

Australian Government Analytical Laboratories

Progress Report

on

**Statistical Population Studies to Support New
Analytical Methodologies using the EPO2000 Project
Urine Samples**

Prepared for

**The Department of Communications Information
Technology and the Arts (DCITA)**

5 December 2003

Summary of Project

The aims of this project are twofold –

1. to determine the variability of the natural isoform pattern of urinary EPO and evaluate whether the patterns are significantly affected by factors such as gender, ethnicity and altitude, and
2. to determine the variability in the isotope ratio of some selected endogenous steroid metabolites and evaluate whether the delta ^{13}C values are significantly affected by factors such as gender, ethnicity and geographical location.

Project Milestones

Achievement Date	Milestone	Milestone
	EPO Glycoforms	CIR Measurements
5 December 2003	<ul style="list-style-type: none">• Organise logistics [how many samples, from whom etc]• Organise samples• Analyse samples	<ul style="list-style-type: none">• Organise logistics• Organise samples• Analyse samples• Evaluation of data
16 April 2004	<ul style="list-style-type: none">• Analyse samples (Contd)• Evaluation of data	<ul style="list-style-type: none">• Analyse samples (Contd)• Evaluation of data
30 June 2004	<ul style="list-style-type: none">• Final report to Anti Doping Research Program Panel	<ul style="list-style-type: none">• Final report to Anti Doping Research Program Panel

Progress to date

- The work on the profiling for both EPO and CIR are well within the milestones and the work will be completed within the stated schedule.

EPO GLYCOFORMS

Milestones 1 and 2 – Organise logistics and Organise samples:

- Urine samples are progressively drawn from the operational sample base. This ensures a supply of recently collected and properly stored urine samples for analysis. The samples are first completely analysed for the requested screen, and reported negative.
- Once cleared, the samples are selected on the basis of available sample volume, and to provide a broad range of sports.
- The samples come from the whole range of sports that undergo testing at ASDTL and include power, endurance, combined power/endurance, skill and aesthetic pursuits.
- Some samples are drawn from the sporting disciplines currently tested most extensively under the current EPO testing regime, but these disciplines, including cycling, rowing, canoeing, cycling and triathlon, are deliberately under-represented.

Milestone 3 – Analyse samples:

- 322 samples have been screened for EPO content.
- 195 males in 32 sports, and 127 females in 28 sports. In total, 36 sports are covered.
- Samples from the five sports that are currently heavily targeted (cycling, canoeing, rowing, swimming, triathlon) make up 13% of the population, reflecting their occurrence in the overall urine-testing program.
- 198 samples were run on 20 gels, and 174 quantifiable results were obtained.
- The preliminary mean and standard deviation for the principal assessment characteristics were:
 - Ratio of largest band coincident with recombinant standard to largest acidic band: 0.9 ± 0.8
 - Ratio of second largest band coincident with recombinant standard to largest acidic band: 0.7 ± 0.7
 - Percentage of signal coincident with, or more basic than, recombinant standard: $37.3 \pm 13.5\%$
 - Further statistical analysis will be evaluated when the complete set of 600 samples have been completed

ISOTOPE RATIO

Milestones 1 and 2 – Organise logistics and Organise samples:

- Urine samples were drawn from the samples available from the EPO2000 project.
- These have been sorted into ethnic groups and suitability such as volume of sample has been assessed.
- The selected samples are analysed using the routine analysis for steroids to determine levels of endogenous metabolites present.

Milestone 3 – Analyse samples:

- So far 1000 samples have been selected and extracted for Isotope Ratio analysis.
- The data from these are being collected in a database for analysis once the complete set is finished.
- Optimisation of procedures for diols has been investigated.
- Samples suitable for this diol analysis are being selected.

Overall the analysis of samples is continuing as planned to allow the statistical analysis later in the year.

Estimated Project Expenditure

Cost Item (detailed breakdown)	Total Approved for Item/Category (Excluding GST)	Amount Spent in Reporting period	Cumulative Amount Spent to Date
Salary, Scientific personnel 10% of SPOA plus on costs 50% of SPOC plus on costs for EPO 30% of PO2 plus on costs for CIR	 \$30,000 \$20,000	 \$22,747 \$13,125	 \$22,747 \$13,125
Salary, technical personnel 100% of technician plus on costs for EPO 30% of technician plus on costs for CIR	 \$48,000 \$13,000	 \$26,124 \$8,443	 \$26,124 \$8,443
Consumables EPO – approximately 60 gels at \$1,800 per gel. CIR – 1100 SPE extractions with GC/MS consumables	 \$90,000 \$33,353	 \$73,800 \$41,450	 \$73,800 \$41,450
Overheads: (indirect costs including support infrastructure, equipment maintenance and repairs, services etc.)	\$30,500	>\$30,500	>\$30,500
Sub-totals	\$264,853	>\$216,189	>\$216,189