

# Saliva Sample Handling Instructions

For our DetectX<sup>®</sup> Immunoassay and Detection Kit

## INTRODUCTION

The use of saliva as a relatively non-invasive sample has become widespread. However its use suffers from a number of potential drawbacks. These are high viscosity, potential discoloration, and particles from food intake. There are also known interferences that can occur depending on the method of collection or processing. These interferences can be high pipetting variability, variable background signal in spectrophotometric signal detection and nonspecific binding.

## MATERIALS NEEDED

- 15 mL centrifuge tubes
- Plastic vials
- Protease inhibitors
- Activated Sodium Orthovanadate. For protocol on preparation see page 7 of PKA Activity Assay kit manual, Catalog No. K027-H for details: [www.arborassays.com/documentation/inserts/K027-H.pdf](http://www.arborassays.com/documentation/inserts/K027-H.pdf)
- Other enzyme inhibitors as appropriate

## PROCEDURE

### General

Whole saliva should be obtained at least 2 hours after eating and rinsing mouth with water to avoid any food borne antigens or materials from affecting the analysis.

### Stable Antigens

Methods of saliva collection vary widely. In our labs we ask volunteers to collect their saliva by allowing the saliva to passively flow into a 15 mL centrifuge tube. For measurements of stable antigens, such as steroids, the saliva is frozen at -20°C. Upon thawing, the saliva is centrifuged at 2,500 x g for 20 minutes and the clear supernatant is pipetted off any precipitated material. Analyze immediately or aliquot and freeze at -20°C.

### Possibly Unstable Antigens

To minimize degradation of unstable antigens, keep samples on ice and immediately add enzyme inhibitors, such as, protease inhibitor cocktail (Sigma, 1 µL/mL whole saliva) and 1 mM of sodium orthovanadate for peptides and proteins, IBMX (Catalog Number P019-100MG and P019-1GM) for cyclic nucleotide measurements, or a general cyclooxygenase inhibitor, such as meclofenamic acid or indomethacin at 15 µM for prostaglandin assays, are added immediately after sample collection. All samples should be kept on ice during the process. The saliva is frozen at -20°C. Upon thawing, the saliva is centrifuged at 2,500 x g for 20 minutes and the clear supernatant is pipetted off any precipitated material. Analyze immediately or aliquot and freeze at -80°C.