



Product Guide for
LudgerTag™ Procainamide
Glycan Labeling Kit with
sodium cyanoborohydride



Product # LT-KPROC-24

Ludger Document # LT-KPROC-24-Guide-v1.0

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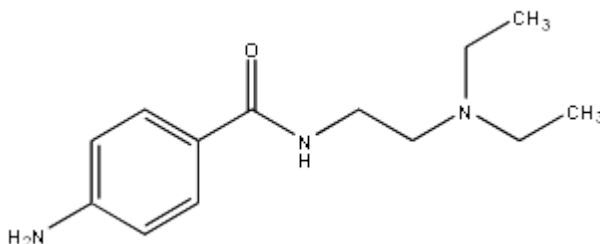
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Specifications for LT-KPROC-24

Application	For labeling of free glycans with procainamide (PROC).
Description	The kit contains reagents for the conjugation of dye to the free reducing end of the glycan by a reductive amination reaction.
Dye Properties	Mass free dye = 235.33. Fluorescence, $\lambda_{\text{ex}} = 310 \text{ nm}$, $\lambda_{\text{em}} = 370 \text{ nm}$.



Number of Samples	12 separate analytical samples per set of labeling reagents (24 samples in total for the kit)
Amount of Sample	From 25 pmol up to 25 nmol glycans per sample.
Suitable Samples	Any purified glycans with free reducing termini can be labeled.
Structural Integrity	No detectable (< 2 mole per cent) loss of sialic acid, fucose, sulfate, or phosphate.
Labeling Selectivity	Essentially stoichiometric labeling.
Storage:	Store at room temperature in the dark. Protect from sources of heat, light, and moisture. The reagents are stable for at least two years as supplied.
Shipping:	The product can be shipped at ambient temperature.
Handling:	Ensure that any glass, plasticware or solvents used are free of glycosidases and environmental carbohydrates. Use powder-free gloves for all sample handling procedures and avoid contamination with environmental carbohydrate. All steps involving labeling reagents must be performed in a dry environment with dry glassware and plasticware. Once individual vials of reagents are opened, their contents should be used immediately and excess then discarded according to local safety rules.
Safety:	For research use only. Not for human or drug use Please read the Safety Data Sheets (SDS's) for all chemicals used. All processes involving labeling reagents should be performed using appropriate personal safety protection - eyeglasses, chemically resistant gloves (e.g. nitrile), etc. - and where appropriate in a laboratory fume cupboard.

Kit Contents



Each kit contains two labeling reaction sets. Each labeling reaction set consists of one vial of each of the following:

Cat. #	Item	Quantity
LT-PROC-01	procainamide dye	16 mg
LT-CYANOB-05	sodium cyanoborohydride	9 mg
LT-ACETIC-DMSO-01	30% acetic acid in DMSO	500 μ L

Additional Reagents and Equipment Required

- Pure water
- Heating block, oven, or similar dry heater (a water bath cannot be used) set at 65°C
- Centrifugal evaporator (e.g. Savant, Heto, or similar)
- Reaction vials (e.g. polypropylene microcentrifuge vials)
- Note: Further consumables are required if doing the optional post-labeling sample cleanup (see Section on Sample Cleanup)

Time Line for Labeling

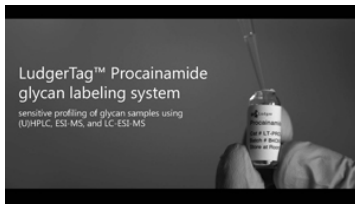
The LudgerTag™ labeling procedure takes 2 hours with just 1 hour for the actual labeling incubation.

Procedure	Time	Elapsed Time (hours)
Transfer samples to reaction tube and dry	45 min	0.75
Make up and add labeling reagent	15 min	1
Incubate samples with reagent	1 hour	2

Printing Out Method Instructions For Your Lab

- If you would like to print out this method for your lab then please print out pages 5, 6 and 7. These pages contain all the information required to use the kit.

Procainamide Glycan Labeling System



For more information on our Procainamide system and to view a presentation, please visit <http://www.ludger.com/procainamide/>

Labeling Method

1 Purify the glycans

- Before labeling the glycans, it is preferable to remove non-carbohydrate contaminants from the samples such as protein/peptide material, salts and detergents. This can be achieved using LudgerClean EB10 cartridges ([LC-EB10-A6](#)) or a LudgerClean Protein Binding Membrane ([LC-PBM-96](#)). Clean up of O-glycans can be achieved using LudgerClean CEX cartridges ([LC-CEX-A6](#)).

2 Transfer samples to reaction vials



- The kit is designed to label up to 25 nmols of glycans per reaction. With a single pure glycan, as little as 5 pmoles per reaction can be labeled and detected in subsequent HPLC and MS analysis. Suitable reaction vials include small polypropylene microcentrifuge vials, vials for PCR work or 96 well PCR plates if performing higher throughput sample analysis.

3 Dry the samples



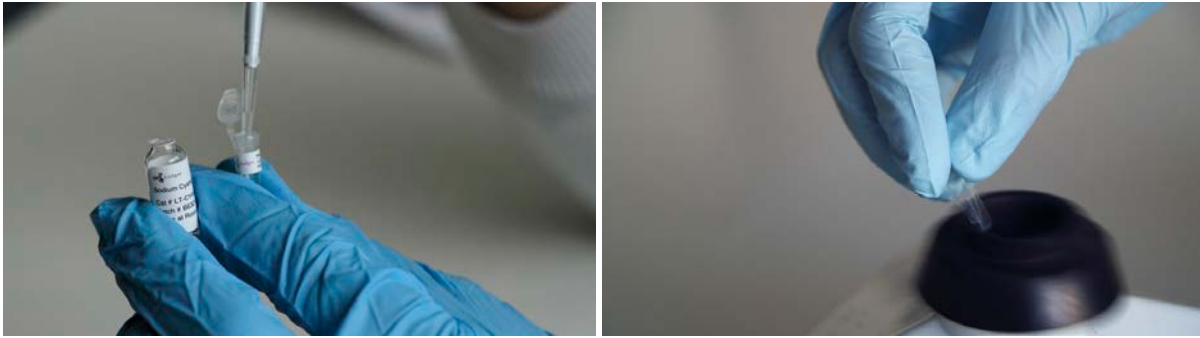
- Dry down the samples.
- Drying down of the samples should be done using a centrifugal evaporator. If this is not possible then freeze drying (lyophilization) can be used with caution (in particular, ensure that the sample dries to a small, compact mass at the very bottom of the vial). Do not subject samples to high temperatures (>28°C) or extremes of pH as these conditions could result in acid catalysed loss of sialic acids (high temperatures, low pH) or epimerization of the glycan reducing terminus (at high pH).

