European Pharmacopoeia Training Session on Biologicals
7-8 February 2017, Strasbourg

A guide through monograph sections with emphasis on synthetic peptides

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Basis for the elaboration of monographs

➢ Products of proven safety, evaluated and approved by competent authorities of Member States;
➢ Impurity profiles for existing, approved synthetic routes;
➢ Use of robust, validated analytical methods;
➢ Technical guides
Individual and General monographs are complementary

Synthetic peptides

- Product of chemical synthesis;
- Molecular mass typically below 5000 Da;
- May contain chemical structures that do not occur naturally in proteins or peptides;
- Generally sufficiently characterised by physico-chemical tests; bioassay not necessary.
Ph. Eur. portfolio for synthetic peptides

- (0471) Calcitonin (salmon)
- (0644) Tetracosactide *
- (0712) Desmopressin
- (0779) Oxytocin conc. solution
- (0780) Oxytocin *
- (0827) Gonadorelin acetate *
- (0949) Somatostatin *

In development:

- (2414) Octreotide (Phpa 28.4)
- (3054) Atosiban
- (3055) Triptorelin
- (3056) Lanreotide
- (3057) Glatiramer

* under revision

Synthetic peptide

**MONOGRAPH SECTION**

**Title**

**INN**

**Formula**

molecular and graphic

**Relative mass CAS number**

**Definition**

- chemical nomenclature
- salt form
- additives (e.g. oxytocin)*
- assay limits:
  - content (anhydrous, acetic acid-free basis)
  - potency (IU/mg) (synthetic peptides: by convention if present e.g. oxytocin, tetracosactide, calcitonin)

**Definition**

Solution of a protein having the primary structure of the hormone or fragment considering the presence of additional amino acid(s) not normally present in the natural hormone (monograph). The protein stimulates the differentiation and proliferation of hormone-targeted cells into hormone-responsive cells.

Content: minimum 10 mg of protein per milligram.

Potency: maximum 0.9 x 10^6 IU per milligram of protein.

* Substances for Pharmaceutical Use (2034): "A monograph is applicable to a substance processed with an excipient only where such processing is mentioned in the definition section of the monograph."

rDNA product

**Title**

**INN**

**Formula**

molecular and graphic

**Relative mass CAS number**

**Definition**

Synthetic peptide rDNA product

Version date

6
### Synthetic peptide

**TERLIPRESSIN**

**Identification**
- no second identification
- often cross-references to Tests and Assay
- specific to product (e.g., glycan analysis for glycoproteins)

**For synthetic peptides:**
- LC + AAAs or LC + NMR (up to ~15aa; dedicated CRS)

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### rDNA product

**FILGRASTIM CONCENTRATED SOLUTION**

**Filgrastim solution concentrate**

**Production**
- absent for synthetic peptides;
- may be present for chemicals;
- extensive for vaccines;
- may contain specific tests for rDNA products

- source materials, manufacturing process, validation, control, in-process testing;
- mandatory for manufacturers;
- independent verification difficult

**Compliance:**
- see General Notices

**Characters**
- Appearance, hygroscopicity, crystallinity, solubility
- useful info for analyst
- not analytical requirement

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* See also Characters section in monographs (5.11)

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**Synthetic peptide rDNA product**

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**Synthetic peptide**

**TERLIPRESSIN**

**Tests**

- purity/impurity assessment
- limits based on specifications and batch data for approved products
- references to general chapters for synthetic peptides:
  - specific optical rotation, potential replacement by chiral chromatography
  - absorbance – whenever appropriate
  - related peptides/substances
  - acetic acid
  - water
  - bacterial endotoxins – no longer present in new monographs*

* Requirements of the general monograph Substances for pharmaceutical use (2034) apply; see European Pharmacopoeia policy on bacterial endotoxins in substances for pharmaceutical use (http://pharmeuropa.edqm.eu)

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**Related peptides/substances – main impurity test**

Identification of impurities and SST

- Impurity identification, SST criteria related to peak separation → Terlipressin impurity mixture CRS (Imp. A, D, L);
- Other SST criteria (S/N, symmetry factor, repeatability) → Terlipressin CRS

* separated as shown in the chromatogram supplied with Terlipressin impurity mixture CRS
Related peptides – main impurity test

**Impurity limits**

- each specified impurity (sometimes sum)
- unspecified impurities (identification threshold)
- total impurities
- reporting threshold

