Potential of Cold Atmospheric Plasma as a new method for the decontamination and sterilization of reusable surgical instruments.

14th World Sterilization Congress
8th National Sterilization Disinfection Congress of Turkey

6th-9th November 2013

Rodolphe Hervé PhD, MSc
The five main functions performed in a hospital sterile service department:

- Cleaning
- Disinfection
- Inspection
- Packaging
- Sterilisation
Decontamination and sterilization
Assessment of instruments cleanliness
Thioflavin T (bright blue) and SYPRO Ruby (amber) dual staining observed on (a-e) a suction canullae and (f) diathermy forceps from a neurosurgery set that were fully reprocessed through a sterile service department. White bars are 100 μm, red bar is 10 μm.

Hervé et al., JHI 2010
Standard reprocessing: neurosurgery instruments

Proteinaceous (total and amyloid) contamination on a suction cannulae isolated from a craniotomy set.
Standard reprocessing: flexible luminal endoscopes

Hervé et al., JHI 2012
Standard reprocessing: endodontic files
Standard reprocessing: endodontic files
Cleaning limitations
Protein removal action of various cleaners

Hervé et al., JHI 2010
Cleaning limitations

Remaining contamination on surfaces
(proportion of hydrophobic amyloid-rich proteins)

Hervé et al., JHI 2010
### Mechanism

<table>
<thead>
<tr>
<th>Physical disruption</th>
<th>Enzymatic degradation</th>
<th>Chemical modification (pH&gt;12)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(detergent, sonication, brushing, flushing)</td>
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</tbody>
</table>

### Potential caveats

| Displacement and/or spreading | Shelf life; control of parameters | Damage to instruments; control of parameters and efficacy |
What is gas plasma?

**Benefits**

- Energetic electrons → chemical dissociation @ low gas temperature
- On-site production of reactive, short-living species e.g. O$_2$•−; O; ¹O$_2$; NO ... OH• and H$_2$O$_2$ → known to act on protein, lipid and DNA
- **Oxidants**: OH•, O$_2$•−; O; ¹O$_2$, H$_2$O$_2$

what is gas plasma? | Shooting Plasma

Nanosecond imaging

Exposure: 1ns
Interval: 100 ns
Testing of first CAP prototype
CAP parameters during initial tests

High purity helium/oxygen mix

5 L/min and 100 ml/min respectively

Target surface set within 1.5 cm

Up to 2 min application
Testing of first CAP prototype

<table>
<thead>
<tr>
<th>Treatment duration</th>
<th>Remaining contamination (μg/mm²)</th>
</tr>
</thead>
<tbody>
<tr>
<td>reference</td>
<td>0.025</td>
</tr>
<tr>
<td>30 sec</td>
<td>0.005</td>
</tr>
<tr>
<td>60 sec</td>
<td>0.005</td>
</tr>
<tr>
<td>90 sec</td>
<td>0.005</td>
</tr>
<tr>
<td>2 min</td>
<td>0.005</td>
</tr>
<tr>
<td>2+1 min</td>
<td>0.005</td>
</tr>
<tr>
<td>2+2 min</td>
<td>0.005</td>
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</tbody>
</table>

Graph showing the remaining contamination for different treatment durations.
Spiked implant wires before CAP treatment
Spiked implant wires after partial CAP treatment
CAP appears equally effective against amyloid proteins

![Graph showing the effectiveness of CAP against amyloid proteins. The x-axis represents the control and CAP-treated samples, while the y-axis shows the residual contamination in pixels per field of view. The graph also displays the amyloid/protein ratio and the amyloid proportion (%).]
The place of CAP in standard reprocessing
The place of CAP in standard reprocessing
The place of CAP in standard reprocessing
what is gas plasma?

**Optimisation**

**Ar-O2-H2O Plasmas**

- up to 1,000 chemical reactions
- threshold dose of each protein-inactivating plasma agent is unknown
- complication with synergy
- plasma diagnostics is complex and not always accessible
- plasma diagnostics in liquid phase – very under-studied field
- if achieved, a **knowledge-based inaction** – indirect **inactivation marker**
- ... empirical strategy – **unreliable** for ensuring efficacy and for system scaling

<table>
<thead>
<tr>
<th>Agents</th>
<th>Plasma Characterisation</th>
</tr>
</thead>
<tbody>
<tr>
<td>O*; OH* etc</td>
<td>Excited species – Optical emission spectroscopy (OES)</td>
</tr>
<tr>
<td>O, H2O-clusters</td>
<td>Ground state – OES not good – MBMS</td>
</tr>
<tr>
<td>OH, H2O2 etc in H2O</td>
<td>Electron spin resonance (ESR) spectroscopy</td>
</tr>
<tr>
<td>UV</td>
<td>Absolute OES</td>
</tr>
<tr>
<td>Ozone</td>
<td>Spatial resolved – UV absorption spectroscopy (UVAS)</td>
</tr>
<tr>
<td>Electron density</td>
<td>Probes not applicable – current density</td>
</tr>
<tr>
<td>Electron energy</td>
<td>Boltzmann plot</td>
</tr>
<tr>
<td>All</td>
<td>Plasma modelling</td>
</tr>
</tbody>
</table>

... additional studies, such as OH scavengers
CAP optimisation

EDIC/EF for rapid and very sensitive quantification of residual proteins and microorganisms.

Refined prion infectivity assays for human strains under development.

Mechanisation and scaling up of the process.

Adaptation into SSDs.
Conclusions

Proteinaceous microcontamination (potentially including PrP\textsuperscript{Sc} in affected countries) is a common problem in clinical settings.

Current standard decontamination procedures suffer from inherent physicochemical limitations.

CAP offers a radically different decontamination mechanism capable of targeting individual atoms, without the problems associated with liquid solutions.

Further development required to adapt the technology to different end users/instruments.
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