4 Prepare the dye solution



- Add 150 μ L of kit component LT-ACETIC-DMSO-01 (30% acetic acid in DMSO) to a vial of procainamide dye (LT-PROC-01) and mix by pipette action until the dye is dissolved. Sometimes heat (30-60°C) is required to help dissolve the dye.
- Add 150 μ L of water to the dissolved dye solution and mix by pipette action.
- Transfer the 300 μ L of dissolved dye solution to a vial of reductant (LT-CYANO-05) and mix by pipette action until the reductant is dissolved. Sometimes heat (30-60°C) is required to help dissolve the reductant.

5 Add labeling reagent to samples



- Add 20 μL of labeling reagent to each glycan sample, cap the vials or PCR plate, mix thoroughly and centrifuge briefly (5-10 seconds) to ensure the labeling solution is at the bottom of the vial.

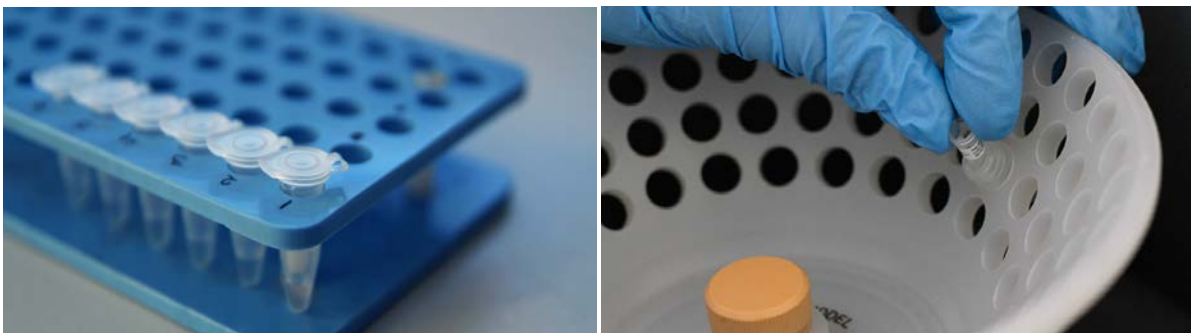
6 Incubate



- Place the reaction vials in an oven or heating block set at 65°C and incubate for 1 hour. We recommend using an oven for the incubation step. An oven provides all around heat for the reaction vials.

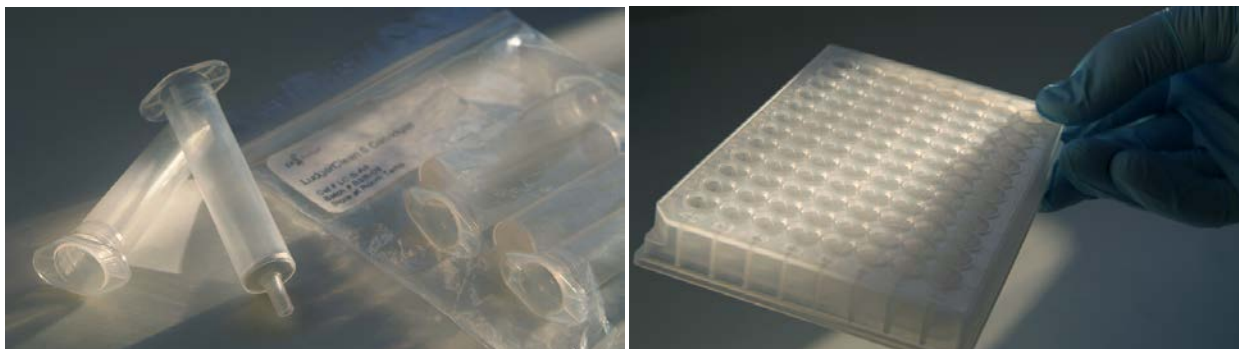
The samples must be completely dissolved in the labeling solution for efficient labeling. To encourage complete dissolution the samples can be vortexed 15 minutes after the start of the incubation then continued.

7 Cool and Centrifuge



- After the incubation period remove the samples from the oven or heating block, allow them to cool to room temperature then briefly centrifuge the reaction vials to ensure the sample is at the bottom of the vial and not in the lid.

LudgerClean™ Post-Labeling Sample Clean-up



For most applications, we recommend that you perform post-labeling sample cleanup to remove non-glycan material, such as excess dye and other labeling reagents, prior to analysis by HPLC, MS or LC-MS.

Benefits of post-labeling cleanup

- Removes free dye and chemicals that may interfere with sample analysis by HPLC and LC-MS.
- Lengthens lifespan of HPLC column.
- Smaller glycans such as O-links close to the start of an HPLC gradient will be detected without interference from the free dye.
- A full range of HPLC columns can be used.
- Sample capacity issues of HPLC columns less likely.

Post-labeling sample clean-up of procainamide (PROC) labeled N-glycans can be achieved using either LudgerClean™ S cartridges (LC-S-A6) or a LudgerClean Procainamide Clean-up Plate (LC-PROC-96) if performing higher throughput sample analysis.

