 70% methanol (MeOH)
- dichloromethane (DCM) (used to add to the balance plate)

Safety and Handling

Ensure that any glass, plastic ware or solvents used with this item are free of carbohydrates. Use powder-free glove for all sample handling procedures and avoid contamination with environmental carbohydrates.

Time Line for Procedure

The permethylation procedure for 96 samples takes approximately 5 to 6 hours when performed manually.

Procedure	Time
Step 1: Permethylation of samples:	
a. Preparation of glycans	As required
b. Addition of DMSO and sample to permethylation plate	15 minutes
c. Incubation	15 minutes
d. Addition of MeI to permethylation plate	15 minutes
e. Incubation	60 minutes
Step 2: Extraction of permethylated samples and storage	
a. Extraction of samples	3 - 4 hours
b. Dry permethylated samples	10 - 30 minutes
c. Addition of 70% MeOH for storage	10 -20 minutes

Method Part 1: Permethylation of glycans

a) Preparation of glycans

- Following N-glycan release, we would advise the user to enrich and purify the N-glycans prior to permethylation in order to increase the efficacy of the reaction. We recommend using the LC-PERMET-96 plate for this purpose. If required, this can be purchased separately from Ludger. After enrichment, the glycans need to be dried down before they can be permethylated. It is preferable for the dried glycans to be in 1.5 mL Eppendorf vials or similar. However, if you have used the LC-PERMET-96 plate for the enrichment and eluted the samples into a 96 well collection plate, the samples can be dried down in the collection plate ready for the next step.

b) Addition of DMSO and dried glycan sample to the permethylation plate

Note: Before you start the experiment please ensure the DMSO is completely defrosted. DMSO is a solid at 19°C.

- Dissolve each of the glycan samples with 300 μ L DMSO. Vortex and centrifuge these samples. Peel the clear seal from the permethylation plate and cut the overhang away. Add the first sample in DMSO to the first well of the permethylation plate and using the same pipette tip disturb the solid at the bottom

of the well. Repeat this procedure with the remaining samples and record the sample positions.

c) Incubation

- Cover the plate with the 96 well silicone plate lid provided, place the permethylation plate on a plate shaker at ~100-150 rpm and incubate for 15 minutes at room temperature.

Note: Cut strips of the silicone 96 well lid / mat to cover the wells containing samples, making sure to cover the sample wells securely.

- After the 15 minute incubation, briefly centrifuge the sample plate to ensure that all the samples are at the bottom of the wells of the plate.

Note: An empty balance plate (LT-BALANCE-PLATE) has been provided as a part of the kit. Ensure the balance plate is appropriately balanced, by adding water (equal weight by volume), in comparison to the sample plate containing DMSO.

d) Addition of Mel to permethylation plate

- Carefully remove the silicone plate lid, to avoid cross contamination of samples, and add 55 μ L of Mel to the wells containing samples. Cover the wells again with the silicone plate lid orientated such that it is positioned to cover the same sample wells as before and seal securely.

e) Incubation

- Make sure the silicone lid is tightly sealed to eliminate the loss of volatile Mel.
- Securely position the permethylation plate on a plate shaker at 100-150 rpm, taking care to observe that there is no spillage or cross contamination of sample contents across the 1.2 mL permethylation plate. Incubate the plate at room temperature for 60 minutes
- After the 60 minute incubation, briefly centrifuge the sample plate to ensure that all the samples are at the bottom of the wells of the plate.

Note: An empty balance plate (LT-BALANCE-PLATE) has been provided as a part of the kit. Ensure the balance plate is appropriately balanced by adding water (equal weight in volume), in comparison to the sample plate containing DMSO and Mel.

Method Part 2: Extraction of permethylated glycans

a) Extraction of permethylated glycans

- Remove the silicon plate lid and add 450 μ L of DCM followed by ~500 μ L of water to each sample well in the permethylation plate.
- For the ease of the extraction steps, transfer the entire contents of each well containing sample (DMSO, DCM and water) to labelled 1.5/2.0 mL Eppendorf vials or glass vials.
- Once the samples are transferred, mix both the organic DCM layer and the aqueous layer by vortexing

each sample. Place the vials in a rack and allow the two layers to separate.

- Discard the top aqueous layer into a chlorinated waste container.
- Add 800 μL of water to each sample to wash the organic layer again. Vortex each sample to ensure good solvent mixing and allow the two layers to separate.
- Discard the top aqueous layer into a chlorinated waste container.
- Repeat the water wash step with further 800 μL amounts of water and test with pH paper until the aqueous layer is no longer basic (if > 7 , repeat with further water washes until the aqueous layer has a $\text{pH} \leq 7$).

b) Dry the permethylated glycans

- Make sure to completely remove the top aqueous layer that is present in each sample vial and discard into a chlorinated waste container. The permethylated glycans are present in the organic, DCM layer. Dry the organic solvent containing the permethylated samples in a centrifugal evaporator.

Note: If the liquid-liquid extraction was performed in a 96-well plate format, use the empty balance plate provided for balancing the centrifugal evaporator. Ensure the balance plate is appropriately balanced, by adding DCM or chloroform (equal weight in volume), in comparison to the sample plate containing permethylated glycans in DCM.

