

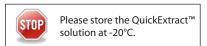
BuccalAmp™ DNA Extraction Kit

QuickExtract™ DNA Extraction Solution 1.0

Catch-All™ Sample Collection Swabs

Cat. Nos. BQ0901S (CR, SC, RB, BS), BQ0908S (CR, SC, RB, BS), BQ0916S (CR, SC, RB, BS), QE09050, QEC091H, QEC89100, MB100BR, and MB100SP





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1. Introduction

The BuccalAmp™ DNA Extraction Kit is a single-tube system for rapid preparation of DNA from human or other mammalian buccal (cheek) swab samples for PCR assays. The kit incorporates the QuickExtract™ DNA Extraction Solution that permits processing of samples using a simplified protocol. The kits provide a choice of buccal swab or brush and include aliquots of the QuickExtract Solution as individual sample tubes. To obtain PCR-ready DNA, just rotate the buccal sample swab in one of these tubes, mix, and heat. No centrifugation step is needed, so sample handling times are short. Thus, the BuccalAmp Kit makes it much easier to process one to hundreds of samples in less than an hour without phenol, chloroform, or other toxic organic solvents. The QuickExtract DNA Extraction Solution is also available separately. DNA yields obtained using the BuccalAmp Kit with Catch-All™ Swabs range from 1-7 µg of DNA per buccal sample. This is enough to perform about 100 PCR assays. The Catch-All Swabs are recommended for greater yields compared to the MasterAmp™ Brushes. The extracted DNA contains MasterAmp PCR Enhancer (with betaine),⁺ which substantially improves product yield and decreases nonspecific product formation with many difficult templates.

Storage: Upon arrival, store the tubes of QuickExtract DNA Extraction Solution 1.0 at -20°C. **Do not** subject the Extraction Solution to repeated freeze/thaw cycles. Store the sterile Catch-All Sample Collection Swabs at room temperature.

Quality Control: The BuccalAmp DNA Extraction Kit is function-tested by assaying for the production of a PCR product from a human X chromosomal marker (Cooperative Human Linkage Center, marker DXS7132), using a buccal cell DNA preparation as template.

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2. Kit Contents

Cat.#		Quantity
BuccalAmp™ DNA Extraction Kits with Catch-All™ Hard Pack Buccal Swab		
BQ0901SCR	1 Kit	(15 Tubes & Swabs)
BQ0908SCR	8 Kits	(120 Tubes & Swabs)
BQ0916SCR	16 Kits	(240 Tubes & Swabs)
BuccalAmp™ DNA Extraction Kits with Catch-All™ Soft Pack Buccal Swab		
BQ0901SSC	1 Kit	(15 Tubes & Swabs)
BQ0908SSC	8 Kits	(120 Tubes & Swabs)
BQ0916SSC	16 Kits	(240 Tubes & Swabs)
BuccalAmp™ DNA Extraction Kits with MasterAmp™ Brush Hard Pack		
BQ0901SRB	1 Kit	(15 Tubes & Brushes)
BQ0908SRB	8 Kits	(120 Tubes & Brushes)
BQ0916SRB	16 Kits	(240 Tubes & Brushes)
BuccalAmp™ DNA Extraction Kits with MasterAmp™ Brush Soft Pack		
BQ0901SBS	1 Kit	(15 Tubes & Brushes)
BQ0908SBS	8 Kits	(120 Tubes & Brushes)
BQ0916SBS	16 Kits	(240 Tubes & Brushes)
QuickExtract™ DNA Extraction Solution 1.0		
QE09050	50 ml	(100 extractions)
Catch-All™ Sample Collection Swab (Hard Pack)		
QEC091H	in hard-pack plastic carrier	100 Buccal Swabs
Catch-All™ Sample Collection Swab (Soft Pack)		
QEC89100		100 Buccal Swabs
MasterAmp™ Buccal Swab Brush (Hard Pack) MB100BR in hard-pack plastic carrier 100 Buccal Brushes		
IVID I UUDK	in hard-pack plastic carrier	TOO DUCCAI Brusnes
MasterAmp™ Buccal Swab Brush (So	oft Pack)	
MB100SP		100 Buccal Brushes

