Document Number:	TD2022LDOC	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

LABORATORY DOCUMENTATION PACKAGES

1.0 Introduction

This *Technical Document* (*TD*) and its annexes outline the requirements for the production of Laboratory Documentation Packages by Laboratories and *ABP* Laboratories, as applicable.

This *TD* includes instructions for producing Laboratory Documentation Packages for results from qualitative Test Methods (applied to Non-Threshold Substances) and quantitative Test Methods (applied to Threshold Substances and the determination of the *Markers* of the steroid profile), as well as for results of the analysis of *ABP* blood *Samples* (see Annex E).

This *TD* also includes the following annexes which list additional documentation that is required for specific analyses:

- Annex A: GC-MS^n for Urine *ABP* (applicable to the steroidal module of the *Athlete Biological Passport*);
- Annex B: GC/C/IRMS (applicable to analyses by Gas Chromatography /Combustion/Isotope Ratio Mass Spectrometry);
- Annex C: ERA (applicable to the analysis of EPO and other Erythropoietin Receptor Agonists (ERAs) using electrophoretic Analytical Methods);
- Annex D: hGH (applicable to the analysis of human Growth Hormone);
- Annex E: Blood *ABP* (applicable to the hematological module of the *Athlete Biological Passport*).

1.1. Production of Laboratory Documentation Packages by Laboratories

If requested by the *Testing* Authority (TA), *Results Management* Authority (RMA) or *WADA*, Laboratory Documentation Packages shall be provided by the Laboratory that reported the results supporting an *Adverse Analytical Finding* (*AAF*) or *Atypical Finding (ATF)*. Laboratories are not required to produce a Laboratory Documentation Package for a *Sample* reported as a Negative Finding, unless requested by a hearing body or disciplinary panel as part of a *Results Management* process or Laboratory disciplinary proceedings.

> *[Comment: Athletes shall only make requests for a Laboratory Documentation Package through the relevant TA or RMA.]*

A Laboratory Documentation Package shall be comprised of the information outlined below to support the result of the Laboratory's analysis of the relevant *Sample*. Laboratory working documents, computer printouts, and similar documents may be in the native language of the Laboratory. The table of contents,

Document Number:	TD2022LDOC	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

summaries and any flowcharts explaining the sequence of steps in the process and any other explanatory portions of the Laboratory Documentation Packages shall be provided at least in English.

The items outlined in this *TD* shall be the only information that the Laboratory includes in the Laboratory Documentation Package for the relevant analyses supporting the *AAF* or *ATF*. Therefore, the Laboratory is not required to provide any additional documentation, such as Standard Operating Procedures (SOP), general quality management documents (*e.g.*, ISO compliance documents), validation or External Quality Assessment Scheme (EQAS) data or any other data or document, in hardcopy or electronic format, not specifically required by this *TD*.

A Laboratory Documentation Package should be provided to the TA, RMA or *WADA* within the timelines stipulated in the *International Standard* for Laboratories (ISL) [1].

Laboratory Documentation Packages may be requested for "A" and "B" *Samples,* including all split portions of the *Sample*. However, Laboratory documents applicable to both "A" and "B" *Samples* (*e.g., Doping Control* Form (DCF), *Sample* receipt documentation, etc.) need only be provided once in the Laboratory Documentation Packages.

This *TD* sets forth formal requirements. Deviations from the requirements set forth herein shall not invalidate the *AAF*(s) or *ATF*(s).

2.0 Formatting Requirements

Laboratory Documentation Packages shall meet the following formatting requirements:

- A Table of Contents;
- Sequentially numbered pages;
- Presentation in a format that will allow proper review by relevant stakeholders such as clearly scanned documents, descriptors, etc. (annotations may be included by the Laboratory to assist interpretation);
- Information that appears on data and forms that refers to other *Samples* may be redacted by the Laboratory;
- Any adjustments to the records in the Laboratory Documentation Package shall be conducted as forensic corrections in accordance with ISO/IEC 17025;
- Data, charts, graphs, etc. shall be clearly described and presented.

  [Comment: Descriptions may be provided in the Table of Contents, page headers, titles, etc.; data and chart details shall be legible.]

Document Number:	TD2022LDOC	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

3.0 Laboratory Documentation Requirements

Laboratory Documentation Packages shall contain the following information:

3.1. Cover Page

- Identification of the Laboratory preparing the Laboratory Documentation Package, including the relevant *Sample* code and whether it is an "A" or a "B" *Sample*;
- A signed statement by the Laboratory Director or authorized delegate certifying that the Laboratory Documentation Package contains authentic copies of original data and forms;
- A declaration specifying that the Laboratory Documentation Package shall be handled as confidential information, shall not be disclosed to third parties or be reproduced or forwarded unless written approval is obtained from the Laboratory;
- A statement certifying that the *Sample* was analyzed according to the relevant *WADA* rules in force (*e.g.*, ISL, *TD*s);
- Any relevant comments.

3.2. Chain of Custody

- List of Laboratory staff involved in the analysis of the *Sample*, including signatures and/or initials and position title(s);

  [Comment: Each individual's complete signature/initials/name shall be provided to assist in the interpretation of the Laboratory Internal Chain of Custody documents.]

- The Laboratory version of the DCF related to the *Sample*. The *Sample's* external chain of custody form shall also be included if provided by the TA;
- The Laboratory's documentation of receipt of the *Sample*, including a declaration about any condition observed upon *Sample* receipt that may adversely impact the integrity of the *Sample* (in accordance with the ISL [1]*;*
- Documentation linking the *Sample* code (collection kit code) to the Laboratory identification code (if available);
- The relevant "A" and/or "B" *Sample* container Laboratory Internal Chain of Custody documentation (see TD LCOC [2]);
- Summary of the chain of custody which is supported by the Laboratory Internal Chain of Custody documentation provided.

