

MasterAmp™ Buccal Swab DNA Extraction Kit

Cat. Nos. MB71030 and MB7901S

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1. Introduction

The MasterAmp™ Buccal Swab DNA Extraction Kit is a fast, simple and inexpensive means for preparing human genomic DNA for amplification. In contrast, extraction of DNA from blood samples involves considerable expense, patient distress and special handling of liquid samples to minimize exposure to blood-borne pathogens. Buccal cell samples can be collected by individuals without extensive training, potentially facilitating the participation of family members in genetic studies.¹ Individual, sterile Buccal Swab Brushes are provided in paper pouches for collection of tissue. Dry brush samples are stable for up to 1 month at 37°C, allowing for remote site collection and transportation. Subsequent DNA extraction requires only heat treatment to lyse the epithelial cells and to degrade compounds inhibitory to amplification.² Following heat treatment and centrifugation to pellet debris, sample DNA is ready for PCR. Yields range from 0.5-3 µg of DNA from each buccal sample. Furthermore, extracted DNA contains MasterAmp PCR Enhancer (with betaine),[†] which substantially improves product yield and decreases nonspecific product formation with many difficult templates.

2. Product Specifications

Storage: Store the tubes of MasterAmp Buccal Swab DNA Extraction Solution at -20°C in a freezer without a defrost cycle. Store the sterile buccal brushes at room temperature.

DNA Extraction Solution: In addition to a proprietary reagent, the DNA Extraction Solution contains 50 mM Tris-HCl (pH 10.5), 1 mM EDTA, and MasterAmp PCR Enhancer at 10X final concentration.

Functional Assay: The MasterAmp Buccal Swab DNA Extraction Kit is assayed by extracting DNA from human buccal cells and amplifying the human CHLC STR marker DXS6797 using MasterAmp AmpliTherm™ DNA Polymerase in conjunction with MasterAmp PCR PreMix C.

3. Kit Contents

Cat. #	Quantity
The MasterAmp™ Buccal Swab DNA Extraction Kit contains enough reagents to perform 30 extractions.	
MasterAmp™ Buccal Swab DNA Extraction Kit (standard)	
MB71030	
Buccal Brushes	30 brushes
DNA Extraction Solution	30 tubes
MasterAmp™ DNA Extraction Solution	
MB7901S	
DNA Extraction Solution	50 ml

4. Related Products

The following products are also available:

- MasterAmp™ Buccal Swab DNA Extraction Kit (remote site testing)
- MasterAmp™ DNA Extraction Solution
- MasterAmp™ Buccal Swab Brushes
- MasterPure™ Nucleic Acid Purification Kits
- FailSafe™ PCR System
- BuccalAmp™ DNA Extraction Kits
- QuickExtract™ DNA Extraction Solution 1.0
- Catch-All™ Sample Collection Swabs

5. DNA Extraction Protocol

Note: *The DNA Extraction Solution contains a bead matrix that may vary in volume among the tubes. This variation does not alter the effectiveness of the DNA Extraction Solution.*

1. Thaw the appropriate number of tubes containing DNA Extraction Solution.
2. Thoroughly rinse out the subject's mouth twice with water. We recommend that subjects abstain from drinking coffee before tissue collection. Alternatively, instruct subjects to gently brush the inside surface of both cheeks with a toothbrush (without toothpaste); follow with a thorough rinsing of the mouth with water.
3. Collect tissue by rolling the Buccal Brush firmly on the inside of the cheek, approximately 20 times on each side, making certain to move the brush over the entire cheek. Either extract the DNA immediately, or air dry the brush for 10-15 minutes at room temperature. Store the dry brushes in the original packaging at 22-37°C for up to one month before extracting the DNA. For longer term storage, place the dry brushes in the original packaging at -20°C for up to 6 months. Yield is directly correlated with the starting amount of buccal cells. If yield is not a concern, use only one brush; if yield must be maximized, use a separate brush for each cheek surface, and if necessary, use a third brush, collecting tissue from both cheeks.
4. Place the Buccal Brush into a tube containing DNA Extraction Solution and rotate the brush a minimum of 5 times. Press the brush against the side of the tube and rotate the brush while removing it from the tube to ensure most of the liquid remains in the tube.
5. Screw the cap on the tube tightly and vortex mix for 10 seconds. Incubate the tube at 60°C for 30 minutes.
6. Vortex mix for 15 seconds.
7. Transfer the tube to 98°C and incubate for 8 minutes.
8. Vortex mix for 15 seconds.
9. Return the tube to 98°C and incubate for an additional 8 minutes.
 10. Vortex mix for 15 seconds. Chill the tube on ice briefly to reduce the temperature.
11. Pellet cellular debris by centrifugation at 4°C for 5 minutes.

12. Carefully transfer the supernatant containing the DNA to a clean tube without including any of the beads.
13. Store the DNA at -20°C , or at -70°C for longer term storage.

The yield of DNA is usually between 2-8 ng/ μl . The concentration of MasterAmp PCR Enhancer (with betaine) in DNA prepared according to the above protocol is 10X. For templates containing high G+C content or secondary structure, we recommend using 15 μl of the supernatant in a 50- μl amplification reaction. This will yield a final concentration of approximately 3X MasterAmp Enhancer (with betaine). For some templates, 3X MasterAmp Enhancer (with betaine) is not optimal. We recommend using less of these templates in a 50- μl amplification reaction (e.g., 2-5 μl).

6. References

1. Richards, B. *et al.*, (1993) *Hum. Molec. Genet.* **2**, 159.
2. Ruano, G. *et al.*, (1992) *BioTechniques* **13**, 266.

**Use of betaine in DNA polymerase reactions, including, but not limited to use for PCR or DNA sequencing, is covered by U.S. Patent No. 6,270,962, European Patent No. 0742838, German Patent No. DE4411588C1, and other patents or patent applications in the U.S. and other countries that are either assigned or exclusively licensed to Epicentre. Purchase of a product from Epicentre that contains betaine and a thermostable DNA polymerase is accompanied by a limited non-exclusive license for the purchaser to use the purchased product solely for life science research, whether the purchaser performs research in a not-for-profit or a for-profit organization. However, if the product does not contain a thermostable DNA polymerase in addition to betaine, all for-profit organizations require a license from Epicentre in order to use betaine or a product that contains betaine in DNA polymerase reactions for research applications, and not-for-profit organizations require a license if the product, the research, or the result of the research is transferred to or obtained for or on behalf of a for-profit organization. Licenses are also available to use betaine in DNA polymerase reactions for human or animal diagnostics, screening, or other fields of use. Please contact Epicentre for information related to such licenses.*

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