Note: Once the sample plate and balance plate are placed in the centrifugal evaporator, spin the plates for 2 minutes without vacuum to allow the DCM to settle. After 2 minutes, the vacuum pump can be switched on to dry the permethylated glycans.

c) Store the samples

- Add 10 μL of 70% MeOH to each sample vial with the dried down permethylated glycan and mix by vortexing. The samples may be stored at temperatures of -20°C before mass spectrometric analysis.

Warranties and liabilities

Ludger warrants that the above product conforms to the attached analytical documents. Should the product fail for reasons other than through misuse Ludger will, at its option, replace free of charge or refund the purchase price. This warranty is exclusive and Ludger makes no other warrants, expressed or implied, including any implied conditions or warranties of merchantability or fitness for any particular purpose.

Ludger shall not be liable for any incidental, consequential or contingent damages.

This product is intended for in vitro research only.

Document Revision Number

Document # LT-PERMET-VP96-guide-v1.0

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Appendix

Safety data sheet - Sodium hydroxide

Version: 1.0

 Date written: 07th October 2015

SECTION 1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND OF THE COMPANY /UNDERTAKING

Product Name	96 well permethylation plate containing sodium hydroxide
Product Catalogue Name	LT-PERMET-PLATE-96
CAS-No.	1310-73-2
Company:	Ludger Ltd Culham Science Centre Abingdon Oxfordshire OX14 3EB
Telephone:	01865 408554
Emergency Telephone:	01865 408554
Email:	info@ludger.com

SECTION 2. HAZARDS IDENTIFICATION

2.1 Classification of the substance or mixture

Classification according to Regulation (EC) No 1272/2008 [EU-GHS/CLP]

Corrosive to metals (Category 1), H290 Skin corrosion (Category 1A), H314
For the full text of the H-Statements mentioned in this Section, see Section 16.

2.2 Label elements



Signal Word: Danger

Hazard Statement(s)

Hazard statement(s)

H290 May be corrosive to metals.

H314 Causes severe skin burns and eye damage.

Precautionary statement(s)

P280 Wear protective gloves/ protective clothing/ eye protection/ face protection.

P303 + P361 + P353 IF ON SKIN (or hair): Take off immediately all contaminated clothing.

Rinse skin with water/shower.

P304 + P340 + P310 IF INHALED: Remove person to fresh air and keep comfortable for breathing. Immediately call a POISON CENTER or doctor/ physician.

P305 + P351 + P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

Supplemental Hazard Statements none

Precautionary Statement(s)

None

2.3 Other hazard information:

This substance/mixture contains no components considered to be either persistent, bioaccumulative and toxic (PBT), or very persistent and very bioaccumulative (vPvB) at levels of 0.1% or higher. section.

SECTION 3. COMPOSITION/INFORMATION ON INGREDIENTS

3.1 Substances

Synonyms: caustic soda
 Formula: NaOH
 Molecular Weight: 40.00 g/mol

Component	Concentration
Name sodium hydroxide	100 %
CAS-No. 1310-73-2	
EC-No. 215-185-5	

SECTION 4. FIRST AID MEASURES

4.1 Description of first aid measures

General Advice

Consult a physician if exposure causes ill effects and if in any doubt. Show this safety data sheet to the physician/ first responder in attendance.

If Ingested

Do NOT induce vomiting. Never give anything by mouth to an unconscious person. Rinse mouth with water. Consult a physician.

If skin is exposed

Take off contaminated clothing and shoes immediately. Wash off with soap and plenty of water. Consult a physician.

If eyes are exposed

Rinse thoroughly with plenty of water for at least 15 minutes and consult a physician.

If inhaled

If breathed in, move person into fresh air. If not breathing, give artificial respiration. Consult a physician.

4.2 Most important symptoms and effects, both acute and delayed

The most important known symptoms and effects are described in the labelling (see section 2.2) and/or in section 11

4.3 Indication of immediate medical attention and special treatment needed

No data available.

SECTION 5. FIRE-FIGHTING MEASURES

5.1 Extinguishing media

Use water spray, alcohol-resistant foam, dry chemical or carbon dioxide.

5.2 Special hazards arising from the substance or mixture

Sodium oxides

5.3 Advice for firefighters

Wear self-contained breathing apparatus for firefighting if necessary.

SECTION 6. ACCIDENTAL RELEASE MEASURES

6.1 Personal precautions, protective equipment and emergency procedures

Wear respiratory protection. Avoid breathing vapours, mist or gas. Ensure adequate ventilation.

Evacuate personnel to safe areas.

For personal protection see section 8.

6.2 Environmental Precautions

Prevent further leakage or spillage if safe to do so. Do not let product enter drains. Discharge into the environment must be avoided.

6.3 Methods and material for containment and cleaning up

6.3 Methods and materials for containment and cleaning up

Soak up with inert absorbent material and dispose of as hazardous waste. Keep in suitable, closed containers for disposal.

6.4 Reference to other sections

For disposal see section 13.

SECTION 7. HANDLING AND STORAGE

7.1 Precautions for safe handling

Avoid inhalation of vapour or mist. For precautions see section 2.2.

7.2 Conditions for safe storage, including any incompatibilities

Store in cool place. Keep container tightly closed in a dry and well-ventilated place. Containers which are opened must be carefully resealed and kept upright to prevent leakage.