Document Number:	TD2022LDOC	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

3.3. Analytical Data

3.3.1. Confirmation Procedure (CP) Data

CP method details to be provided within the documentation:

- SOP title or identification code of the CP method applied;
- Instrument type/identification code;
- Description of the composition of each positive quality control (PQC) sample(s) analyzed in the same batch;
- The monitored ions/transitions in the method for identification of the target Analyte(s) (for GC-MS^n and/or LC-MS^n procedures);
- "A" and/or "B" *Sample* Laboratory Internal Chain of Custody documentation for the CP relevant to the storage and handling of the *Sample* container (if not provided under 3.2 above);
- CP Aliquot Laboratory Internal Chain of Custody documentation [2];
- CP analytical instrument sequence file;

  [Comment: A copy of the original file (preferably generated by the analytical instrument software), which demonstrates the identification and order of analysis of each Sample analyzed in the CP.]

- CP chromatographic and spectral data (for GC-MS^n and/or LC-MS^n procedures):
  - Positive QC sample(s);
  - Negative QC sample(s); and
  - Aliquot(s) analyzed to conclude the *AAF(s)*;

    [Comment: The Laboratory shall demonstrate that the CP data is traceable to the Laboratory Internal Chain of Custody documentation. CP data shall be copies of the original data which was evaluated by the Laboratory to support the conclusion of an AAF or ATF.]

- For GC-MS^n and/or LC-MS^n procedures, identification data demonstrating compliance with the TD IDCR [3] including:
  - A summary table with relative abundances (RAs) of diagnostic ions, retention time (RT) data and relevant calculation results;

    [Comment: The Laboratory is not required to quantify or estimate a concentration for a Non-Threshold Substance not subject to a Minimum Reporting Level (MRL) [1].]

  - The applicable criteria utilized to identify the target Analyte(s) and report an *AAF or ATF*;
  - The summary table shall include signed/initialed (or electronic signature/validated LIMS record) statements that the results meet the applicable criteria.

    [Comment: For example, "Pass/Fail" as a statement of compliance with the relevant criteria.]

- Statement that there was no deviation from the CP SOP.

Document Number:	TD2022LDOC	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

[Comment: If a deviation exists (e.g., a change in the split ratio or a dilution of the derivatized Sample due to Sample overload in the instrument; application of an additional cleanup step; or an explanation for the re-analysis of the Sample with a new Aliquot) then documentation of the deviation(s) from the written CPs shall be provided.]

- A signed and dated statement of acceptable performance based on the evaluation of the analytical instrument which was used to generate the *Sample*'s CP data.

[Comment: For example: "Instrument [identification] meets performance criteria based on the Laboratory SOP and QC data". This statement shall be signed and dated by the operator performing the evaluation.]

3.3.2. Additional Documentation for Non-Threshold Substances with an *MRL* only

A summary of the method used to estimate the concentration of target Analyte(s) of Non-Threshold Substances with an *MRL* (see TD MRPL [13]).

[Comment: The estimation of concentration for Non-Threshold Substances with an MRL shall only be conducted in the "A" CP (TD MRPL [13]) in order to report an AAF or ATF.]

- CP chromatographic and spectral data for:
  - The internal standard;
  - The single-point calibrator;
  - The independent Quality Control (QC) sample; and
  - The *Sample* Aliquot.
- Summary table that includes the calculation to estimate the concentration for the target Analyte(s);
- The confirmed urine Specific Gravity (SG). If an adjustment for SG is necessary (for SG > 1.018) [13], then the resulting adjusted concentrations shall be provided.

3.3.3. Additional Documentation for Quantitative CP Methods only (Threshold Substances)

A summary of the quantitative data for the Threshold Substance(s) (see TD DL [4] or applicable *TD* [5, 7-9] or Laboratory Guidelines [6]), including:

[Comment:

- *For those Threshold Substances of exogenous origin, which are analyzed by chromatography-based Analytical Methods, reporting requirements are specified in the TD DL [4]. For the "B" Sample confirmation of exogenous Threshold Substances [4], a quantitative CP is not necessary [1]. In such cases, the Laboratory shall only establish the presence (i.e., the identity) of the Threshold Substance or its Metabolite(s) or Marker(s) in the "B" Sample in accordance with the TD IDCR [3].*
- *For endogenous Threshold Substances (human Growth Hormone - hGH, human Chorionic Gonadotropin - hCG), these requirements are included in specific TDs or Laboratory Guidelines (TD GH [5], Laboratory Guidelines on hGH Biomarkers Test [6] and Annex D of this TD for hGH; TD CG/LH [7] for hCG). For the "B" Sample confirmation of endogenous Threshold Substances, the quantitative CP shall establish that the identified Threshold Substance or its Metabolite(s) or Marker(s) is present in the "B" Sample at a concentration and/or ratio and/or score of measured analytical values greater than (>) the*

Document Number:	TD2022LDOC	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

Threshold, and/or that the Threshold Substance or its Metabolite(s) or Marker(s) is of exogenous origin[1].