Post-labeling sample clean-up of PROC labeled O-glycans can be achieved using LudgerClean™ S cartridges (LC-S-A6).

This is summarised in Table 1.

Type of glycan	LudgerClean Product	Product Code
PROC labeled N-Glycans	LudgerClean Procainamide Clean-up Plate	LC-PROC-96
	LudgerClean™ S cartridges	LC-S-A6
PROC labeled O-Glycans	LudgerClean™ S cartridges	LC-S-A6

Table 1: Which LudgerClean products should you use for cleaning up PROC labeled N-glycans or PROC labeled O-glycans

Analysis of LudgerTag™ Procainamide Labeled Glycans

LudgerTag™ procainamide labeled glycans may be studied by a number of different analytical methods including (U)HPLC, ESI mass spectrometry and LC-ESI-MS.

Example UHPLC conditions – conditions vary from instrument to instrument. Check these conditions are optimal for your equipment.

- Sample made up in solvent equivalent to starting gradient e.g. 76% acetonitrile.
- Column: Waters BEH Glycan column 15 cm x 2.1 mm.
- Column temperature: 40°C
- Fluorescence detector settings: Excitation wavelength: 310 nm, Emission wavelength: 370 nm.
- Solvent A: 50 mM ammonium formate buffer pH 4.4 (Ludger Product: LS-N-BUFFX40)
- Solvent B: Acetonitrile

Time (min)	% solvent B	Flow Rate (mL/min)
0.00	76	0.4
53.5	51	0.4
55.5	0	0.25
57.5	0	0.25
59.5	76	0.25
65.5	76	0.25
66.5	76	0.4
70.0	76	0.4

Table 2: Long UHPLC gradient for samples where the glycan profile is unknown and where glycans may be large and/or highly sialylated.

The use of this UHPLC gradient is demonstrated for the separation of N-glycans that have been released from human IgG and erythropoietin followed by procainamide labeling (LT-KPROC-V24) and clean-up (LC-PROC-96) (Figures 1 and 2). Once the range of glycans present in your sample has been determined the UHPLC gradient can be optimised and significantly shortened (down to 15 minutes on some UHPLC instruments).

LC-MS analysis of procainamide labeled N-glycans allows the user to obtain both a fluorescent chromatogram of the separated N-glycans (Figure 1) and an ESI-MS spectrum (Figures 3 and 4) that can be combined to give a greater overall picture of N-glycan structures present. The MS data can be used to assign m/z masses to each of the peaks in the fluorescent chromatogram thereby giving possible structural identity, in the form of monosaccharide composition, to each of the peaks (Figure 5 and Table 3). Single peaks in the fluorescent chromatogram can be determined to have multiple glycan structures present by ESI-MS analysis (Figure 4).

The repeatability (precision) of the procainamide labeling and cleanup procedure for N-glycans released from human IgG glycans shows low RSD values (<5% RSD for peaks with a relative % area >0.7 %) (Figure 6 and Table 4).