Storage class (TRGS 510): Non-combustible, corrosive hazardous materials

7.3 Specific end uses

Apart from the uses mentioned in section 1.2 no other specific uses are stipulated

SECTION 8. EXPOSURE CONTROLS/PERSONAL PROTECTION

8.1 Control parameters

Component	CAS-No.	ValueForm of exposure	Control parameters	Basis
Sodium hydroxide	1310-73-2	STEL	2 mg/m ³	UK. EH40 WEL - Workplace Exposure Limits

8.2 Exposure controls

Appropriate engineering controls

Handle in accordance with good industrial hygiene and safety practice. Wash hands before breaks and at the end of workday.

Personal Protective Equipment

Eye / face protection

Tightly fitting safety goggles. Faceshield (8-inch minimum). Use equipment for eye protection tested and approved under appropriate government standards such as NIOSH (US) or EN 166(EU).

Skin protection

Handle with gloves. Gloves must be inspected prior to use. Use proper glove removal technique (without touching glove's outer surface) to avoid skin contact with this product. Dispose of contaminated gloves after use in accordance with applicable laws and good laboratory practices. Wash and dry hands.

If used in solution, or mixed with other substances, and under conditions which differ from EN 374, contact the supplier of the CE approved gloves. This recommendation is advisory only and must be evaluated by an industrial hygienist and safety officer familiar with the specific situation of anticipated use by our customers. It should not be construed as offering an approval for any specific use scenario.

Body Protection

Complete suit protecting against chemicals, The type of protective equipment must be selected according to the concentration and amount of the dangerous substance at the specific workplace.

Respiratory protection

Where risk assessment shows air-purifying respirators are appropriate use a full-face respirator with multi-purpose combination (US) or type ABEK (EN 14387) respirator cartridges as a backup to engineering controls. If the respirator is the sole means of protection, use a full-face supplied air respirator. Use respirators and components tested and approved under appropriate government standards such as NIOSH (US) or CEN (EU).

Thermal hazards

No information available.

SECTION 9. PHYSICAL AND CHEMICAL PROPERTIES

9.1 Information on basic physical and chemical properties

Appearance	Form: liquid, Colour: colourless
Odour threshold	No data available
pH	No data available
Freezing/Melting Point	-12-10°C
Initial boiling point and boiling range	105 - 140 °C
Flash Point	Not applicable
Evaporation rate	No data available
Flammability	No data available
Upper/lower flammability or explosive limits	No data available
Vapour Pressure, Pa at temperature degree C	< 24 hPa at 20 °C
Relative Density	1.515 g/mL at 25 °C
Solubility in water and solvents (mg/l)	completely miscible, soluble
Partition coefficient	No data available
Autoignition temperature	No data available
Decomposition temperature	No data available
Viscosity	No data available
Explosive properties	No data available
Oxidising properties	No data available

9.2 Other information

Relative vapour density 1.38 - (Air = 1.0)

SECTION 10. STABILITY AND REACTIVITY**10.1 Reactivity**

No data available

10.2 Chemical stability

Stable under recommended storage conditions.

10.3 Possibility of hazardous reactions

No data available

10.4 Conditions to Avoid

No data available

10.5 Incompatible materials

Water, acids, Organic materials, Chlorinated solvents, Aluminium, Phosphorus, Tin/tin oxides, Zinc.

10.6 Hazardous decomposition products

Other decomposition products - No data available In the event of fire: see section 5

SECTION 11. TOXICOLOGICAL INFORMATION**11.1 Information on toxicological effects****Acute toxicity**

No data available

Skin corrosion/irritation

No data available.

Serious eye damage/irritation

No data available

Respiratory or skin sensitisation

No data available

Germ cell mutagenicity

No data available

Reproductive toxicity

No data available

STOT-single exposure

No data available

STOT-repeated exposure

No data available

Aspiration hazard.

No data available

Potential Health Hazards

Corrosive and causes severe burns.

Signs and symptoms of exposure

Burning sensation, Cough, wheezing, laryngitis, Shortness of breath, spasm, inflammation and edema of the larynx, spasm, inflammation and edema of the bronchi, pneumonitis, pulmonary edema, Material is extremely destructive to tissue of the mucous membranes and upper respiratory tract, eyes, and skin.

SECTION 12. ECOLOGICAL INFORMATION

12.1 Toxicity

Toxicity to fish LC50 Gambusia affinis (Mosquito fish) - 125 mg/l - 96 h
 Toxicity to daphnia and
 other aquatic invertebrates Immobilization EC50 - Daphnia - 40.38 mg/l - 48 h

12.2 Persistence and degradability

No data available

12.3 Bioaccumulative potential

No data available

12.4. Mobility in soil

No data available

12.5. Results of PBT and vPvB assessment

PBT/vPvB assessment not available as chemical safety assessment not required/not conducted

12.6. Other adverse effects

Harmful to aquatic life.

SECTION 13. DISPOSAL CONSIDERATIONS

13.1 Waste treatment methods

Offer surplus and non-recyclable solutions to a licensed disposal company.

Contaminated packaging

Dispose of as unused product.