- *For other quantitative CPs, such as GC-MS^n for the Markers of the urinary steroid profile or GC/C/IRMS analysis, details are provided in the TD EAAS [8] and TD IRMS [9] and in Annexes A and B, respectively, of this TD.]*

- The calibration curve;
- The mean concentration (or ratio or score) from triplicate (3x) determinations as well as the individual concentrations determined for all the *Sample* Aliquots determined with appropriate units (as applicable);
- The nominal and measured concentrations of the QC sample(s) in addition to the acceptance criteria with a statement that the QC(s) test results pass the acceptance criteria;
- The Laboratory result for the Threshold Substance in the *Sample* (units), as the mean value from triplicate determinations;
- The confirmed urine SG. If an adjustment for SG is necessary (for SG > 1.018), then the resulting adjusted *Decision Limit* (DL_{adj}) [4] shall be provided;
- The Measurement Uncertainty (MU) details:
  - A statement that the relative u_c (%) for results at levels close to the Threshold does not exceed the maximum permissible relative u_{c_Max}(%) in Table 1 of the TD DL [4] or applicable *TD* [5, 7-9] or Laboratory Guidelines [6].

[Comment: The summary table provided shall compile the necessary data and applicable criteria utilized to evaluate the quantitative results obtained for the target Analyte(s) in order to report an AAF or ATF.]

3.4. Laboratory Test Report(s)

Laboratory Documentation Packages shall include the Laboratory (*ADAMS*) Test Report(s) including the relevant Laboratory Test Report(s) from the Laboratory which performed subcontracted analyses, if applicable.

[Comment: In the case of quantitative CPs, the ADAMS Test Report shall include details in compliance with the TD DL [4] or applicable TD [5, 7-9] or Laboratory Guidelines [6].]

[1] For endogenous Threshold Substances, the Threshold values have been established based on reference population statistics, and already incorporate the Measurement Uncertainty. Therefore, the Threshold constitutes the *DL*.

Document Number:	TD2022LDOC	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

3.5. Subcontracted Analysis

If an *AAF* or an *ATF* resulted (in whole or in part) from a subcontracted analysis, then the subcontracted Laboratory shall provide the documentation (as described in this *TD*) to the Laboratory (which subcontracted the analysis and reported the result into *ADAMS*) for the preparation of the Laboratory Documentation Package for the TA, RMA or *WADA*. The Laboratory Documentation Package shall clearly describe the steps conducted by each Laboratory.

Document Number:	TD2022LDOC–Annex A: Urine *ABP*	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

Urine LABORATORY DOCUMENTATION PACKAGE for the GC-MS^n CP of the Steroid Profile *Markers*

and

Urine LABORATORY CERTIFICATE OF ANALYSIS for the GC-MS^n ITP of the Steroid Profile *Markers*

The requirements of this Annex of the TD2022LDOC are relevant to Laboratories analyzing urine *Samples* in support of the steroidal module of the *Athlete Biological Passport* (*ABP*).

This Annex of TD2022LDOC outlines the requirements for the production of a Urine Laboratory Documentation Package for the *ABP* or a Urine Laboratory Certificate of Analysis for the *ABP*. The Laboratory may be requested by the relevant *Athlete* Passport Management Unit (APMU), Expert Panel or *WADA* to provide these types of documentation to support an *Adverse Passport Finding (APF)*.

> *[Comment: Athletes shall only make requests for a Urine ABP Laboratory Documentation Package or a Urine ABP Laboratory Certificate of Analysis through the relevant Testing Authority or Results Management Authority.]*

It is only mandatory to have a Urine *ABP* Laboratory Documentation Package for those confirmed test results that are deemed essential by the APMU or Expert Panel. Laboratories are not required to produce a Urine *ABP* Laboratory Documentation Package for ITP results of a *Sample* that is judged to confirm the baseline level of a steroid *Marker* by an APMU or Expert Panel. In such case, Laboratories shall provide a Urine *ABP* Laboratory Certificate of Analysis, in accordance with the requirements indicated in Article 3 of this *TD* Annex A, upon request by an APMU or Expert Panel.

Deviations from this *TD* Annex A shall not invalidate an *APF*.

1.0 Formatting Requirements

A Urine *ABP* Laboratory Documentation Package shall meet the formatting requirements as detailed in Article 2.0 of the TD2022LDOC.

2.0 Urine *ABP* Laboratory Documentation Package Requirements

2.1 Cover Page

The cover page shall meet the requirements detailed in Article 3.1 of the TD2022LDOC.

2.2 Chain of Custody

The chain of custody documentation shall meet the requirements detailed in Article 3.2 of the TD2022LDOC and the TD LCOC [2].

2.3 GC-MS^n Confirmation Procedure (CP) data

- A general description of the CP method details *(e.g.,* scheme/sequence of key analysis steps*)*, including:
  - o Standard Operating Procedure (SOP) title or identification code of the CP method applied;
  - o Instrument type/Identification code;
  - o Description of quality control (QC) sample(s) analyzed in the same batch;
  - o The monitored ions/transitions in the method for identification of the target Analyte(s).
- "A" and/or "B" *Sample* Laboratory Internal Chain of Custody documentation [2] for the CP, which is relevant to the storage and handling of the *Sample* container (if not provided under 2.2 above);
- CP Aliquot Laboratory Internal Chain of Custody documentation [2];
- CP analytical instrument sequence file;

  [Comment: A copy of the original sequence file (preferably generated by the analytical instrument software), which demonstrates the identification and order of analysis of each Sample analyzed in the Confirmation Procedure.]
- *Sample* preparation details:
  - o Data on controlling for efficiency of hydrolysis;
  - o Data on controlling for completeness of derivatization.
- CP GC-MS^n chromatographic and spectral data:

  [Comment: CP data shall be copies of the original data which were evaluated by the Laboratory to support the conclusion of an APF.]