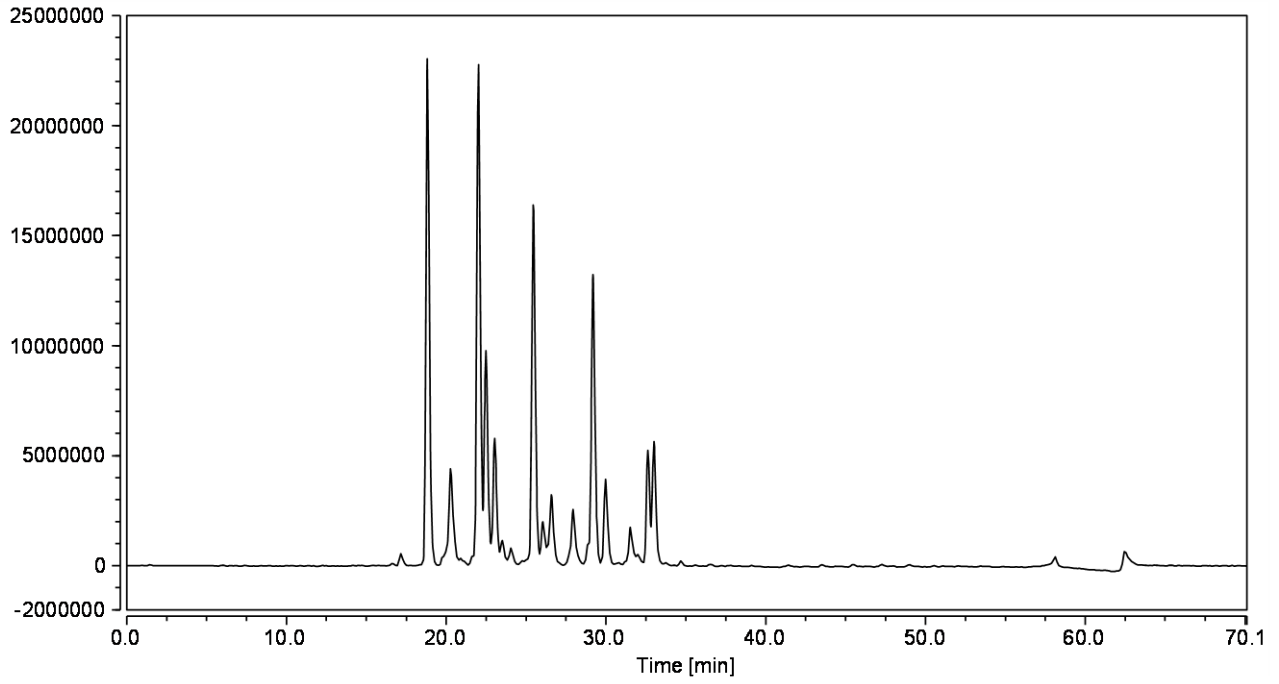


Figure 1: Procainamide labeled human IgG glycans after LC-PROC-96 clean-up

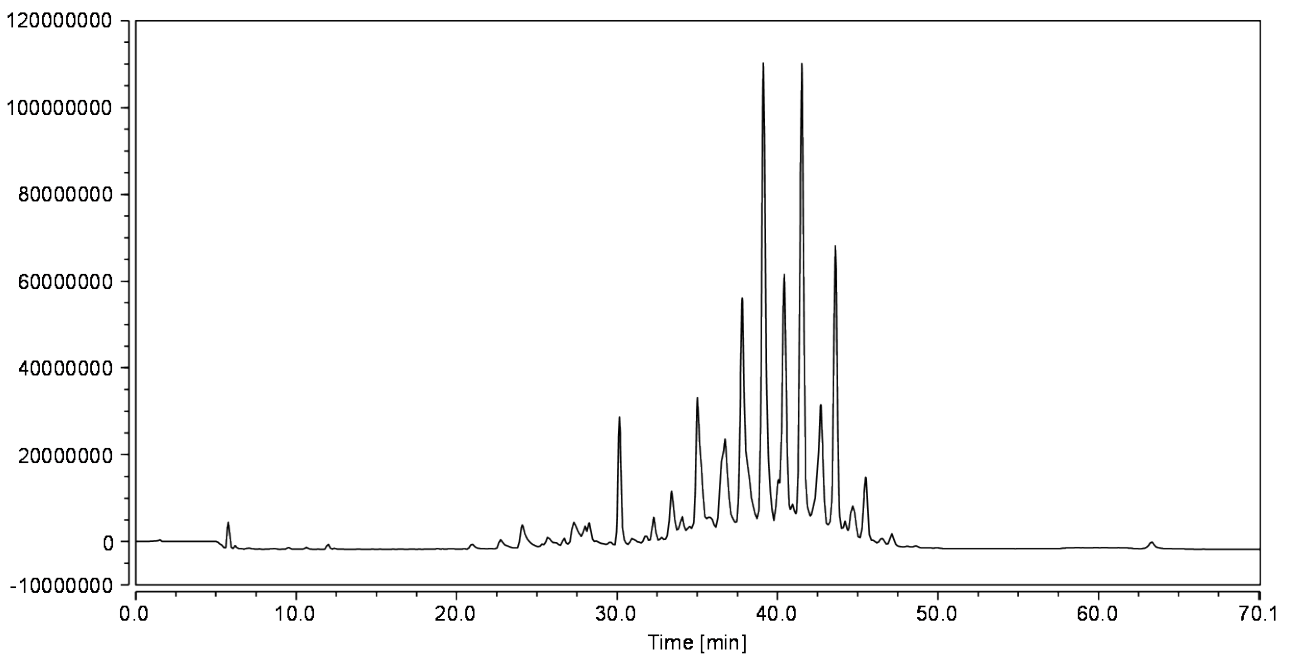


Figure 2: Procainamide labeled erythropoietin glycans after LC-PROC-96 clean-up

References

1. Kozak RP, Tortosa CB, Fernandes DL, Spencer DI. Comparison of procainamide and 2-aminobenzamide labeling for profiling and identification of glycans by liquid chromatography with fluorescence detection coupled to electrospray ionization-mass spectrometry. *Anal Biochem.* 2015; 486: 38-40. doi: 10.1016/j.ab.2015.06.006.
2. Klapoetke S, Zhang J, Becht S, Gu X, Ding X. The evaluation of a novel approach for the profiling and identification of N-linked glycan with a procainamide tag by HPLC with fluorescent and mass spectrometric detection. *J Pharm Biomed Anal.* 2010; 53(3): 315-24. doi: 10.1016/j.jpba.2010.03.045.
3. Pabst M, Kolarich D, Pörtl G, Dalik T, Lubec G, Hofinger A, Altmann F. Comparison of fluorescent labels for oligosaccharides and introduction of a new postlabeling purification method. *Anal Biochem.* 2009; 384(2): 263-73. doi: 10.1016/j.ab.2008.09.041.
4. Liu R, Giddens J, McClung CM, Magnelli PE, Wang LX, Guthrie EP. Evaluation of a glycoengineered monoclonal antibody via LC-MS analysis in combination with multiple enzymatic digestion. *MAbs.* 2016; 8(2): 340-6. doi: 10.1080/19420862.2015.1113361.

The Reductive Amination Reaction

1. Schiff's base formation.

This requires a glycan with a free reducing terminus which is in equilibrium between the ring closed (cyclic) and ring open (acyclic) forms. The primary amino group of the dye performs a nucleophilic attack on the carbonyl carbon of the acyclic reducing terminal residue to form a partially stable Schiff's base.

2. Reduction of the Schiff's base.

The Schiff's base imine group is chemically reduced to give a stable labeled glycan.