SECTION 14. TRANSPORT INFORMATION

14.1 UN Number

ADR/RID: 1824 IMDG: - IATA: 1824

14.2 UN Proper Shipping Name

ADR/RID: sodium hydroxide solid
 IMDG: sodium hydroxide solid
 IATA: sodium hydroxide solid

14.3 Transport hazard class(es)

ADR/RID: 8 IMDG: 8 IATA: 8

14.4 Packing group

ADR/RID: II IMDG: II IATA:II

14.5 Environmental hazards

ADR/RID: No IMDG: marine pollutant: No IATA: No

14.6 Special precautions for user

No data available

SECTION 15. REGULATORY INFORMATION**15.1. Safety, health and environmental regulations/legislation specific for the substance or mixture**

No data available.

15.2 Chemical Safety Assessment

For this product a chemical safety assessment was not carried out.

SECTION 16. OTHER INFORMATION

Full text of H-Statements referred to under sections 2 and 3.

H290	May be corrosive to metals.
H314	Causes severe skin burns and eye damage.
Met. Corr.	Corrosive to metals
Skin Corr.	Skin corrosion

The advice offered is derived from the current available information on the hazardous materials in this product and its component(s). Consideration has been made regarding the quantities offered in the pre-dispensed container. The advice offered is, therefore, not all-inclusive nor should it be taken as the descriptive of the compound generally.

Safety data sheet – dimethyl sulfoxide

Version: 1.1

 Date written: 07th October 2011

 Date reviewed: 9th February 2016

SECTION 1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND OF THE COMPANY / UNDERTAKING

Product Name	dimethyl sulfoxide
Product Catalogue Name	LT-PERMET-DMSO-96
CAS-No.	67-68-5
Company:	Ludger Ltd Culham Science Centre Abingdon Oxford OX14 3EB
Telephone:	01865 408554
Emergency Telephone:	01865 408554
Email:	info@ludger.com

SECTION 2. HAZARDS IDENTIFICATION

2.1 Classification of the substance or mixture

Classification according to Regulation (EC) No. 1272/2008 [EU-GHS-CLP]

Not a hazardous substance or mixture according to Regulation (EC) No. 1272/2008 [EU-GHS-CLP].

2.2 Label elements

The product does not require to be labelled in accordance with EC directives or respective national laws.

Signal Word: None

Hazard Statement(s)

None

Precautionary Statement(s)

None

2.3 Other hazard information:

None

SECTION 3. COMPOSITION/INFORMATION ON INGREDIENTS

3.1 Substances

Synonyms:	DMSO methyl sulfoxide dimethyl sulfoxide
Formula:	C ₂ H ₆ OS
Molecular Weight:	78.13g/mol

Component		Concentration
Name	Dimethyl Sulfoxide	-
CAS-No.	67-68-5	
EC-No.	200-664-3	

SECTION 4. FIRST AID MEASURES

4.1 Description of first aid measures

General Advice

Consult a physician if exposure causes ill effects and if in any doubt. Show this safety data sheet to the physician/ first responder in attendance.

If Ingested

Do NOT induce vomiting. Rinse mouth well with water. Never give anything by mouth to an unconscious person.

If skin is exposed

Wash off with plenty of soap and water.

If eyes are exposed

Flush eyes with plenty of water/ eye wash solution as a precaution.

If inhaled

Move affected person to fresh air. If not breathing give artificial respiration.

4.2 Most important symptoms and effects, both acute and delayed

Effects due to ingestion may include: Nausea, Fatigue and Headache.

4.3 Indication of immediate medical attention and special treatment needed

No data available.

SECTION 5. FIRE-FIGHTING MEASURES

5.1 Extinguishing media

Small fires: Use extinguishing media such as "alcohol" foam, dry chemical or carbon dioxide.

Large fires: Use extinguishing media such as water, from a far away distance as possible. Use very large quantities of water as mist or spray to flood the fire and the combustible material. Cool all affected containers with large quantities of water.

5.2 Special hazards arising from the substance or mixture

Carbon oxides, Sulphur oxides

5.3 Advice for fire fighters

Wear self contained breathing apparatus for fire fighting if necessary, to spray cool water on any unopened containers near the fire.

SECTION 6. ACCIDENTAL RELEASE MEASURES

6.1 Personal precautions, protective equipment and emergency procedures

Avoid breathing vapours, gas or mist. Remove all sources of ignition. Beware of vapours accumulating to form explosive concentrations. Vapours can accumulate in low areas.

6.4 Environmental Precautions

Prevent further leakage or spillage if safe to do so, e.g. with spill mats. Do not let the product enter drains.

6.5 Methods and material for containment and cleaning up

Contain the spillage and put the collected material into a suitable container with a secure lid. Wash the area well, do not let run off into the drains, collect as waste.

6.4 Reference to other sections

See section 13 for disposal of waste material(s).

SECTION 7. HANDLING AND STORAGE

7.1 Precautions for safe handling

Avoid inhalation of vapour or mist. Keep away from sources of ignition- No smoking.
Take measures to prevent the build up of electrostatic charge.

7.2 Conditions for safe storage, including any incompatibilities

Store in a cool place. Keep container closed in a dry well ventilated place.

7.3 Specific end uses

No data available

SECTION 8. EXPOSURE CONTROLS/PERSONAL PROTECTION

8.1 Control parameters

Components with workplace control parameters.

Contains no substances with occupational exposure limit values.

8.3 Exposure controls

Appropriate engineering controls

Handle in accordance with good laboratory hygiene and safety practice. Wash hands before breaks and at the end of the day.