  o Calibration curve or concentrations of the calibration standards for all confirmed *Markers* of the steroid profile;

  o Clearly integrated chromatograms for the relevant *Markers* of the steroid profile and their respective (deuterated) Internal Standards;

  o Identification data of the chromatographic peaks of the relevant *Markers* demonstrating compliance with the TD IDCR [3], including:
    - QC sample(s);
    - *Sample*;
    - A summary table with relative abundances (RAs) of diagnostic ions, retention time (RT) data and relevant calculation results;
    - The applicable criteria utilized to identify the target *Marker*(s);

  [Comment: It is not necessary to perform the GC-MS^n confirmatory identification of the steroid Markers twice, both during the initial GC-MS^n confirmation and during the subsequent GC/C/IRMS analysis. However, the identification of the steroid Markers (i.e., target compounds) is still mandatory prior to reporting an Adverse

Analytical Finding (AAF) or an Atypical Finding (ATF) based on GC/C/IRMS results (see TD EAAS [8] and TD IRMS [9]). The confirmatory identification of the Markers during the initial confirmation by GC-MS^n becomes relevant when advancing an Adverse Passport Finding (APF) based on the altered values (concentrations, ratios) of the Markers without a corroborative positive GC/C/IRMS result).]

- The summary table shall include signed/initialed statements (or electronic signature/validated LIMS record) that the results meet the applicable criteria.

[Comment: For example, "Pass/Fail" as a statement of compliance with relevant criteria.]

- "A" *Sample* GC-MS^n (n ≥ 1) CP:
  - o Confirmed SG of the "A" *Sample*;
  - o Confirmed values of the *Markers* of the steroid profile for:
    - QC sample(s); and
    - *Sample*;

    [Comment: ADAMS printout of Sample record containing this information may be provided to address this requirement.

    In addition, the acceptance criteria for the concentrations of the Markers in the QC(s) shall be provided with a statement that the QC(s) test results pass the acceptance criteria.]
  - o The associated u_c expressed in units;
  - o Statement that the associated u_c (%) for the *Markers* of the steroid profile does not exceed the maximum permissible relative u_{c_Max} (%) specified in the TD EAAS [8];
  - o Confirmed values of:
    - 5α-androstanedione (5αAND) concentration; and/or
    - 5β-androstanedione (5βAND) concentration, and
    - ratio of 5αAND/A; and/or
    - ratio of 5βAND/Etio;
    - ratio of T_{free}/T_{total}.

    [Comment: the steroid ratios specified above shall be as determined from the respective steroid concentrations (and not as ratios of chromatographic peaks or areas).]
  - o Confirmation results about the presence/absence of substance(s) that may alter the steroid profile, including reporting the estimated concentrations of:
    - ethyl-glucuronide (if ≥ 5 µg/mL),
    - carboxy-finasteride (if ≥ 5 ng/mL),
    - 4-hydroxy- and/or 6-hydroxy-dustasteride (if ≥ 5 ng/mL),
    - ketoconazole (if ≥ 100 ng/mL),

- fluconazole (if ≥ 500 ng/mL), and
- miconazole (if ≥ 1,000 ng/mL).

- "B" *Sample* GC-MS^n (n ≥ 1) CP:

  o Confirmed SG of the "B" *Sample*;

  o If the "A" *Sample* has not been reported as an *AAF* for the *Marker*(s) of the steroid profile based on the results of the GC/C/IRMS analysis, but the steroid profile CP by GC-MS^n has been requested for the "B" *Sample*, then the Laboratory shall include the results of the "B" GC-MS^n confirmation of the steroid profile as described for the "A" *Sample*.

- Statement that there was no deviation from the CP SOP.

  [Comment: If a deviation exists (for example, a change in the split ratio or a dilution of the derivatized Sample due to Sample overload in the instrument; application of an additional cleanup step; or an explanation for the re-analysis of the Sample with a new Aliquot) then documentation of the deviation(s) from the written CPs shall be provided.]

- Statement of acceptable performance based on the evaluation of the analytical instrument which was used to generate the *Sample*'s CP data.

  [Comment: For example: "Instrument [identification] meets performance criteria based on the Laboratory SOP and QC data". This statement shall be signed and dated by the analyst performing the evaluation.]

3.0 Urine *ABP* Laboratory Certificate of Analysis Requirements

A Urine *ABP* Laboratory Certificate of Analysis shall only contain the following information:

3.1. Cover Page

A signed and dated document by the Laboratory Director or authorized delegate including:

- Identification of the Laboratory preparing the Urine *ABP* Laboratory Certificate of Analysis, including the relevant *Sample* code;
- A statement certifying that the Urine *ABP* Laboratory Certificate of Analysis contains authentic copies of original data and forms;
- A statement specifying that the Urine *ABP* Laboratory Certificate of Analysis shall be handled as confidential information, which shall not be disclosed to third parties and shall not be reproduced or forwarded unless written approval is obtained from the Laboratory;
- A statement certifying that the *Sample* was analyzed according to the relevant *WADA* rules in force (*e.g.*, ISL, *TD*s);
- Any relevant comments.

3.2. ITP GC-MS^n Data

The ITP GC-MS^n analysis of the *Sample* steroid profile, including

- SG of the "A" *Sample*;
- Chromatographic printout for all *Markers* of the steroid profile;
- The measured values of the *Markers* of the steroid profile;
- The associated u_c expressed in units;
- The presence or absence in the *Sample* of substance(s) that may alter the steroid profile (see TD EAAS [8]);
- *Sample* code;
- Analysis date and time;
- Instrument identification code.