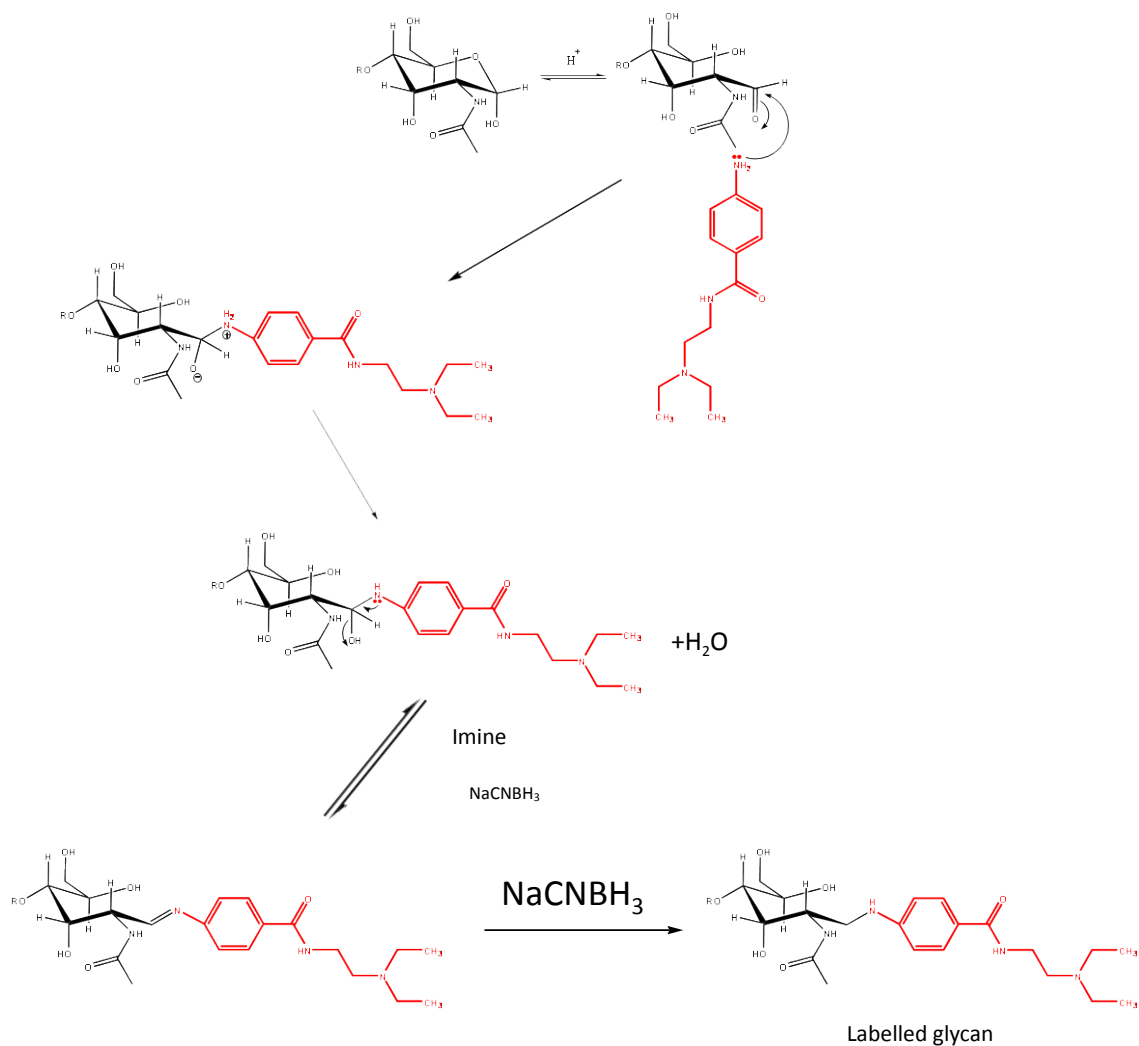


Figure 3: Labeling of a glycan with procainamide (PROC) by reductive amination.

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-KPROC-24-Guide-v1.0

SAFETY DATA SHEET

Version: 1.0 Date written: 12th January 2015

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

Product Name	Procainamide Dye
Product Catalogue Name	LT-PROC-01
CAS-No.	614-39-1
Company:	Ludger Ltd Culham Science Centre Abingdon Oxfordshire OX14 3EB
Telephone:	01865 408554
Emergency Telephone:	01865 408554
Email:	info@ludger.com

2. HAZARDS IDENTIFICATION

2.1 Classification of the substance or mixture

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

Acute toxicity, Oral (Category 4)
 Skin irritation (Category 2)
 Eye irritation (Category 2)
 Specific target organ toxicity - single exposure (Category 3)

2.2 Label elements



Signal Word: Warning

Hazard Statement(s)

H302 Harmful if swallowed.
 H315 Causes skin irritation.
 H319 Causes serious eye irritation.
 H335 May cause respiratory irritation.

Precautionary Statement(s)

P261	Avoid breathing dust/ fume/gas/mist/vapours/spray
P305+P351+P338	IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do so. Continue rinsing.

2.3 Other hazard information:

No supplemental hazard statements.

3. COMPOSITION/INFORMATION ON INGREDIENTS

3.1 Substances

Synonyms: Procainamide hydrochloride; 4-Amino-N-(2-diethylaminoethyl)benzamide hydrochloride; 4-Aminobenzoic acid 2-diethylaminoethylamide

Formula: $C_{13}H_{21}N_3O \cdot HCl$

Molecular Weight: Procainamide hydrochloride: 271.79 g/mol

Component		Concentration
Name	Procainamide Dye	-
CAS-No.	614-39-1	
EC-No.	210-381-7	

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

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.

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, nitrogen oxides (NOx), Hydrogen chloride gas.

5.3 Advice for firefighters

Wear self contained breathing apparatus for fire fighting if necessary.

6. ACCIDENTAL RELEASE MEASURES

6.1 Personal precautions, protective equipment and emergency procedures

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

6.2 Environmental Precautions

Do not let product enter drains.

6.3 Methods and material for containment and cleaning up

Pick up and arrange disposal without creating dust. Sweep up and shovel. Keep in suitable, closed containers for disposal.

6.4 Reference to other sections

Please refer to section 13.

7. HANDLING AND STORAGE

7.1 Precautions for safe handling

Avoid contact with skin and eyes. Avoid formation of dust and aerosols. Provide appropriate exhaust ventilation at places where dust is formed. Normal measures for preventive fire protection.

7.2 Conditions for safe storage, including any incompatibilities

Store in a cool, dark place. Keep the container tightly closed in a dry well ventilated place.

7.3 Specific end uses

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

8. EXPOSURE CONTROLS/PERSONAL PROTECTION

8.1 Control parameters

Contains no substances with occupational exposure limit values.

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

Safety glasses with side-shields conforming to EN166 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.

