Feb 21, 2024

Hydrogen peroxide decontamination of eDNA dedicated material

DOI
dx.doi.org/10.17504/protocols.io.kxygx36ozg8j/v1

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DOI: https://dx.doi.org/10.17504/protocols.io.kxygx36ozg8j/v1

Protocol Citation: Marine Vautier, Cyrielle Galiegue 2024. Hydrogen peroxide decontamination of eDNA dedicated material. **protocols.io https://dx.doi.org/10.17504/protocols.io.kxygx36ozg8j/v1**

Protocol status: Working
We use this protocol and it's working

Created: February 16, 2024

Last Modified: February 21, 2024

Protocol Integer ID: 95332

Keywords: DNA, decontamination, material, eDNA, environmental DNA, hydrogen peroxide, H2O2, hydrogen peroxide decontamination, decontamination protocol, concentrated hydrogen peroxide, use of concentrated hydrogen peroxide, decontamination protocol such as the one, material decontamination step, environmental dna, traces of dna, h2o2, autoclave, possible traces of dna, dna, edna sampling, edna

Abstract

The purpose of this protocol is to detail the **material decontamination** steps to be performed to **remove all traces of DNA** from **environmental DNA (eDNA) dedicated material.**

The use of **concentrated hydrogen peroxide (H_2O_2) solution** requires **careful handling** under a **ventilated hood** with **personal protective equipment** *(gloves and* protective gown and goggles*).*

Note: The autoclave only sterilises equipment and does not eliminate possible traces of DNA. Therefore, materials intended for eDNA sampling must follow a decontamination protocol such as the one proposed here.

Guidelines

The main steps of the protocol are:

- Material preparation
- Material decontamination
- Waste management

Materials

- **Material:**

- All material to be decontaminated.
- Waste container for disposal of diluted 10% hydrogen peroxide solution.
- Personal protective equipment (gloves, gown and goggles).

- **Reagents:**

- Concentrated hydrogen peroxide solution H_2O_2 - to be diluted to a final concentration of 10%

Troubleshooting

Safety warnings

- The use of concentrated hydrogen peroxide solution requires careful handling under a ventilated hood with personal protective equipment *(gloves and* protective gown and goggles*).*

Please see **SAFETY DATA SHEET_Hydrogen Peroxide solution** attached:

SAFETY DATA SHEET_Hydrogen P... 453KB

Before start

- **Preparation of the material:**

- Concentrated hydrogen peroxide solution H_2O_2 - to be diluted to a final concentration of 10%.
- Prepare all material to be decontaminated (removal of DNA contamination).
- Prepare a waste container for the diluted hydrogen peroxide solution.
-Personal protective equipment (*gloves and* protective gown and goggles).

- **Pre and post equipment cleaning:**

- Specific workstation with ventilated hood

- **The following precautions should be taken:**

- Wear personal protective equipment (*gloves and* protective gown and goggles) throughout the handling.
- All operations must be carried out under a special work station with a ventilated hood.

Material preparation

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- *Pre and post equipment cleaning.*
- *Preparation of all material to be decontaminated.*
- *Provide a waste container to dispose of the hydrogen peroxide solution.*
- *Personal protective equipment (gloves and protective gown and goggles) and ventilated hood.*

Material decontamination

2 The aim of this decontamination protocol is to eliminate all traces of DNA present on the material to be used for eDNA analysis.

Note: Handle under ventilated hood and wear personal protective equipment (gloves and protective gown and goggles).

- Switch on the hood ventilation.
- Dilute the concentrated hydrogen peroxide solution H_2O_2 with distilled water to a final concentration of 10%.
- In the case of **bottles or containers:**

- It is possible to pour the concentrated hydrogen peroxide solution directly into the bottle or container and top up with distilled water to dilute the solution. Make sure that the bottle is not completely full so that it can be shaken well.
- Close and shake the bottle or container vigorously.
- Empty the diluted hydrogen peroxide into another bottle to be decontaminated (*if applicable*) or dispose of in a suitable waste container. *Up to ten bottles can be decontaminated with the same hydrogen peroxide solution.*
- Top up with distilled water before shaking again to remove any traces of hydrogen peroxide.
- Repeat this rinsing process 3 times.
- Dry and store the material in a clean room.

Note: After decontamination, do not touch the neck of the bottle and the inside of the caps, and ideally, dry in an incubator at 60/80°C.

- For **small equipment** (filtration equipment, tubes...):

- Take a retention tray and immerse the small equipement in the diluted hydrogen peroxide solution for at least one hour.
- Thoroughly rinse with distilled water (3 times).
- Dry and store the material in a clean room.

Note : Ideally, dry in an incubator at 60/80°C.

- At the end of handling, provide a suitable waste container to dispose of the diluted hydrogen peroxide solution 10% H_2O_2.

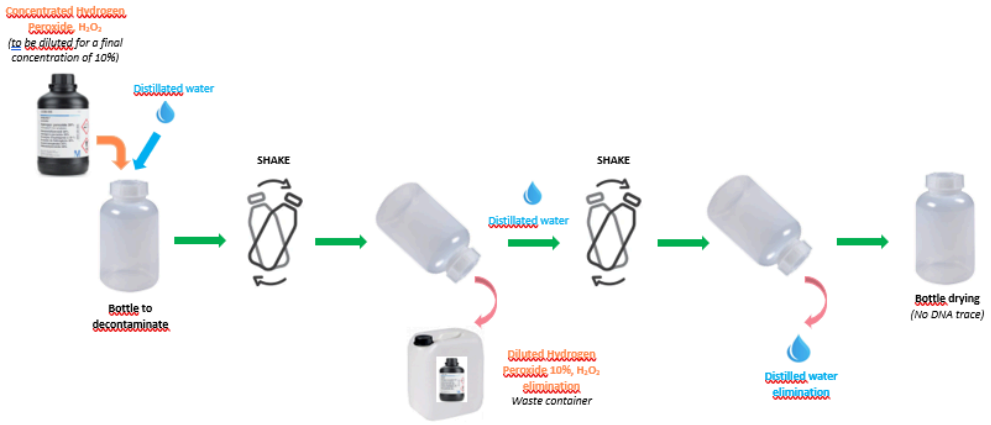

Diagram of bottle decontamination steps

Waste Management

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- The waste container containing the hydrogen peroxide solution must be labelled and disposed of as hazardous liquid waste. Under no circumstances should this waste be released to the environment.