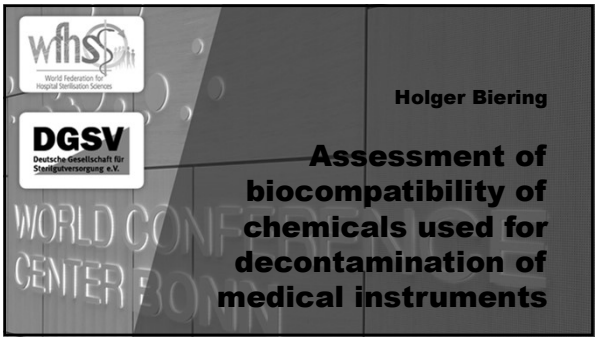


Holger Biering

Assessment of biocompatibility of chemicals used for decontamination of medical instruments

Agenda

- **Why biocompatibility assessments of process chemicals?**
- **Objectives for biocompatibility expert working groups.**
- **Test protocol to assess biocompatibility.**
- **Determination of acceptance level at user site.**
- **Summary.**

WHY BIOCOMPATIBILITY ASSESSMENTS OF PROCESS CHEMICALS?

- **European Medical Device Directive requires risk assessment of safety-related characteristics of medical devices before first use.**
- **Same safety level for processed medical devices like new one.**
- **Manufactures of washer-disinfectors must specify tolerable residues according to ISO 15883.**
- **ISO 15883 describes no methods how to do this.**
- **One element is the biocompatibility of process chemical residues.**

OBJECTIVES FOR BIOCOMPATIBILITY EXPERT WORKING GROUPS

- **Process chemical manufactures used different test protocols for biocompatibility assessments.**
- **Set up expert working groups by the industrial organization of German process chemical manufactures (IHO) with following goals:**
  - Development of a common test protocol to asses the biocompatibility of process chemical residues.
  - Formulation of uniform methodologies for determination of tolerable residual amounts.

TEST PROTOCOL TO ASSESS BIOCOMPATIBILITY

- **First decision in the working group:**
  Test protocol should be based on ISO 10993 "Biological evaluation of medical devices".
- **ISO 10993 Part 1 "Evaluation and testing within a risk management system" describes required tests depending on**
  – nature of body contact and
  – contact duration
  between medical device and human tissue and/or body fluid.

- **Following tests are proposed for surgical instruments, rigid and flexible endoscopes with limited contact time (<24h):**
  – Sensitization
  – Irritation
  – Systematic toxicity (acute)
  – Cytotoxicity
  – Haemocompatibility (in some cases)

- **Following tests are proposed for surgical instruments, rigid and flexible endoscopes with limited contact time (<24h):**
  – Sensitization → If applicable: Formula can be evaluated based on raw material data
  – Irritation → If applicable: Formula can be evaluated based on raw material data
  – Systematic toxicity (acute) → If applicable: Formula can be evaluated based on raw material data
  – Cytotoxicity → Tests are required
  – Haemocompatibility (in some cases) → Tests are required

- **Following tests are proposed for surgical instruments, rigid and flexible endoscopes with limited contact time (<24h):**
  – Sensitization → If applicable: Formula can be evaluated based on raw material data
  – Irritation → If applicable: Formula can be evaluated based on raw material data
  – Systematic toxicity (acute) → If applicable: Formula can be evaluated based on raw material data
  – **Cytotoxicity** → **Tests are required**
  – Haemocompatibility (in some cases) → **Tests are required**

Cytotoxicity tests

Products tested:

- **Product A: liquid disinfectant containing 10-25% glutaraldehyde, 10-25% ethanol and water.**
- **Product B: liquid two component disinfectant, Component 1 containing 1-5% peracetic acid, 8-35% hydrogen peroxide, <10% acetic acid and water, Component 2 containing 2-5% sodium hydroxide and water.**
- **Product C: liquid disinfectant and detergent containing <10% quaternary ammonium compound (QAC), <10% diamine, non-ionic surfactants, solvents, complexing agents and water.**
- **Product D: liquid detergent containing 5-15% fatty alcohol alkoxylate, solvent and water.**