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

For nuisance exposures use type P95 (US) or type P1 (EU EN 143) particle respirator. For higher level protection use type OV/AG/P99 (US) or type ABEK-P2 (EU EN 143) respirator cartridges. Use respirators and components tested and approved under appropriate government standards such as NIOSH (US) or CEN (EU).

9. PHYSICAL AND CHEMICAL PROPERTIES

9.1 Information on basic physical and chemical properties

Appearance	Form: solid
Odour	No data available
Odour threshold	No data available
pH	No data available
Freezing/Melting Point	Melting point/range: 167 - 169 °C - lit.
Initial boiling point and boiling range	No data available
Flash Point	No data available
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	No data available
Relative Density	No data available
Solubility in water and solvents (mg/l)	No data available
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

No data available

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

Strong oxidizing agents

10.6 Hazardous decomposition products

Other decomposition products - No data available

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

R LD50 Oral - rat - 1,509 mg/kg

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

Carcinogenicity

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

Reproductive toxicity

No data available

STOT-single exposure

Inhalation - May cause respiratory irritation.

STOT-repeated exposure

No data available

Aspiration hazard.

No data available

Potential Health Hazards

Inhalation	May be harmful if inhaled. Causes respiratory tract irritation.
Ingestion	Harmful if swallowed.
Skin	May be harmful if absorbed through the skin. Causes skin irritation.
Eyes	Causes serious eye irritation.

Signs and symptoms of exposure

To the best of our knowledge the chemical, physical and toxicological properties have not been thoroughly investigated.

12. ECOLOGICAL INFORMATION

12.1 Toxicity

No data available

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

No data available

13. DISPOSAL CONSIDERATIONS

13.1 Waste treatment methods

Contact a licensed waste disposal service to collect/dispose of any waste material.

Contaminated packaging

Treat as an unopened/ unused product.

14. TRANSPORT INFORMATION

14.1 UN Number

ADR/RID: - IMDG: - IATA: -

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: -

14.4 Packing group

ADR/RID: - IMDG: - IATA: -

14.5 Environmental hazards

ADR/RID: no IMDG Marine pollutant: no IATA: no

14.6 Special precautions for user

No data available

15. REGULATORY INFORMATION

This safety datasheet 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

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

Note that the label elements, the Risk and Safety phrases (now Hazard and Precautionary statements) that used to be in Section 15 are now in Section 2.

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

Version: 1.1

Date written: 28th September 2011

Date reviewed 9th February 2016

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

Product Name	sodium cyanoborohydride
Product Catalogue Name	LT-CYANOB-05
CAS-No.	25895-60-7
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 the Regulation (EC) No. 1272/2008 [EU-GHS/CLP]

Flammable solids (category 1)
 Substances, which in contact with water emit flammable gases (category 1)
 Acute toxicity, Oral (category 2)
 Acute toxicity, Inhalation (category 2)
 Acute toxicity, Dermal (category 2)
 Skin corrosion (category 1B)
 Acute aquatic toxicity (category 1)
 Chronic aquatic toxicity (category 1)

2.2 Label elements



Signal Word: DANGER

Hazard Statement(s)

H228	Flammable solid
H260	In contact with water releases flammable gases which may ignite spontaneously.
H300	Fatal if swallowed.
H310	Fatal in contact with skin.
H314	Causes severe burns and eye damage.
H330	Fatal if inhaled.
H410	Very toxic to aquatic life with long lasting effects.

Precautionary Statement(s)

P210	Keep away from heat/sparks/open flames/hot surfaces. No smoking.
P223	Keep away from any possible contact with water, because of violent reaction and flash fire.
P231+P232	Handle under inert gas. Protect from moisture.

P260	Do not breathe dust/fume/gas/mist/vapours/spray.
P370+P378	In case of Fire: Use dry sand, dry chemical or alcohol-resistant foam for extinction.
P422	Store contents under inert gas.

2.3 Other hazard information (EU)

EUH032	Contact with acids liberates very toxic gas.
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SECTION 3. COMPOSITION/INFORMATION ON INGREDIENTS

3.1 Substances

Synonyms:	Sodium Cyanotrihydridoborate
Formula:	CH ₃ BNNa

Component	Concentration
Sodium Cyanotrihydroborate	100 %
CAS-No. 25895-60-7	-
EC-No. 247-317-2	

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 doctor/ first responder in attendance.

If Ingested

Do NOT induce vomiting. Rinse mouth well with water, unless person(s) is unconscious.

If skin is exposed

Remove contaminated clothing/shoes immediately. Wash affected area(s) with water and soap.

If eyes are exposed

Wash eye(s) with plenty of water for at least 15 minutes, if unsure seek medical advice.

If inhaled

Move into a source of fresh air, if not breathing give artificial respiration.

4.2 Most important symptoms and effects, both acute and delayed

Burning sensation, coughing, wheezing, laryngitis, shortness of breath, head ache, nausea, vomiting. To the best of our knowledge the chemical, physical and toxicological properties have not been thoroughly investigated. Absorption into the body leads to the formation of methemoglobin which in sufficient concentration causes cyanosis. Onset may be delayed two to four hours or longer.

4.3 Indication of immediate medical attention and special treatment needed

No Data available

SECTION 5. FIRE-FIGHTING MEASURES

5.1 Extinguishing media

Dry powder

5.2 Special hazards arising from the substance or mixture

Carbon oxides, Nitrogen oxides, Hydrogen cyanide (Hydrocyanic acid), Borane/boron oxides.

5.3 Advice for firefighters

Wear self contained breathing apparatus for fire fighting if necessary.

SECTION 6. ACCIDENTAL RELEASE MEASURES

6.1 Personal precautions, protective equipment and emergency procedures.

Wear respiratory protection; gently sweep up to avoid dust formation. Avoid breathing vapours, mist or gas. Ensure adequate ventilation, remove all sources of ignition. Evacuate personnel to a safe area; avoid breathing in dust/gas or mist.