Personal Protective Equipment

Eye / face protection

Use equipment for eye protection tested and approved under appropriate government standards such as NIOSH (US) or EN 166 (EU).

Skin protection

Handle with gloves, which should be inspected before use. Use proper glove removal technique (removal without the outside of the glove touching the skin) to avoid contact with the skin/chemical. Dispose of contaminated gloves as Laboratory waste in accordance with applicable laws and good laboratory practices. Wash and dry hands.

Gloves should be of the standard that will stratify the specifications of EU directive 89/696/EEC and the standard EN 374 derived from it.

Body Protection

The type of protective clothing must be selected according to the amount of substance at the specific workplace being used. Impervious coats or laboratory coats.

Respiratory protection

Use substance in an operation fume hood/ outside venting extraction cupboard. Wear full face respirator if appropriate to use, must be tested and approved under appropriate government standards such as NIOSH (US) or CEN (EU).

SECTION 9. PHYSICAL AND CHEMICAL PROPERTIES

9.1 Information on basic physical and chemical properties

Appearance	Form: Liquid, clear
	Colour: Colourless
Odour	No data available
Odour threshold	No data available
pH	No data available
Freezing/Melting Point	Melting point/range: 16-19°C
Initial boiling point and boiling range	189°C
Flash Point	87°C – Closed cup
Evaporation rate	No data available
Flammability	No data available
Upper/lower flammability or explosive limits	Upper explosion limit: 42% (V)
	Lower explosion limit: 3.5% (V)
Vapour Pressure, Pa at temperature degree C	0.55hPa at 20°C

Relative Density	1.1g/mL
Solubility in water and solvents	Completely miscible
Partition coefficient: n-octanol/water	log Pow: - 2.03
Auto ignition temperature	No data available
Decomposition temperature	No data available
Viscosity	No data available
Explosive properties	No data available
Oxidising properties	No data available

9.2 Other information

No data available

SECTION 10. STABILITY AND REACTIVITY

10.1 Reactivity

No data available

10.2 Chemical stability

No data available

10.3 Possibility of hazardous reactions

No data available

10.4 Conditions to Avoid

Heat, flames and sparks

10.5 Incompatible materials

Acid chlorides, Phosphorus halides, Strong acids, Strong oxidizing agents and strong reducing agents.

10.6 Hazardous decomposition products

Other decomposition products – No data available

SECTION 11. TOXICOLOGICAL INFORMATION

11.1 Information on toxicological effects

Acute toxicity

LD50 Oral – Rat – 14,500mg/kg

LC50 Inhalation – Rat – 4h – 40250ppm

LD50 Dermal – Rabbit - > 5,000mg/kg

Skin corrosion/irritation

Skin – Rabbit – No skin irritation – 4h

Serious eye damage/irritation

Eyes – Rabbit – Mild eye irritation

Respiratory or skin sensitisation

No data available

Germ cell mutagenicity

Genotoxicity in vitro – Mouse – lymphocyte

Cytogenetic analysis

Genotoxicity in vitro – Mouse – lymphocyte

Mutation in mammalian somatic cells

Genotoxicity in vivo – Rat – Intraperitoneal

Cytogenetic analysis

Genotoxicity in vivo - Mouse – Intraperitoneal

DNA damage

Carcinogenicity

Carcinogenicity – Rat – Oral

Tumorigenic: Equivocal tumorigenic agent by RTECS criteria. Skin and Appendages: Others: Tumors.

Carcinogenicity – Mouse – Oral

Tumorigenic: Equivocal tumorigenic agent by RTECS criteria. Lukaemia skin and appendages: Other: Tumors.

IARC: No component of this product presents at levels greater than or equal to 0.1% is identified as probable, possible or confirmed human carcinogen by IARC.

Reproductive toxicity

Reproductive toxicity – Rat – Intraperitoneal

Effects on fertility: Abortion

Reproductive toxicity – Rat – Intraperitoneal

Effects on fertility: Post – implantation mortality (e.g. dead and/or resorbed implants per total number of implants).

Reproductive toxicity – Rat – Subcutaneous

Effects on fertility: Post – implantation mortality (e.g. dead and/or resorbed implants per total number of implants). Effects on fertility: Litter size (e.g. # fetuses per litter; measured before birth).

Reproductive toxicity – Mouse – Oral

Effects on fertility: Pre-implantation mortality (e.g. reduction in number of implants per female; total number of implants per corpora lutea). Effects on Embryo or fetus: Fetotoxicity (except death, e.g. stunted fetus). Specific developmental abnormalities: Musculoskeletal system.

Reproductive toxicity – Mouse – Intraperitoneal

Effects on embryo or fetus: Fetotoxicity (except death, e.g. stunted fetus). Specific developmental abnormalities: Musculoskeletal system.

STOT-single exposure

No data available

STOT-repeated exposure

No data available

Aspiration hazard.

No data available

Potential Health Hazards

Inhalation

May be harmful if inhaled. May cause respiratory tract irritation.

Ingestion

May be harmful if swallowed.

Skin

May be harmful if absorbed through skin. May cause skin irritation.

Eyes

May cause eye irritation.

Aggravated Medical Condition

Avoid contact with DMSO solutions containing toxic materials or materials with unknown toxicological properties. Dimethyl sulfoxide is readily absorbed through the skin and may carry such materials into the body.