Document Number:	TD2022LDOC–Annex B: GC/C/IRMS	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

LABORATORY DOCUMENTATION PACKAGE FOR GC/C/IRMS ANALYSIS

This Annex of the TD2022LDOC includes instructions for producing Laboratory Documentation Packages for confirmatory analysis results supporting an *Adverse Analytical Finding* (*AAF*) or an *Atypical Finding* (*ATF*) based on the application of Gas Chromatography/Combustion/Isotope Ratio Mass Spectrometry (GC/C/IRMS).

1.0 Formatting Requirements

A GC/C/IRMS Laboratory Documentation Package shall meet the formatting requirements detailed in Article 2.0 of the TD2022LDOC.

2.0 Laboratory Documentation

2.1. Chain of Custody

The chain of custody documentation shall meet the requirements detailed in Article 3.2 of the TD2022LDOC and the TD LCOC [2].

2.2. Confirmation Procedure Analytical data

- If an adjustment for SG is necessary (for SG > 1.018) [9, 10], then the SG of the *Sample* and the resulting adjusted concentration of the Target Compound (TC) shall be provided;
- Analysis description (*e.g.,* scheme/sequence of key analysis steps);
- *Sample* preparation:
  - Documentation demonstrating the order of sequence injection;
  - Statement on the verification of retention time (RT) stability.
- GC/C/IRMS analysis:
  - Data on CO_2 pulses stability test and statement on when the linearity signal was checked last;
  - CP analytical instrument sequence file;

    [Comment: A copy of the file (preferably generated by the analytical instrument software) which demonstrates the order of analysis of each Sample in the CP.]
  - GC/C/IRMS Test Results for relevant Target Compounds (TCs) (which produced the *AAF* or *ATF*) and Endogenous Reference Compounds (ERCs), including:
    - Chromatograms with the integration and annotation of the peaks;
    - $\delta^{13}C$ values (before and after correction for acetylation, if applicable); and
    - $|\Delta\delta^{13}C|$ values.

These results shall be produced for:

- The Reference Material (RM);
  - The acceptance criteria for the $\delta^{13}C$ determinations of the TCs and ERCs in the RM shall be provided;
  - It shall be stated whether the RM test results pass the acceptance criteria.
- The negative (QCN) and positive quality control (QCP) samples;
  - The acceptance criteria for the $\delta^{13}C$ determinations of the TCs and ERC in the QC samples shall be provided;
  - It shall be stated whether the QC test results pass the acceptance criteria.
- The *Sample*
  - Summary of results: Worksheet with $\delta^{13}C$ values, associated u_c (expressed in ‰) and $|\Delta\delta^{13}C|$ values for the relevant TCs and ERCs.

- GC-MS analysis

  o Mass spectrum of each relevant TC and ERC (average and not apex) in the *Sample* and a comparison with mass spectrum obtained from a reference preparation;

  o Proof of identification of the peaks of the relevant TC(s) and ERCs in accordance with TD IDCR [3] requirements;
    - A summary table with RAs of diagnostic ions, RT data and relevant calculation results;
    - The applicable criteria utilized to identify the target Analyte(s);
    - The summary table shall include signed/initialed (or electronic signature/validated LIMS record) statements that the results meet the applicable criteria.

      [Comment: For example, "Pass/Fail" as a statement of compliance with the relevant criteria.]

  o A statement about steroid peak purity.
- A statement on the criteria that were fulfilled, as per the TD IRMS [9], to report an *AAF*.

  [Comment: the TD IRMS criteria to report an AAF may be found in the ADAMS Test Report.]
- Second Opinion (if requested).

2.3. Laboratory Test Report(s)

The Test Report documentation as detailed in Article 3.4 of the TD2022LDOC and the TD IRMS [9].

2.4. Subcontracted Analysis

A subcontracted analysis shall meet the requirements detailed in Article 3.5 of the TD2022LDOC.

Document Number:	TD2022LDOC–Annex C: ERA	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

LABORATORY DOCUMENTATION PACKAGE FOR ERA ANALYSIS BY ELECTROPHORETIC ANALYTICAL METHODS

This Annex of the TD2022LDOC includes instructions for producing Laboratory Documentation Packages for results supporting an *Adverse Analytical Finding* (*AAF*) or *Atypical Finding* (*ATF*) reported for Erythropoietin Receptor Agonists (ERAs) when using polyacrylamide gel electrophoretic (PAGE) Analytical Methods.

[Comment: Erythropoietin Receptor Agonists (ERAs), as defined in the Prohibited List, include erythropoietin and its analogs and mimetics. These substances were previously known by the name of Erythropoiesis Stimulating Agents (ESA). Their analysis is covered in the TD EPO[11].]

1.0 Formatting Requirements

An ERA Laboratory Documentation Package shall meet the formatting requirements as detailed in Article 2.0 of the TD2022LDOC.

2.0 Laboratory Documentation

2.1. Chain of Custody

The chain of custody shall meet the requirements detailed in Article 3.2 of the TD2022LDOC and the TD LCOC [2].

2.2. Analytical data

2.2.1. Initial *Testing* Procedure (ITP)

Provision of the ITP data is optional (at the Laboratory's discretion):

- Test description

  [Comment: For example, description of the key steps in the IEF-PAGE or SAR-/SDS-PAGE procedure, including method used for ERA immunopurification.]

- *Sample* sequence description (content and lane position on the gel);
- ITP results including gel images and report (*e.g.,* GASepo Analysis Report) on:
  - Negative control sample (QCN);
  - Reference standard solutions used to define basic, acidic and endogenous areas in IEF-PAGE or apparent molecular mass in SAR-PAGE and SDS-PAGE;
  - Test sensitivity controls (if applicable); and
  - *Sample* Aliquot.