6.2 Environmental Precautions

Prevent further leakage or spillage if safe to do so. Do not let the chemical enter the drainage system and further discharge into the environment must be avoided.

6.3 Methods and material for containment and cleaning up

Contain the spill with matting if necessary and then collect using either an electrically protected vacuum cleaner or by damp brushing (not wet) and putting the collected waste into a secure dry container, do not flush with water. Dispose according to local regulations.

6.4 Reference to other sections

For disposal regulations see section 13.

SECTION 7. HANDLING AND STORAGE

7.1 Precautions for safe handling

Avoid contact with skin and eyes, avoid formation of dust when handling. Provide appropriate exhaust ventilation in work areas where dust could be formed. Keep away from sources of ignition (No Smoking) and 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 the container tightly closed in a dry and well ventilated place. Never allow the product to get into contact with water during storage as it is moisture sensitive. Do not store near acids. Handle and open the container with care. Hygroscopic. Handle when open under an inert gas.

7.3 Specific end uses

No data available

SECTION 8. EXPOSURE CONTROLS/PERSONAL PROTECTION

8.1 Control parameters

Contains no substances with occupational exposure limit values.

8.2 Exposure controls

Appropriate engineering controls

Avoid contact with skin, eyes and clothing. Wash hands before breaks and immediately after handling the product.

Personal protective equipment

Eye/Face protection

Safety glasses to be worn following good laboratory practice. Eye protection should be tested and approved under appropriate government standards such as EN 166 (EU) or NIOSH (US).

Skin protection

Handle with gloves at all times following good laboratory practice. Gloves must be inspected before use and to be removed in proper glove removal technique (without touching the gloves outer

surface) to avoid skin contact. Dispose of contaminated gloves after use as contaminated waste, in accordance with local regulations. Wash and dry hands.

Gloves to be within the specifications of EU directive 89/686/EEC and the standard EN 374 derived from it.

Body protection

Laboratory coat or other protective laboratory clothing should be worn when handling the product.

Respiratory protection

Use extraction unit/fume hood when using the product.

SECTION 9. PHYSICAL AND CHEMICAL PROPERTIES

9.1 Information on basic physical and chemical properties

Appearance	Form: Powder
	Colour: Beige
Odour	No data available
Odour threshold	No data available
pH	No data available
Freezing/Melting Point	Melting point/range :> 242°C
Initial boiling point and boiling range	No data available
Flash Point	No data available
Evaporation rate	No data available
Flammability (solids and gases)	The substance or mixture is a flammable solid with the subcategory 1.
Upper/lower flammability or explosive limits	No data available
Vapour Pressure	No data available
Relative Density	No data available
Solubility in water and solvents	No data available
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

None 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

Reacts violently with water.

10.4 Conditions to Avoid

Do not allow water to enter container because of violent reaction.

Heat, flames and sparks. Extremes of temperature and direct sunlight. Exposure to sunlight.

10.5 Incompatible materials

Do not store near acids, oxidising agents.

10.6 Hazardous decomposition products

Reacts with water to form: - Hydrogen gas

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

Carcinogenicity

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

Reproductive toxicity

No data available

STOT (specific target organ toxicity) -single exposure

No data available

STOT (specific target organ toxicity) -repeated exposure

No data available

Aspiration hazard.

No data available

Potential health effects

Inhalation
of the

May be fatal if inhaled. Material is extremely destructive to the tissue
mucus membranes and upper respiratory tract.

Ingestion

May be fatal if swallowed. Causes burns.

Skin

May be fatal if absorbed through the skin. Causes skin burns.

Eyes

Causes burns to the eyes.

As there is less than 10mg per vial, complete toxicological information is not available.

Signs and symptoms of Exposure

Burning sensation, coughing, wheezing, laryngitis, shortness of breath, head ache, nausea, vomiting. To the best of our knowledge the chemical, physical and toxicological properties have not been thoroughly investigated. Absorption into the body leads to the formation of methemoglobin which in sufficient concentration causes cyanosis. Onset may be delayed two to four hours or longer.

SECTION 12. ECOLOGICAL INFORMATION

12.1 Toxicity

No data available

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

Very toxic to aquatic life with long lasting effects.

SECTION 13. DISPOSAL CONSIDERATIONS

13.1 Waste treatment methods

Product or/and collected waste from spillage.

Burn in a chemical incinerator equipped with an afterburner and scrubber but take extra precautions when igniting as this material is highly flammable. Or to contact a licensed disposal company and arrange disposal, inform the company of the nature of the waste.

Contaminated packaging

Dispose of as the unused product, with a licensed disposal company.

SECTION 14. TRANSPORT INFORMATION

14.1 UN Number

ADR/RID: 3179

IMDG: 3179

IATA: 3179

14.2 UN Proper Shipping Name

ADR/RID: FLAMMABLE SOLID, TOXIC, INORGANIC, N.O.S. (Sodium cyanotrihydroborate)

IMDG: FLAMMABLE SOLID, TOXIC, INORGANIC, N.O.S. (Sodium cyanotrihydroborate)

IATA: Flammable solid, toxic, inorganic, n.o.s. (Sodium cyanotrihydroborate)

14.3 Transport hazard class (es)

ADR/RID: 4.1 (6.1)

IMDG: 4.1 (6.1)

IATA: 4.1 (6.1)

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

No data available

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

Version: 1.0

Date written: 31st March 2015

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

Product Name	acetic acid / dimethyl sulfoxide solution
Product Catalogue Name	LT-ACETIC-DMSO-01
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]

Flammable liquids (Category 3)

Skin corrosion (Category 1A)

2.2 Label elements



Signal Word: Danger

Hazard Statement(s)

H226

Flammable liquid and vapour

H314

Causes severe skin burns and eye damage.