**Substances for Pharmaceutical Use (2034)**:

<table>
<thead>
<tr>
<th>Reporting threshold</th>
<th>Identification threshold</th>
<th>Qualification threshold</th>
</tr>
</thead>
<tbody>
<tr>
<td>&gt; 0.1 per cent</td>
<td>&gt; 0.5 per cent</td>
<td>&gt; 1.0 per cent</td>
</tr>
</tbody>
</table>

**Synthetic peptide**

**MONOGRAPH SECTION**

**rDNA product**

**Transparency section**

- present in synthetic peptide monographs
- controlled by related substances test (synthetic peptides)
- not necessary exhaustive – impurities known to and shown to be detected by the test - based on information obtained and verified during monograph elaboration/revision
**New impurity profiles:**
**Directive 2003/63/EC**

*pro memoria*

“However, where a starting material in the European Pharmacopoeia … has been prepared by a method liable to leave impurities not controlled in the pharmacopoeia monograph, these impurities and their maximum tolerance limits must be declared and a suitable test procedure must be described.”

“In cases where a specification contained in a monograph of the European Pharmacopoeia (...) might be insufficient to ensure the quality of the substance, the competent authorities may request more appropriate specifications from the marketing authorisation holder…”

**Monograph revision**

*pro memoria*

**Impurities control has to be updated for newly authorised products/sources:**

“[Where] a monograph … [may] be insufficient … the competent authorities shall inform the European Pharmacopoeia. The marketing authorisation holder shall provide the European Pharmacopoeia with the details of the alleged insufficiency and the additional specifications applied.”
Inorganic impurities

- Sulfated ash (2.4.14): global determination of foreign cations – present in some monographs for synthetic peptides: (e.g. leuprorelin)
- Tests for known and identified inorganic impurities (e.g. elemental impurities, reagents, ligand catalysts, inorganic salts, filter aids) – present when appropriate in other monographs

Residual solvents

- tests included only for class 1 solvents and class 3 when they exceed 0.5%
- absent in monographs for synthetic peptides
- covered by:
  - Substances for pharmaceutical use (2034)
  - Residual solvents (5.4)

Synthetic peptide

TERLIPRESSIN
Tetrapressinum

Content: 95.0 per cent to 105.0 per cent (anhydrous, acetic acid-free basis)

Assay: (…)

- synthetic peptides: comparative chromatographic procedures using defined CRS as a standard
- content: anhydrous, acetic acid-free basis
- often asymmetric limits

rDNA product

FILGRASTIM CONCENTRATED SOLUTION
Filgrastim solutio concentrata

Content: minimum 0.9 mg of protein per milliliter

Potency: minimum 0.9 x 10^8 IU per milligram of protein

Assay: Protein: Liquid chromatography (2.2.29) as described in the test for related substances with the following modifications. Calculate the percentage content of filgrastim (C32H39HNO29) taking into account the assigned content of C32H39HNO29 in reference CRS.

The potency of the preparation to be examined is determined by comparison of the dilutions of the test preparation with the dilutions of the International Standard of filgrastim or with a reference preparation calibrated in International Units. The International Unit is the activity contained in a stated amount of the appropriate International Standard. The equivalence in International Units of the International Standard is stated by the World Health Organization.
**Storage**

- not mandatory
- decided by competent authority (may decide to make it mandatory)
- storage → to ensure compliance with the monographs
- Conventional expressions (e.g. *in an airtight container*) defined in General Notices

**Labelling**

- covered by national and international regulations
- not comprehensive
- only statements necessary to demonstrate (non-) compliance are mandatory (e.g. nominal value for excipients)
- Label → container, package, leaflet, CoA

General Notices (1) apply to all monographs and other texts. See the information section on general monographs (cover pages)
Knowledge Database – additional source of information

- **Ongoing revision**
  - scope
  - state of work
  - last issue of Pharmeuropa where the draft was published

**Associated Reference Standards**

**Practical Info (e.g. column brand)**

**CEP holders**

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**Summary**

- Individual monographs and relevant general monographs/chapters are complementary
- Referenced general chapters become mandatory
- Monograph sections:
  - **Title**
  - **Formula & CAS**
  - **Mass**
  - **Definition**
  - **Production***
  - **Characters**
  - **Identification**
  - **Tests**
  - **Assay**
  - **Storage**
  - **Labelling**
  - **Impurities***

*Absent in monographs for synthetic peptides or products of rDNA technology

- Not mandatory monograph sections: Characters, Storage
- Consult Knowledge Database for additional monograph-associated information
Thank you for your attention!