Signs and symptoms of exposure

Effects due to ingestion may include; Nausea, Fatigue, Headache.

Additional Information

RTECS: PV6210000

SECTION 12. ECOLOGICAL INFORMATION

12.1 Toxicity

Toxicity to Fish 96h	LC50-Pimephales promelas (fathead minnow) – 34,000mg/l -
Toxicity to daphnia and other Aquatic invertebrates	LC50-Oncorhynchus mykiss (rainbow trout) – 34,000mg/l-96h
Toxicity to algae	EC50-Daphnia pulex (water fleas) – 27,500mg/l EC50-Lepomis macrochirus (bluegill) - >400,000mg/l-96h

12.2 Persistence and degradability

No data available

12.3 Bioaccumulative potential

No data available

12.4. Mobility in soil

No data available

12.5. Results of PBT and vPvB assessment

No data available

12.6. Other adverse effects

No data available

SECTION 13. DISPOSAL CONSIDERATIONS

13.1 Waste treatment methods

This combustible material may be burned in a chemical incinerator equipped with an afterburner and scrubber or to be disposed of by a licensed professional waste disposal company.

Contaminated packaging

Dispose of as the unused product.

SECTION 14. TRANSPORT INFORMATION

14.1 UN Number

ADR/RID: -	IMDG: -	IATA: -
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14.2 UN Proper Shipping Name

ADR/RID:	Not Dangerous Goods
IMDG:	Not Dangerous Goods
IATA:	Not Dangerous Goods

14.3 Transport hazard class (es)

ADR/RID: -	IMDG: -	IATA: -
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14.4 Packing group

ADR/RID: -	IMDG: -	IATA: -
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14.5 Environmental hazards

ADR/RID: No	IMDG Marine pollutant: No	IATA: No
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14.6 Special precautions for user

No data available

SECTION 15. REGULATORY INFORMATION

This safety data sheet complies with the requirements of Regulation (EC) No. 1907/2006

15.1. Safety, health and environmental regulations/legislation specific for the substance or mixture

No data available

15.2 Chemical Safety Assessment

No data available

Please note that the label elements that used to go in Section 15 are now in Section 2.

SECTION 16. OTHER INFORMATION

The advice offered is derived from the current available information on the hazardous materials in this product and its component(s). Consideration has been made regarding the quantities offered in the pre-dispensed container. The advice offered is, therefore, not all-inclusive nor should it be taken as the descriptive of the compound generally.

SAFETY DATA SHEET - dichloromethane

Version: 1.0

Date written: 07th October 2015**SECTION 1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND OF THE COMPANY****/ UNDERTAKING**

Product Name **dichloromethane**

Product Catalogue Name **LT-PERMET-DCM-96**

CAS-No. 75-09-2

Company: Ludger Ltd
 Culham Science Centre
 Abingdon
 Oxfordshire
 OX14 3EB

Telephone: 01865 408554

Emergency Telephone: 01865 408554

Email: info@ludger.com

SECTION 2. HAZARDS IDENTIFICATION**2.1 Classification of the substance or mixture****Classification according to Regulation (EC) No 1272/2008 [EU-GHS/CLP]**

Skin irritation (Category 2), H315 Eye irritation (Category 2), H319 Carcinogenicity (Category 2), H351

Specific target organ toxicity - single exposure (Category 3), Central nervous system, H336

Specific target organ toxicity - single exposure (Category 3), Respiratory system, H335

Specific target organ toxicity - repeated exposure (Category 2), Liver, Blood, Central nervous system, H373

For the full text of the H-Statements mentioned in this Section, see Section 16.

2.2 Label elements

Signal Word: Warning

Hazard Statement(s)

H315 Causes skin irritation.

H319 Causes serious eye irritation.

H335 May cause respiratory irritation.

H336 May cause drowsiness or dizziness.

H351 Suspected of causing cancer.

H373 May cause damage to organs (Liver, Blood, Central nervous system) through prolonged or repeated exposure.

Precautionary Statement(s)

P261 Avoid breathing vapours.

P281 Use personal protective equipment as required.

P305 + P351 + P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Supplemental Hazard Statements none

2.3 Other hazard information:

This substance/mixture contains no components considered to be either persistent, bioaccumulative and toxic (PBT), or very persistent and very bioaccumulative (vPvB) at levels of 0.1% or higher.

SECTION 3. COMPOSITION/INFORMATION ON INGREDIENTS

3.1 Substances

Synonyms: methylene chloride (DCM)

Formula: CH₂Cl₂

Component		Concentration
Name	methylene chloride	
CAS-No.	75-09-2	
EC-No.	200-838-9	

For the full text of the H-Statements mentioned in this Section, see Section 16.

SECTION 4. FIRST AID MEASURES

4.1 Description of first aid measures

General Advice

Consult a physician. Show this safety data sheet to the doctor in attendance.

If Ingested

Never give anything by mouth to an unconscious person. Rinse mouth with water. Consult a physician.

If skin is exposed

Wash off with soap and plenty of water. Consult a physician.

If eyes are exposed

Rinse thoroughly with plenty of water for at least 15 minutes and consult a physician.

If inhaled

If breathed in, move person into fresh air. If not breathing, give artificial respiration. Consult a physician.

