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# Stringency wash buffer V.1

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MPI EVA Ancient DNA C...

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## Abstract

Protocol for the preparation of Stringency wash buffer for automated single-stranded DNA library preparation using the ssDNA2.0 method (Gansauge et al. 2020).

### References

Gansauge, M.-T., Aximu-Petri, A., Nagel, S., & Meyer, M. (2020). Manual and automated preparation of single-stranded DNA libraries for the sequencing of DNA from ancient biological remains and other sources of highly degraded DNA. Nature Protocols, 15, 2279-2300.

### Note

This protocol describes the preparation of 500 ml buffer.

### Materials

| | Reagent/consumable | Supplier | Catalogue number |
|---|---|---|---|
| | ***Reagents*** | | |
| | Water | Sigma Aldrich/Merck | 1153332500 |
| | 20x SSC buffer | Thermo Fisher Scientific | AM9770 |
| | 20% SDS solution | Thermo Fisher Scientific | AM9820 |
| | ***Consumables*** | | |
| | Square media bottle 500 ml | VWR | 391-0630 |
| | 50 ml serological pipet | Corning BV | 357550 |
| | 5 ml serological pipet | Corning BV | 357543 |

### Equipment

- Serological pipette controller (e.g. battery-powered pipetting aid ROTILABO, cat. no. TC16.1)

### Protocol

1. Prepare the buffer in a 500 ml square media bottle by adding the following reagents. Use the glass pipette for transfer of large volumes (> 1 ml). Mix reagents by shaking the bottle.

| | Reagent | Volume | Final concentration in reaction |
|---|---|---|---|
| | Water | 495 ml | |
| | 20x SSC buffer | 2.5 ml | 0.1x |
| | 20% SDS solution | 2.5 ml | 0.1% |

| | Reagent | Volume | Final concentration in reaction |
|---|---|---|---|
| | *sum* | *500 ml* | |

Note

**[Note]**

It is also acceptable to use the scale of the bottle to fill up to 400 ml with water, then adding the remaining 95 ml using the glass pipette.

2. Review the protocol in which the buffer is used to determine whether the buffer should be decontaminated using UV treatment. Instructions for UV-decontamination are provided in the Appendix.

Note

**[Labeling]**

Label the bottle with the buffer name, batch ID, date and the initials of the person who prepared the buffer.

Attention: Every single bottle prepared at the same day gets a new batch ID. Name the batches with Roman numerals (e.g. batch I, batch II, etc.)

3. Store the buffer at room temperature until used. Shelf life is at least two months from preparation.

Note

**[Note]**

Note the lot numbers and the date and initials of the reagents used for the preparation in Labfolder (orange fields).

**Appendix**

Document

NAME

# UV decontamination of reagents/buffers

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Preview