

# JETQUICK protocol for the purification of DNA from whole blood, serum, plasma or other body fluids

**Reconstitute buffers KX and K2 with absolute ethanol as stated on the bottle's label!**

**Prepare GENOMED protease and RNase A as stated on the respective labels! Store the dissolved enzymes in single-use aliquots at -20°C. Avoid multiple freezing/thawing once the enzymes are dissolved in water.**

**The JETQUICK Blood & Cell Culture Kit 50 (Cat. No. 440 050) is designed for 50 preparations, the JETQUICK Blood & Cell Culture Kit 250 (Cat. No. 440 250) for 250 preparations from 200 µl of whole blood.**

1.) Pipette into a 1.5 ml or 2.0 ml reaction tube (i.e. Eppendorf) **up to 200 µl** of whole blood, serum, plasma, buffy coat oder another body fluid.

2.) Add **20 µl GENOMED protease** (20 mg/ml) and **200 µl Buffer K1** to this sample and **mix very thoroughly** either by vortexing or by inverting of the tube.

**Do NOT add GENOMED protease directly to buffer K1. First mix the blood sample with the enzyme, mix, then add buffer K1.**

**Buffer K1:** Contains guanidine hydrochloride and a detergent. These substances are irritants. Use with proper precaution! Wear gloves and safety goggles!

**OPTIONAL:** If one wants to prepare RNA-free genomic DNA, add **10 µl of RNase A solution** (20 mg/ml) **prior** to the addition of buffer K1.

3.) Incubate for **10 min at 58°C**.

4.) Add **200 µl of absolute ethanol** to the mixture and mix **immediately and very thoroughly** in order to prevent any precipitation of nucleic acids due to too high local alcohol concentrations.  
*Do not use other alcohols than ethanol, because other alcohols may cause inconsistent yields.*

5.) Assemble a JETQUICK micro-spin column with a 2 ml receiver tube. Apply the sample from step 4 into the JETQUICK micro-spin column without moistening the rim of the micro-spin cup and centrifuge the sample for **1 min at 10.000 x g** (approximately 10.600 rpm) through the silica membrane.

6.) Discard the flowthrough, re-assemble the Micro-Spin unit with the receiver tube and wash the JETQUICK column by applying **500 µl buffer KX reconstituted with ethanol** and centrifuging for **1 min at 10.000 x g** (approximately 10.600 rpm).

7.) Discard the flowthrough, re-assemble the Micro-Spin unit with the receiver tube and wash the JETQUICK column by applying **500 µl buffer K2 reconstituted with ethanol** and centrifuging for **1 min at 10.000 x g** (approximately 10.600 rpm).

- 8.) Discard the flowthrough, re-assemble the micro-spin and the receiver tube and centrifuge the empty tube again for **1 min at full speed (~13,000 rpm)** in order to clear the silica membrane from residual liquid.
  
- 9.) Insert the JETQUICK micro-spin tube into a new, sterile 1.5 ml reaction tube and elute the DNA from the membrane with **200 µl of 10 mM Tris-HCl buffer (pH 8,5)** or simply water.

*The elution buffer should be used prewarmed to 70 °C and pipetted directly onto the center of the silica membrane. Take care that the whole membrane comes into contact with the elution buffer.*

*Incubate the spin column for **2 min at room temperature** after application of the elution buffer and centrifuge subsequently for **2 min at 10.000 x g** (approximately 10.600 rpm). The eluate now contains the pure DNA that can be further processed immediately.*

*Yield can be increased by up to 15% doing a second elution step with the same 200 µl of DNA-containing eluate. Preheat the 200 µl of eluate to 70 °C, fill it into the micro-spin device and proceed as described before.*

*If lower DNA yields (< 3 µg) are to be expected, the volume of the elution buffer can be reduced to **100 µl** in order to obtain a higher DNA concentration in the eluate.*

*For expected DNA yields that are even < 1 µg, one can consider the reduction of the elution buffer volume to **50 µl**. However, any reduction of the elution buffer volume will lead to a somewhat reduced overall DNA yield of approximately 10-15%.*