3. Related Products

The following products are also available:

- FailSafe™ PCR System
- TAQXpedite[™] PCR System (Fast end point)
- MasterPure™ Complete DNA and RNA Purification Kit
- MasterAmp[™] *Taq, Tth, Tfl*, and AmpliTherm[™] DNA Polymerases

4. Rapid DNA Extraction Protocol

- Label the appropriate number of tubes containing QuickExtract DNA Extraction Solution 1.0.
- 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) followed by a thorough rinsing of the mouth with water.
- 3. Collect tissue by rolling the Catch-All sample collection swab firmly on the inside of the cheek, approximately 20 times on each side, making certain to move the brush over the entire cheek. If storing or transporting the sample, air dry the swab for 10-15 minutes at room temperature. Store the dry swab in the original packaging at 22-37°C for up to one week before extracting the DNA. For longer storage, place the dry swabs 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 swab; if yield must be maximized, use a separate swab for each cheek surface, and if necessary, use a third swab, collecting tissue from both cheeks.
- 4. Place the swab end of the Catch-All sample collection swab into a tube containing QuickExtract DNA extraction solution and rotate the brush a minimum of five 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 65°C for 1 minute.
- 6. Vortex mix for 15 seconds.
- 7. Transfer the tube to 98°C and incubate for 2 minutes.
- 8. Vortex mix for 15 seconds.
- 9. Quantitate DNA yield by fluorimetry using Hoechst dye 33258 to avoid an over estimation given by A_{260} readings.¹
- 10. Store the DNA at -20° C, or at -70° C for longterm storage.

The yield of DNA is usually between 2-14 ng/ μ l. The QuickExtract DNA Extraction Solution 1.0 contains the MasterAmp PCR Enhancer (with betaine). The presence of this reagent may change the annealing temperature of a given primer pair. We recommend using 5 μ l of extracted DNA in a 50- μ l PCR amplification reaction. For target sequences containing high G+C content or secondary structure, we recommend using 5-15 μ l of the extracted sample.

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5. OuickExtract DNA Extraction Protocol

The QuickExtract DNA Extraction Solution, currently available separately or as a component of the BuccalAmp DNA Extraction Kit, provides an extremely efficient method for extracting PCR-ready genomic DNA from diverse samples. The following protocol has been used to extract PCR-ready genomic DNA from HeLa cells, hair follicles, tail snips, bacterial cells, fingernails, and feathers.

- 1. Label the appropriate number of tubes containing 0.5 ml of QuickExtract Solution.
- 2. Place one sample in each tube, for example:
 - 10⁴ counted human cervical carcinoma tissue culture (HeLa) cells.
 - a 0.5-1 cm region of a single plucked human hair with follicle.
 - a 0.5-1 cm section of a mouse tail snip, finely diced using a fresh blade.
 - one single E. coli colony picked from a plate.
 - a 0.5-1 cm guill-end of a breast feather that was plucked and stored at 4°C.
- 3. Vortex mix for 15 seconds.
- 4. Transfer the tube to 65°C and incubate for 6 minutes (15 minutes for fingernails).
- 5. Vortex mix for 15 seconds.
- 6. Transfer the tube to 98°C and incubate for 2 minutes.
- 7. Store the DNA at -20° C, or at -70° C for long term storage.
- 8. Use 5 μl or less of the extracted DNA for each PCR amplification (see Fig. 1).

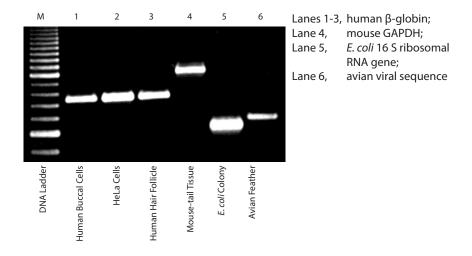


Figure 1. Genomic DNA extracted from a variety of tissues or cells using the QuickExtract™ DNA Extraction Solution were consistently amplified using the FailSafe PCR System.

6. Reference

 Ausubel, F. et al., (eds.) (1995) Current Protocols in Molecular Biology (CD ROM ver. 3.7.5) John Wiley and Sons, New York, Appendix 3D.

*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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