Document Number:	TD2022LDOC–Annex C: ERA	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

- Statement on quality control, instrument operation and other test validity data.

  [Comment: For example, "The overall system performance is demonstrated by the quality control samples of the ITP. It is considered to be valid for the entire procedure".]

- Conclusion from ITP

  [Comment: For example, "The band in Sample x shows a faint, diffuse area above the corresponding endogenous band on the SAR-PAGE gel; therefore, the presence of recombinant EPO cannot be excluded. Consequently, this result is considered a Presumptive Adverse Analytical Finding and the Sample shall be subjected to a Confirmation Procedure".]

2.2.2. Confirmation Procedure (CP)

- Test Description

  [Comment: For example, description of the key steps in the SAR-PAGE procedure, including method used for ERA immunopurification.]

- *Sample* sequence description (content and lane position on the gel);
- Confirmation results including gel images and report (*e.g.,* GASepo Analysis Report) on:
  - Negative control sample (QCN);
  - Positive control sample(s) (QCP);
  - Reference standard solution(s) used to define basic, acidic and endogenous areas in IEF-PAGE or apparent molecular mass in SDS-PAGE and SAR-PAGE;
  - Test sensitivity control(s) (if applicable); and
  - *Sample* Aliquot.
- Statement on quality control, instrument operation and other test validity data.

  [Comment: For example, "The overall system performance is demonstrated by the positive and negative control samples of the Confirmation Procedure. It is considered to be valid for the entire procedure".]

- Conclusion from CP.

  [Comment: For example, "The band in Sample x shows a faint, diffuse area above the corresponding band for endogenous EPO on the SAR-PAGE gel; therefore, the presence of recombinant EPO is confirmed according with the WADA TD EPO [11]. Consequently, a second opinion for this Sample shall be requested".]

- Second Opinion (signed by a member of the *WADA* EPO Working Group (see TD EPO [11]).

Document Number:	TD2022LDOC–Annex C: ERA	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

2.2.3. Additional Analyses to Determine rEPO Findings

2.2.3.1 ERA Analysis on Blood *Samples* for VAR-EPO [11]

When there is a finding for rEPO in urine or blood *Samples* requiring further investigation under Annex B of the TD EPO and other blood *Samples* from the *Athlete* are analyzed for ERAs to establish whether the *Athlete* is a carrier of the *EPO* c.577del variant (see TD EPO [11]), the Laboratory shall include *WADA*'s written instructions on how to report the finding under investigation (based on the ERA blood results) in the Laboratory Documentation Package.

2.2.3.2 DNA Analysis

If necessary, a DNA analysis targeting the *EPO* gene (exon 5 or region encompassing c.577) in blood *Samples* shall be conducted (as described in the TD EPO [11]) and the test results included in the Laboratory Documentation Package, including:

- DNA Analysis Test Description

  [Comment: For example, description of the DNA sequencing platform (e.g., Sanger) and the key steps in the DNA Analysis procedure.]

- Description of the *Sample* subjected to DNA analysis (*Sample* code, *Testing* Authority, Date of Collection, matrix *e.g.,* whole blood/serum/plasma) if different from the *Sample* under investigation;
- DNA sequencing analysis images and results (or copy of DNA test report):
  - Quality Control sample(s);
  - *Sample* Aliquot.
- DNA Analysis Test Report with conclusion.

  The DNA Analysis Test Report shall conclude on whether or not the blood *Sample* tested indicates that the associated *Athlete* is a carrier of the *EPO* c.577del variant.

  [Comment: For example, "The EPO sequencing results conclude that the Athlete that provided the blood Sample tested is a carrier of the EPO c.577del variant" or "The EPO sequencing results conclude that the Athlete that provided the blood Sample tested is not a carrier of the EPO c.577del variant".]

- *WADA*'s written instructions on how to report the finding under investigation (based on the results of the DNA analysis).

2.3. Laboratory Test Report(s)

The Test Report documentation as detailed in Article 3.4 of the TD2022LDOC and the TD EPO [11].

2.4. Subcontracted Analysis

A subcontracted analysis shall meet the requirements detailed in Article 3.5 of the TD2022LDOC.

Document Number:	TD2022LDOC–Annex D: hGH	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

LABORATORY DOCUMENTATION PACKAGE FOR hGH ANALYSIS

This Annex of the TD2022LDOC includes instructions for producing Laboratory Documentation Packages for Confirmation Procedure (CP) results supporting an *Adverse Analytical Finding* (*AAF*) or an *Atypical Finding* (*ATF*) reported for human Growth Hormone (hGH).

1.0 Formatting Requirements

An hGH Laboratory Documentation Package shall meet the formatting requirements as detailed in Article 2.0 of the TD2022LDOC.

2.0 Laboratory Documentation

2.1. Chain of Custody

The chain of custody documentation shall meet the requirements detailed in Article 3.2 of the TD2022LDOC and the TD LCOC [2].

2.2. CP Analytical Data

- Summary test description, including
  - Scheme/sequence of key analysis steps;
  - Kit lot numbers if applying the Isoforms Test;
  - IGF-I and P-III-NP assay pairs and kit lot numbers if applying the Biomarkers Test.
- Statement of acceptable performance based on the evaluation of the analytical instrument, which was used to generate the *Sample*'s CP data.