Precautionary Statement(s)

P280

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

 P305+P351+P338
contact

IF IN EYES: Rinse cautiously with water for several minutes. Remove

lenses, if present and safe to do so. Continue rinsing.

P310

Immediately call a POISON CENTRE or doctor/ physician.

2.3 Other hazard information:

None

SECTION 3. COMPOSITION/INFORMATION ON INGREDIENTS

3. 1 Substances

Synonyms:

 DMSO, methyl sulfoxide, dimethyl sulfoxide
Glacial acetic acid

Formula:

 DMSO: C₂H₆OS
Acetic Acid: C₂H₄O₂

Molecular Weight: DMSO: 78.13 g/mol
Acetic Acid: 60.05 g/mol

Component		Concentration
Name	Dimethyl Sulfoxide	70%
CAS-No.	67-68-5	
EC-No.	200-664-3	
Name	Acetic Acid	30%
CAS-No.	64-19-7	
EC-No.	200-580-7	
Index-No.	607-002-00-6	

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

Remove all contaminated clothing immediately; wash the area well with plenty of soap and water.

If eyes are exposed

Flush eyes with plenty of water/ eye wash solution for at least 15 minutes, if present and safe to do so, remove contact lenses and continue rinsing.

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

Nausea, Fatigue and Headache. To the best of our knowledge, the chemical, physical and toxicological properties have not been thoroughly investigated.

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.

ACETIC ACID

CAS-No.	Value	Control Parameters	Update	Basis
64-19-7	TWA	10ppm 25mg/m ³	1991-07-05	Europe. Commission Directive 91/322/EEC on establishing indicative limit on values.
Remarks	Indicative			

DMSO 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	Strong
Odour threshold	No data available
pH	No data available
Freezing/Melting Point	No data available
Initial boiling point and boiling range	No data available
Flash Point	No data available
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	No data available
Relative Density	No data available
Solubility in water and solvents	Completely miscible
Partition coefficient: n-octanol/water	No data available
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 oxidizing agents and strong reducing agents, soluble carbonates and phosphates, hydroxides, metals, peroxides, permanganates, e.g. potassium permanganate, Amines and Alcohols.

10.6 Hazardous decomposition products

Other decomposition products – No data available

SECTION 11. TOXICOLOGICAL INFORMATION

11.1 Information on toxicological effects

DMSO

Acute toxicity

LD50 Oral – Rat – 14,500mg/kg

LC50 Inhalation – Rat – 4h – 40250ppm

LD50 Dermal – Rabbit - > 5,000mg/kg

Acetic Acid

Acute toxicity

LD50 Oral – Rat – 3,310 mg/kg

LC50 Inhalation – Mouse – 1h - 5620ppm

Remarks: Sense Organs and Special Senses (Nose, Eyes, Ears and Taste): Eyes: Conjunctive irritation. Eyes: Other. Blood: Other changes.

LD50 Dermal – Rabbit – 1,112 mg/kg

DMSO

Skin corrosion/irritation

Skin – Rabbit – No skin irritation – 4h

Acetic Acid

Skin corrosion/irritation

Skin – Rabbit – Mild skin irritation – 24h

DMSO

Serious eye damage/irritation

Eyes – Rabbit – Mild eye irritation

Acetic Acid

Serious eye damage/irritation

Eyes – Rabbit – Corrosive to eyes.

Respiratory or skin sensitisation

May cause sensitization by skin contact.

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

Harmful if inhaled. Causes serious respiratory tract irritation.

Ingestion

Harmful if swallowed. Causes burns.

Skin

May be harmful if absorbed through skin. Causes skin burns.

Eyes

Causes eye irritation/ burns.

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

Nausea, Fatigue, Headache. To the best of our knowledge, the chemical, physical and toxicological properties have not been thoroughly investigated.

Additional Information

RTECS: PV6210000

RTECS: AF1225000

SECTION 12. ECOLOGICAL INFORMATION
12.1 Toxicity

DMSO

Toxicity to Fish LC50-Pimephales promelas (fathead minnow) – 34,000mg/l - 96h

LC50-Oncorhynchus mykiss (rainbow trout) – 34,000mg/l-96h

Toxicity to daphnia/Aquatic invertebrates EC50-Daphnia pulex (water fleas) – 27,500mg/l

Toxicity to algae EC50-Lepomis macrochirus (bluegill) - >400,000mg/l-96h

Acetic Acid

Toxicity to Fish LC50 – Leuciscus idus (Golden Orfe) – 410.00mg/l – 48h

LC50 – Cyprinus carpio (Carp) – 49.00mg/l – 48h

LC50 – Pimephales promelas (Fathead minnow) – 79.00 - 88.00mg/l – 96h

LC50 – Lepomis macrochirus – 75mg/l – 96h

Toxicity to Daphnia/Aquatic invertebrates EC50 – Daphnia magna (Water flea) – 65.00mg/l – 48h

12.2 Persistence and degradability

Biodegradability

Remarks: Expected to be biodegradable.

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

Biochemical Oxygen Demand (BOD) - 880mg/g

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

DMSO ADR/RID: -

IMDG: -

IATA: -

Acetic Acid ADR/RID: 2789

IMDG: 2789

IATA: 2789

14.2 UN Proper Shipping Name

DMSO ADR/RID: Not Dangerous Goods

IMDG: Not Dangerous Goods

IATA: Not Dangerous Goods

Acetic Acid	ADR/RID:	ACETIC ACID, GLACIAL
	IMDG:	ACETIC ACID, GLACIAL
	IATA:	Acetic Acid, glacial

14.3 Transport hazard class (es)

DMSO	ADR/RID: -	IMDG: -	IATA: -
Acetic Acid	ADR/RID: 8 (3)	IMDG: 8 (3)	IATA: 8 (3)

14.4 Packing group

DMSO	ADR/RID: -	IMDG: -	IATA: -
Acetic Acid	ADR/RID: II	IMDG: II	IATA: II

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.