Cytotoxicity tests

First test level: Detection of concentration limits in solutions:

- **Solutions with different concentration (1.0, 0.1, 0.01, 0.001, and 0.0001 Vol-%) are prepared.**
- **These solutions are mixed with the cell culture medium (Dulbecco's Modified Eagle Medium-DMEM).**
- **Aliquots of 100 µl are pipetted into the cell culture plate.**
- **Incubation for 72 ± 6 h at 37 ± 1 °C.**
- **Measurement of protein content.**
- **Calculation of proliferation inhibition.**

Cytotoxicity tests

First test level: Detection of concentration limits in solutions:

Concentration		Formulation			
Vol-%	ppm	Product A	Product B	Product C	Product D
1	10,000	100	100	100	100
0.1	1,000	100	100	100	100
0.01	100	100	12	100	100
0.001	10	8	0	65	78
0.0001	1	0	2	22	23

Values greater than 30 % proliferation inhabitation – in red – are classified as being cytotoxic

Product A: Glutaraldehyde based disinfectant
Product B: Buffered Peracetic Acid based disinfectant
Product C: QAC + Diamine based disinfectant and detergent
Product D: Neutral cleaner

Cytotoxicity tests

Second test level: Cytotoxicity to Process Challenge Devices (PCD's):

- **Following PCD materials are used:**
  - Stainless steel X20Cr13, brushed surface, representative of non-cutting surgical instruments.
  - Silicon rubber, representative of anaesthesia equipment.
- **Solutions with different concentration (1.0, 0.1, 0.01 and 0.001 Vol-%) are prepared.**
- **PCD's are immersed for 1 h in the test solution, then dries for 15 sec on paper and 1 h at room temperature.**
- **PCD's are eluted 1.5 Vol-% DMSO in cell culture medium (DMDM).**
- **Aliquots of 100 µl were pipetted into the cell culture plate.**
- **Incubation for 72 ± 6 hat 37 ± 1 °C, measurement of protein content and calculation of proliferation inhibition.**

Cytotoxicity tests

Second test level: Cytotoxicity to PCD's made of stainless steel:

Concentration		Formulation			
Vol-%	ppm	Product A	Product B	Product C	Product D
1	10,000	0	15	100	71
0.1	1,000	5	20	79	33
0.01	100	3	13	33	17
0.001	10	3	19	33	17

Values greater than 30 % proliferation inhabitation – in red – are classified as being cytotoxic

Product A: Glutaraldehyde based disinfectant
Product B: Buffered Peracetic Acid based disinfectant
Product C: QAC + Diamine based disinfectant and detergent
Product D: Neutral cleaner

Cytotoxicity tests

Second test level: Cytotoxicity to PCD's made of silicon rubber:

Concentration		Formulation			
Vol-%	ppm	Product A	Product B	Product C	Product D
1	10,000	25	5	100	59
0.1	1,000	11	0	100	33
0.01	100	2	0	47	8
0.001	10	20	12	25	5

Values greater than 30 % proliferation inhabitation – in red – are classified as being cytotoxic

Product A: Glutaralaldehyde based disinfectant
Product B: Buffered Peracetic Acid based disinfectant
Product C: QAC + Diamine based disinfectant and detergent
Product D: Neutral cleaner

Summary Cytotoxicity tests

- **Disinfectants are cytotoxic in diluted solutions in declining intensity:**

  QAC/Amine -> Glutaraldehyde -> buffered Peracetic Acid.
- **Adsorption effects on surfaces seems to be dominant related to cytotoxicity potential of products on stainless steel and silicone rubber:**
  - Glutaraldehyde and buffered Peracetic acid have low adsorption potential on both materials
    => no cytotoxicity up to 1 Vol%
  - QAC has high adsorption potential on both materials
    => cytotoxic effects up to 0,01 Vol%.
- **Cytotoxic behaviours of non-ionic surfactants seems to be dominant related to neutral cleaner:**

  => cytotoxic effects up to 0,001 Vol% in solution and 0,1 Vol% on both PCD materials.