4.2 Most important symptoms and effects, both acute and delayed

The most important known symptoms and effects are described in the labelling (see section 2.2) and/or in section 11

4.3 Indication of immediate medical attention and special treatment needed

No data available

SECTION 5. FIRE-FIGHTING MEASURES

5.1 Extinguishing media

Use water spray, alcohol-resistant foam, dry chemical or carbon dioxide.

5.2 Special hazards arising from the substance or mixture

Carbon oxides, Hydrogen chloride gas

5.3 Advice for firefighters

Wear self-contained breathing apparatus for firefighting if necessary.

SECTION 6. ACCIDENTAL RELEASE MEASURES

6.1 Personal precautions, protective equipment and emergency procedures

Use personal protective equipment. Avoid breathing vapours, mist or gas. Ensure adequate ventilation. Evacuate personnel to safe areas.

For personal protection see section 8.

6.6 Environmental Precautions

Prevent further leakage or spillage if safe to do so. Do not let product enter drains.

6.7 Methods and material for containment and cleaning up

Soak up with inert absorbent material and dispose of as hazardous waste. Keep in suitable, closed containers for disposal.

6.4 Reference to other sections

For disposal see section 13.

SECTION 7. HANDLING AND STORAGE

7.1 Precautions for safe handling

Avoid contact with skin and eyes. Avoid inhalation of vapour or mist. For precautions see section 2.2.

7.2 Conditions for safe storage, including any incompatibilities

Store in cool place. Keep container tightly closed in a dry and well-ventilated place. Containers which are opened must be carefully resealed and kept upright to prevent leakage.

Heat sensitive. Store under inert gas.

Storage class (TRGS 510): Non-combustible, acute toxic Cat.3 / toxic hazardous materials or hazardous materials causing chronic effects

7.3 Specific end uses

Apart from the uses mentioned in section 1.2 no other specific uses are stipulated.

SECTION 8. EXPOSURE CONTROLS/PERSONAL PROTECTION

8.1 Control parameters

Component	CAS-No.	ValueForm of exposure	Control parameters	Basis
Methylene chloride	75-09-2	STEL	300 ppm 1,060 mg/m ³	UK. EH40 WEL - Workplace Exposure Limits
	Remarks	Can be absorbed through skin. The assigned substances are those for which there are concerns that dermal absorption will lead to systemic toxicity.		

8.4 Exposure controls

Appropriate engineering controls

Handle in accordance with good industrial hygiene and safety practice. Wash hands before breaks and at the end of workday.

Personal Protective Equipment

Eye / face protection

Face shield and safety glasses Use equipment for eye protection tested and approved under appropriate government standards such as NIOSH (US) or EN 166(EU).

Skin protection

Handle with gloves. Gloves must be inspected prior to use. Use proper glove removal technique (without touching glove's outer surface) to avoid skin contact with this product. Dispose of contaminated gloves after use in accordance with applicable laws and good laboratory practices. Wash and dry hands.

The selected protective gloves have to satisfy the specifications of EU Directive 89/686/EEC and the standard EN 374 derived from it.

If used in solution, or mixed with other substances, and under conditions which differ from EN 374, contact the supplier of the CE approved gloves. This recommendation is advisory only and must be evaluated by an industrial hygienist and safety officer familiar with the specific situation of anticipated use by our customers. It should not be construed as offering an approval for any specific use scenario.

Body Protection

Complete suit protecting against chemicals, The type of protective equipment must be selected according to the concentration and amount of the dangerous substance at the specific workplace.

Respiratory protection

Where risk assessment shows air-purifying respirators are appropriate use a full-face respirator with multi-purpose combination (US) or type AXBEK (EN 14387) respirator cartridges as a backup to engineering controls. If the respirator is the sole means of protection, use a full-face supplied air respirator. Use respirators and components tested and approved under appropriate government standards such as NIOSH (US) or CEN (EU).

Thermal hazards

No information available.

SECTION 9. PHYSICAL AND CHEMICAL PROPERTIES

9.2 Information on basic physical and chemical properties

Appearance	Form: liquid, Colour: colourless
Odour threshold	No data available
pH	No data available
Freezing/Melting Point	-97 °C
Initial boiling point and boiling range	39.8 - 40 °C
Flash Point	Not applicable
Evaporation rate	0.71
Flammability	No data available
Upper/lower flammability or explosive limits	Upper explosion limit: 19 %(V) Lower explosion limit: 12 %(V)
Vapour Pressure, Pa at temperature degree C	470.9 hPa at 20.0 °C
Relative Density	1.325 g/mL at 25 °C

Solubility in water and solvents (mg/l)	slightly soluble
Partition coefficient	log Pow: 1.25
Autoignition temperature	556.1 °C 662.0 °C
Decomposition temperature	No data available
Viscosity	No data available
Explosive properties	No data available
Oxidising properties	No data available

9.2 Other information

Relative vapour density 2.93 - (Air = 1.0)

SECTION 10. STABILITY AND REACTIVITY

10.1 Reactivity

No data available

10.2 Chemical stability

Stable under recommended storage conditions.

Contains the following stabiliser(s): 2-Methyl-2-butene (>0.005 - <0.015 %)

10.3 Possibility of hazardous reactions

No data available

10.4 Conditions to Avoid

Heat, flames and sparks. Exposure to sunlight.