  [Comment: For example: "Instrument [identification] meets performance criteria based on the Laboratory SOP and QC data". This statement shall be signed and dated by the analyst performing the evaluation.]
- Assays' calibration curve;
- Sequence of analysis;
- Test data for negative (QCN) and positive quality control (QCP) sample(s) and *Sample,* including:
  - Isoforms Test [5]
    - The REC and PIT concentrations, expressed to three (3) decimal places, for the three (3) *Sample* Aliquots analyzed using kit-1 and kit-2;
    - The mean concentrations from the triplicate determinations expressed to three (3) decimal places;
    - The Relative Standard Deviation (RSD, %) of the triplicate determinations;

- The resulting REC/PIT ratios (ratio-1; ratio-2), expressed to two (2) decimal places, calculated from the corresponding mean REC and PIT concentrations from the triplicate determinations;
- The applicable (kit, gender of the *Athlete*) *Decision Limit*(s) (*DL*); and
- The u_c (%) at values close to the *DL* as determined by the Laboratory during method validation.

o Biomarkers Test [6]

- The IGF-I and P-III-NP concentrations (expressed to the nearest integer for IGF-I and two decimal places for P-III-NP) for the three (3) *Sample* Aliquots analyzed with two (2) different IGF-I / P-III-NP assay pair combinations;
- The mean concentrations from the triplicate determinations (expressed to the nearest integer for IGF-I and two decimal places for P-III-NP);

[Comment: When the bottom-up LC-MS/MS or LC-HRMS method is used for IGF-I quantification during the CP, the Laboratory shall report the IGF-I concentrations (triplicate determinations, mean concentration) determined from the quantification of T1 and T2 peptides, as well as the calculated difference between these mean (T1, T2) concentrations. The Laboratory shall also report the average (overall) IGF-I concentration determined from the quantification of T1 and T2 [6].]

- The GH-2000 scores, expressed to two (2) decimal places, calculated from the natural logarithms (ln) of the mean concentrations (ng/mL) of IGF-I and P-III-NP;

[Comment: When the bottom-up LC-MS/MS or LC-HRMS method is used for IGF-I quantification during the CP, the GH-2000 score is calculated from the natural logarithm (ln) of the average (overall) concentration (ng/mL) of IGF-I determined from the quantification of T1 and T2 [6].]

- The *applicable DL*(s) (assay pair, gender of the *Athlete*); and
- The u_c at values close to the *DL* as determined by the Laboratory during method validation.

- The acceptance criteria for the concentrations and ratios/scores of each QC sample, and a statement on whether the QC test results passed the acceptance criteria.

2.3. Laboratory Test Report(s)

- Laboratory Test Report from *ADAMS,* including the conclusion from the CP;

Example Isoforms Test [5]:

"The analysis of the *Sample* using the hGH differential immunoassays has produced the following analytical values of assay ratios: 2.52 for kit "1" and 2.40 for kit "2", which are greater than the corresponding *DL*s of 1.84 and 1.91, respectively. The relative combined standard uncertainty (u_c, %) estimated by the Laboratory at levels close to the *DL* is 15% for kit "1" and 17% for kit "2". This constitutes an *Adverse Analytical Finding* for hGH".

Document Number:	TD2022LDOC–Annex D: hGH	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

Example Biomarkers Test [6]:

"The analysis of the *Sample* with the hGH Biomarkers Test has produced the following GH-2000 scores: 10.90 for assay pair '1' [IDS IGF-I + Centaur P-III-NP] and 9.90 for assay pair '2' [LC-MS/MS IGF-I + Orion P-III-NP], which are greater than the corresponding male-specific *DL*s of 10.61 and 9.70, respectively. The combined standard uncertainty (u_c) estimated by the Laboratory at levels close to the *DL* is 0.40 for assay pair '1' and 0.35 for assay pair '2'. This constitutes an *Adverse Analytical Finding* for hGH".

- Relevant Laboratory Test Report(s) from subcontracted analyses, if any.

2.4. Subcontracted Analysis

A subcontracted analysis shall meet the requirements detailed in Article 3.5 of the TD2022LDOC.

Document Number:	TD2022LDOC–Annex E: Blood ABP	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

Blood *ABP* LABORATORY DOCUMENTATION PACKAGE and Blood *ABP* LABORATORY CERTIFICATE OF ANALYSIS

The requirements of this Annex of the TD2022LDOC are relevant to *ABP* blood *Samples* analyzed in support of the hematological module of the *Athlete Biological Passport* (*ABP*).

This *TD* Annex outlines the requirements for the production of a Blood *ABP* Laboratory Documentation Package or a Blood *ABP* Laboratory Certificate of Analysis. The Laboratory or *ABP* Laboratory may be requested by the relevant *Athlete* Passport Management Unit (APMU), Expert Panel or *WADA* to provide these types of documentation to support an *Adverse Passport Finding (APF).*

> *[Comment: Athletes shall only make requests for a Blood ABP Laboratory Documentation Package or a Blood ABP Laboratory Certificate of Analysis through the relevant Testing Authority or Results Management Authority.]*

It is only mandatory to have a Blood *ABP* Laboratory Documentation Package for those test results that are deemed essential by the APMU or Expert Panel. Laboratories and *ABP* Laboratories are not required to produce a Blood *ABP* Laboratory Documentation Package for a *ABP* blood *Sample* that is judged to confirm the baseline level of a blood *Marker* by an APMU or Expert Panel. In such case, Laboratories and *ABP* Laboratories shall provide a Blood *ABP* Laboratory Certificate of Analysis in accordance with the requirements as indicated in Article 3.0 of this *TD* Annex, upon request by an APMU or Expert Panel.

Deviations from this *TD* Annex shall not invalidate the blood *APF*.

1.0 Formatting Requirements

A Blood *ABP* Laboratory Documentation Package shall meet the formatting requirements as detailed in Article 2.0 of the TD2022LDOC.

