# Investigating a dual sIgA and alpha-amylase Point of Care test in the sporting environment

Dunbar J, Hazell G, & Jehanli A. IPRO Interactive, Wallingford, UK.

## Introduction

The use of salivary biomarker responses has gathered momentum in recent years in sports, exercise and behavioural sciences. A Point of Care (POC) platform using Lateral Flow Device (LFD) technology, which takes just over 10 minutes to measure salivary IgA, has previously been validated (Coad et al., 2015).

Another analyte gaining popularity is salivary alpha-amylase (sAA), an acute stress biomarker, which can be measured on the IPRO POC platform using a novel antibody capture, rather than enzymatic method (Dunbar et al., 2015).

The ability to multiplex sampling would save processing time and speed up delivery of data in the applied setting when the assessment of more than one biomarker is required and such a POC test would certainly give a significant time advantage over standard laboratory techniques, which often reveal data to sporting squads only days later.

This paper assesses a new POC LFD for the rapid determination of IgA / sAA in comparison to a novel antibody capture laboratory ELISA determination.

### Methods

A total of 56 saliva samples were taken from 3 cohorts of English Premier League soccer players using IPRO Oral Fluid Collection (OFC) kits. The OFC kits collect 0.5mL of oral fluid and contain a colour changing volume adequacy indicator within the swab, giving collection times typically in the range of 20-50 seconds (Jehanli et al., 2011).

The samples analysed in this study were taken during routine monitoring: before training sessions, during a competitive season. The samples were analysed immediately to determine sIgA and sAA concentrations via the dual analyte POC LFD.

Two drops of saliva/buffer mix from the OFC were added to the sample window of the sIgA / sAA LFD. The liquid runs the length of the test strip via lateral flow, creating a control and two test lines visible in the test window. Ten minutes after the sample is added, the test line intensities were measured using an IPRO LFD Reader. The test line intensities are inversely proportional to the sIgA and sAA concentrations in the sample giving a quantitative value. The same samples were then taken to a laboratory for ELISA analysis within 4 hours of sample collection.

Measurement range on ELISA was 18.5-600  $\mu g/ml$  for sIgA and 20 - 4000  $\mu g/ml$  for sAA; on the the POC test it was 18.5 - 900  $\mu g/ml$  for sIgA and 20 - 4000  $\mu g/ml$  for sAA.

## Results

The sIgA values ranged from 33 - 500  $\mu$ g/ml on ELISA and 28- 684  $\mu$ g/ml on the LFD; whilst sAA concentrations measured via ELISA ranged from 68 - 1698  $\mu$ g/ml and with the POC test from 52 - 4000  $\mu$ g/ml. The Pearson correlation between test types was r = 0.90 (95% CI, 0.84 - 0.94) for IgA and r= 0.87 (95% CI, 0.78 - 0.92) for the sAA, thus showing good validity for both assays, but absolute values for the sAA POC tended to be higher than ELISA.

### Figure 1: sIgA ELISA & IPRO LFD



One of the most important aspects of such technology for the applied user is the repeatability of measurement. Athletes are more concerned about how their readings vary on a longitudinal basis, rather than how the LFD performs in comparison to the ELISA, so good repeatability is important.

Six of the samples were run as six replicates and the values, mean, S.D. and Coefficient of Variation (CV) of these replicates (expressed as a percentage) are displayed in Table 1.

#### Figure 2: sAA ELISA & IPRO LFD

Table 1: Repeatability (CV %) of six replicates of the dual sIgA / sAA LFD in six different samples.

Sample	IgA (µg/ml)	CV (%)	sAA (µg/ml)	CV (%)
1	160	6.78	3543	14.2
2	55	14.4	329	9.91
3	168	6.22	1188	11.3
4	20	3.42	127	12.2
5	417	6.43	251	16
6	181	5.6	68	15.5
Mean		7.15		13.2

#### **Conclusion / Practical Implications**

The point of care test shows good agreement with the ELISA method for the determination of sIgA and sAA. Given the quick data turnaround and efficiency in terms of cost, it represents a suitable alternative method for use in sports teams. Given the fact that both IgA and sAA concentrations can now be performed on site, in the training environment, alongside other markers such as cortisol on the same device; this test represents a true paradigm shift in the way athletes can be monitored, in that results are gained within twelve minutes from sample collection and subsequent intervention strategies can be applied immediately where appropriate.

### References

Coad S, Mclellan C, Whitehouse T & Gray B (2015) Validity and reliability of a novel salivary immunoassay for Individual Profiling in Applied Sports Science. *Research in Sports Medicine* 23 (2): 140-150.

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The IPRO POC system in use in football





Innovation Centre Howbery Park Benson Lane Wallingford, 0X10 8BA, UK



www.iprointeractive.com joe.dunbar@iprointeractive.com 01491 822622 f /iprointeractiv 07889 920473 @@iprointeractiv

12th International Society of Exercise Immunology Symposium, Vienna, Austria 2015