10.5 Incompatible materials

Alkali metals, Aluminum, Strong oxidizing agents, Bases, Amines, Magnesium, Strong acids and strong bases, Vinyl compounds

10.6 Hazardous decomposition products

Other decomposition products - No data available In the event of fire: see section 5.

SECTION 11. TOXICOLOGICAL INFORMATION

11.1 Information on toxicological effects

Acute toxicity

LD50 Oral - Rat - > 2,000 mg/kg

LC50 Inhalation - Rat - 52,000 mg/m³

LD50 Dermal - Rat - > 2,000 mg/kg (OECD Test Guideline 402)

Skin corrosion/irritation

Skin - Rabbit

Result: Irritating to skin. - 24 h (Draize Test)

Serious eye damage/irritation

Eyes - Rabbit

Result: Irritating to eyes. - 24 h (Draize Test)

Respiratory or skin sensitisation

No data available

Germ cell mutagenicity

Rat
DNA damage

Reproductive toxicity

No data available

STOT-single exposure

May cause respiratory irritation. May cause drowsiness or dizziness

STOT-repeated exposure

Inhalation - May cause damage to organs through prolonged or repeated exposure. - Central nervous system

Oral - May cause damage to organs through prolonged or repeated exposure. - Liver, Blood

Aspiration hazard.

No data available

Potential Health Hazards

Corrosive and causes severe burns.

Signs and symptoms of exposure

Dichloromethane is metabolized in the body producing carbon monoxide which increases and sustains carboxyhemoglobin levels in the blood, reducing the oxygen-carrying capacity of the blood., Acts as a simple asphyxiant by displacing air., anesthetic effects, Difficulty in breathing, Headache, Dizziness, Prolonged or repeated contact with skin may cause:, defatting, Dermatitis, Contact with eyes can cause:, Redness, Blurred vision, Provokes tears., Effects due to ingestion may include:, Gastrointestinal discomfort, Central nervous system depression, Paresthesia., Drowsiness, Convulsions, Conjunctivitis., Pulmonary edema. Effects may be delayed., Irregular breathing., Stomach/intestinal disorders, Nausea, Vomiting, Increased liver enzymes., Weakness, Heavy or prolonged skin exposure may result in the absorption of harmful amounts of material., Abdominal pain.

SECTION 12. ECOLOGICAL INFORMATION**12.1 Toxicity**

Toxicity to fish LC50 - Pimephales promelas (fathead minnow) - 193.00 mg/l - 96 h NOEC -
Cyprinodon variegatus (sheepshead minnow) - 130 mg/l - 96 h

Toxicity to daphnia and other aquatic invertebrates.

12.2 Persistence and degradability

Biodegradability Result: < 26 % - Not readily biodegradable.
(OECD Test Guideline 301C)

12.3 Bioaccumulative potential

Does not bioaccumulate.

12.4. Mobility in soil

No data available

12.5. Results of PBT and vPvB assessment

This substance/mixture contains no components considered to be either persistent, bioaccumulative and toxic (PBT), or very persistent and very bioaccumulative (vPvB) at levels of 0.1% or higher.

12.6. Other adverse effects

No data available

SECTION 13. DISPOSAL CONSIDERATIONS

13.1 Waste treatment methods

Product

Offer surplus and non-recyclable solutions to a licensed disposal company.

Contaminated packaging

Dispose of as unused product.

SECTION 14. TRANSPORT INFORMATION

14.1 UN Number

ADR/RID: 1593

IMDG: -1593

IATA: 1593

14.2 UN Proper Shipping Name

ADR/RID:

DICHLOROMETHANE

IMDG:

DICHLOROMETHANE

IATA: Dichloromethane

14.3 Transport hazard class(es)

ADR/RID: 6.1

IMDG: 6.1

IATA: 6.1

14.4 Packing group

ADR/RID: III

IMDG: III

IATA: III

14.5 Environmental hazards

ADR/RID: No

IMDG: marine pollutant: No

IATA: No

14.6 Special precautions for user

No data available

SECTION 15. REGULATORY INFORMATION

15.1. Safety, health and environmental regulations/legislation specific for the substance or mixture

Methylene chloride CAS-No.: 75-09-2

REACH - Restrictions on the manufacture, placing on the market and use of certain dangerous substances, preparations and articles (Annex XVII)

Paint strippers containing dichloromethane in a concentration equal to or greater than 0,1 % by weight shall not be: (a) placed on the market for the first time for supply to the general public or to professionals after 6 December 2010; (b) placed on the market for supply to the general public or to professionals after 6 December 2011; (c) used by professionals after 6 June 2012.

See Commission Regulation (EU) No 276/2010 for Conditions of restriction.

15.2 Chemical Safety Assessment

A Chemical Safety Assessment has been carried out for this substance.

SECTION 16. OTHER INFORMATION

Full text of H-Statements referred to under sections 2 and 3.

- H315 Causes skin irritation.
- H319 Causes serious eye irritation.
- H335 May cause respiratory irritation.
- H336 May cause drowsiness or dizziness.
- H351 Suspected of causing cancer.
- H373 May cause damage to organs (/\$/*_ORGAN_REPEAT/\$/) through prolonged or repeated exposure.

The advice offered is derived from the current available information on the hazardous materials in this product and its component(s). Consideration has been made regarding the quantities offered in the pre-dispensed container. The advice offered is, therefore, not all-inclusive nor should it be taken as the descriptive of the compound generally.