2.0 Blood *ABP* Laboratory Documentation Package Requirements

2.1. Cover Page

The cover page shall meet the requirements detailed in Article 3.1 of the TD2022LDOC.

2.2. A copy of the *ABP* blood *Sample*'s temperature data logger report (if the report associated with the *ABP* blood *Sample* result is not submitted in *ADAMS*).

2.3. Chain of Custody

The chain of custody documentation shall meet the requirements detailed in Article 3.2 of the TD2022LDOC and the TD LCOC [2].

Document Number:	TD2022LDOC–Annex E: Blood ABP	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

2.4. Analytical Data

- Original Sysmex printouts of all *ABP* blood *Sample* full blood count and scattergrams, including:
  - *ABP* blood *Sample* code;
  - Analysis date and time; and
  - Instrument identification and serial number.
- *ABP* blood *Sample* and XN-checks (levels 1, 2 and 3) quality control (QC) results summary table, including:
  - Results of all *ABP* blood *Sample* analyses (minimum two);
  - All XN-check QC levels from the same batch as the *ABP* blood *Sample*;
  - Acceptance criteria; and
  - Statements of acceptance.

  [Comment: The summary table provided shall compile the necessary data and applicable criteria as per the TD BAR [12].]
- XN-CHECK manufacturer assay sheets for each QC level; and
- *ADAMS* record printout which contains:
  - Date and time of submission of the results into *ADAMS*;
  - Date and time of *ABP* blood *Sample* reception;
  - Date and time of *ABP* blood *Sample* analysis;
  - Sport/discipline;
  - *Testing* Authority (TA), *Results Management* Authority (RMA), *Sample* Collection Authority (SCA); and
  - Reported test results for the blood *Markers* of the *ABP* blood *Sample*.

3.0 Blood *ABP* Laboratory Certificate of Analysis Requirements

A Blood *ABP* Laboratory Certificate of Analysis shall only contain the following information:

3.1. Cover Page

A signed and dated document by the Laboratory Director or the Director of the *ABP* Laboratory or authorized delegate including:

- Identification of the Laboratory or the *ABP* Laboratory preparing the Blood *ABP* Laboratory Certificate of Analysis;
- The relevant *ABP* blood *Sample* code;

- A statement certifying that the Blood *ABP* Laboratory Certificate of Analysis contains authentic copies of original data and forms;
- A statement specifying that the Blood *ABP* Laboratory Certificate of Analysis shall be handled as confidential information which shall not be disclosed to third parties and shall not be reproduced or forwarded unless written approval is obtained from the Laboratory or the *ABP* Laboratory;
- A declaration certifying that the *ABP* blood *Sample* was analyzed according to the relevant *WADA* rules in force (*e.g.*, ISL, *TD*s); and
- Any relevant comments.

3.2. Original Sysmex Printout

The original instrument printouts of the accepted and reported *ABP* blood *Sample* analysis, including:

- Full blood count and scattergram;
- *ABP* blood *Sample* code;
- Analysis date and time; and
- Instrument identification and serial number.

Document Number:	TD2022LDOC-References	Version Number:	1.0
Written by: Reviewed by:	*WADA* Science *WADA* Laboratory Expert Advisory Group	Approved by:	*WADA* Executive Committee
Date:	24 November 2021	Effective Date:	1 January 2022

REFERENCES

[1] The World Anti-Doping *Code International Standard* for Laboratories (ISL).

[2] *WADA Technical Document* TD LCOC: Laboratory Internal Chain of Custody.

[3] *WADA Technical Document* TD IDCR: Minimum Criteria for Chromatographic-Mass Spectrometric Confirmation of the Identity of Analytes for *Doping Control* Purposes.

[4] *WADA Technical Document* TD DL: *Decision Limits* for the Confirmatory Quantification of Exogenous Threshold Substances by Chromatography-based Analytical Methods.

[5] *WADA Technical Document* TD GH: Human Growth Hormone (hGH) Isoform Differential Immunoassays for *Doping Control* Analyses.

[6] *WADA* Laboratory Guidelines on human Growth Hormone Biomarkers Test for *Doping Control* Analyses.

[7] *WADA Technical Document* TD CG/LH: Reporting and Management of Urinary Chorionic Gonadotrophin (hCG) and Luteinizing Hormone (LH) Findings in Male *Athletes*.

[8] *WADA Technical Document* TD EAAS: Measurement and Reporting of Endogenous Anabolic Androgenic Steroid (EAAS) *Markers* of the Urinary Steroid Profile.

[9] *WADA Technical Document* TD IRMS: Detection of Synthetic Forms of *Prohibited Substances* by GC/C/IRMS.

[10] *WADA Technical Document* TD NA: Harmonization of Analysis and Reporting of 19-Norsteroids Related to Nandrolone.

[11] *WADA Technical Document* TD EPO: Harmonization of Analysis and Reporting of EPO and other Erythropoietin Receptor Agonists (ERAs) by Polyacrilamide Gel Electrophoretic (PAGE) Analytical Methods.

[12] *WADA Technical Document* TD BAR: Blood Analytical Requirements *Athlete Biological Passport* Operating Guidelines & Compilation of Required Elements.

[13] *WADA Technical Document* TD MRPL: Minimum Required Performance Levels and Applicable *Minimum Reporting Levels* for Non-Threshold Substances Analyzed by Chromatographic - Mass Spectrometric Analytical Methods.

[Current versions of WADA ISL, Technical Documents and Laboratory Guidelines may be found at https://www.wada-ama.org/en/what-we-do/science-medical/laboratories]