WG-Proposal for assessment of biocompatibility

- **Experimental detection of cytotoxic properties of process chemicals**
  - in diluted solution (first test level) and if necessary
  - of product residues on various surfaces relevant for the intended application (second test level).
- **Assessment of systemic toxicity, irritation and sensitization potential**
  based on already available data for the respective raw materials.
- **Experimental detection of haemocompatibility of process chemicals depending on the intended use of reprocessed medical devices.**
- **Evaluation of all data within the framework of biocompatibility assessment**
  => definition of acceptance value in µg/cm² or µg/instrument.

DETERMINATION OF ACCEPTANCE LEVEL AT USER SITE

Surgical instruments

Automated processing in washer-disinfectors

Measurement of conductivity at the end of the process in final rinse water:

- **Indirect method**
- **Applicable, if acceptance level in solutions is high enough.**
- **Mainly used for validation of thermal disinfection processes in combination with alkaline cleaners and neutralizer.**
- **Not applicable for most of neutral cleaner, antimicrobial cleaner and disinfectants.**

Lit.: Biering H, Glasmacher R, Hermann M, Schrader E: Biocompatibilty of medical devices after automated reprocessing in washer-disinfectors. Central Service 2011; 19(5): 334-339.

Surgical instruments

Manual processing

Residue extraction from medical device surface:

- **Direct method proposed by IHO working group**
- **Crile clamps are used as PCD's.**
- **Residue extraction after processing with demineralized water.**
- **Analytical detection of key components of used process chemicals.**
- **Applicable for all types of process chemicals.**

Lit.: Tschoerner M: Methods for determination of tolerable process chemical residues after manual processing. Central Service 2017; in print.

Thermolabile Endoscopes

Automated processing in washer-disinfectors

- **Disinfectant residue extraction from endoscope surface:**
  - Direct method
  - Residues are extracted from distal end.
  - Analytical detection of glutaraldehyde.

Lit.: 1. Emmrich M, Bloß R, Martiny H: Glutaraldehyde(GA) Residues in Flexible Endoscopes. Part I: Development of an Analytical Method for Detection of GA Residues. Central Service 2014; 22(1): 46-49.
2. Emmrich M, Bloß R, Martiny H: Glutaraldehyde(GA) Residues in Flexible Endoscopes. Part II: Analytical Method and Factors for Detection of GA Residues. Central Service 2014; 22(1): 84-87.

- **Disinfectant residue determination in final rinse water:**
  - Indirect method
  - Applicable, if acceptance in solutions is high enough.
  - Analytical detection of peracetic acid.

Thermolabile Endoscopes

Automated processing in washer-disinfectors

Residue extraction from surface of PCD's:

- **Method proposed by IHO working group**
- **Polyurethane blocks are used as PCD's.**
- **Residue extraction after processing with demineralized water.**
- **Analytical detection of key components of used process chemicals.**
- **Applicable for all types of process chemicals.**

Lit.: Biering H: Determination of tolerable process chemical residues after reprocessing thermolabile endoscopes. Central Service 2016; 24(3): 160-164.

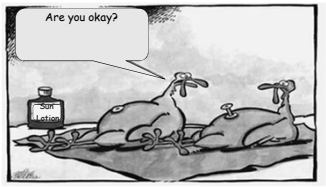
SUMMARY

Steps for biocompatibility assessment and validation/verification at user site:

- **Determination of tolerable residual amount of the respective products.**
- **Definition of conductivity values in the final rinse water for alkaline cleaners and neutralizer.**
- **Investigation of adsorption and extraction profiles of process chemicals with respect to medical devices.**
- **Development and provision of analytical methods for determination of tolerable residual amount.**

Members of three working groups (in alphabetic order):

Dr. Holger Biering	**Dr. Richard Bloß**
Dr. Erik Brückner	**Dr. Kai Groh**
Dr. Thomas-Jörg Henning	**Dr. Elmar Hjorth**
Markus Kamer	**Dagmar Martini**
Alexander Müller	**Dr. Andreas Otte**
Axel Schneider	**Michael Schreiner**
Anna-Maria Sprünken	**Dr. Matthias Tschoerner**



Thank you for your attention!