



## Research article

# Microbiological evaluation of ultraviolet C light-emitting diodes for disinfection of medical instruments

Hannah Siwe<sup>a,b</sup>, Annelies Aerssens<sup>c</sup>, Mieke V. Flour<sup>b</sup>, Silke Ternest<sup>a,c</sup>,  
Leen Van Simaey<sup>d</sup>, Duncan Verstraeten<sup>b</sup>, Alain F. Kalmar<sup>e</sup>, Isabel Leroux-  
Roels<sup>c,f,1,\*</sup>, Philip Meuleman<sup>a,1,\*\*</sup>, Piet Cools<sup>d,1,\*\*\*</sup>

<sup>a</sup> Laboratory of Liver Infectious Diseases, Department of Diagnostic Sciences, Faculty of Medicine and Health Sciences, Ghent University, Corneel Heymanslaan 10, entrance 38, Medical Research Building 2, 9000, Ghent, Belgium

<sup>b</sup> Research and Development, eLEDricity, Hundelgemsesteenweg 446A, 9820, Merelbeke, Belgium

<sup>c</sup> Department of Infection Control, Ghent University Hospital, Corneel Heymanslaan 10, entrance 12, Clinical Building 12E, 9000, Ghent, Belgium

<sup>d</sup> Laboratory Bacteriology Research, Department of Diagnostic Sciences, Faculty of Medicine and Health Sciences, Ghent University, Corneel Heymanslaan 10, entrance 38, Medical Research Building 2, 9000, Ghent, Belgium

<sup>e</sup> Department of Electronics and Information Systems, IBI<sup>2</sup>Tech, Ghent University, Technologiepark-Zwijnaarde 126, 9052, Ghent, Belgium

<sup>f</sup> Department of Diagnostic Sciences, Faculty of Medicine and Health Sciences, Ghent University, Corneel Heymanslaan 10, entrance 38, Medical Research Building 2, 9000, Ghent, Belgium

## ARTICLE INFO

## Keywords:

UVC LED  
Sustainability  
*Staphylococcus aureus*  
Reprocessing

## ABSTRACT

**Background:** Despite the many guidelines for reprocessing of medical instruments, challenges persist such as microbial resistance to biocides, corrosive effects on materials, and time-consuming reprocessing procedures. Ultraviolet (UV) C light-emitting diode (LED) chambers might provide a solution but the integration in healthcare is still in its infancy. Here, we evaluated the efficacy of a novel ZAPARAY™ UVC LED chamber as a time and energy-efficient alternative for reprocessing of medical instruments for which current disinfection protocols exhibit limitations.

**Methods:** We verified the disinfection efficacy of the UVC LED chamber on a Petri dish and contaminated several medical devices with *Staphylococcus aureus* ATCC 25923. The bacterial reduction was assessed after 5 min of UVC LED exposure. Additionally, we investigated the impact of rinsing before UVC exposure.

**Results:** We demonstrated a bacterial reduction of 9 log<sub>10</sub> on a Petri dish. Non-rinsed dental tools exhibited varied reduction levels ranging from a 3.23 log<sub>10</sub> to a 6.25 log<sub>10</sub> reduction. Rinsing alone yielded an average reduction of 2.7 log<sub>10</sub> and additional UVC exposure further reduced the bacterial load by an average of 3.65 log<sub>10</sub>. We showed an average 4.90 log<sub>10</sub> reduction on thermistors, 2 log<sub>10</sub> or less on orthodontic pliers, and no reduction on handpieces.

**Conclusions:** This study demonstrates that UVC LED chambers may be used as a standardized substitute for specific (manual) disinfection procedures of certain medical devices, offering a

\* Corresponding author.

\*\* Corresponding author.

\*\*\* Corresponding author.

E-mail addresses: [Isabel.LerouxRoels@UZGent.be](mailto:Isabel.LerouxRoels@UZGent.be) (I. Leroux-Roels), [Philip.Meuleman@UGent.be](mailto:Philip.Meuleman@UGent.be) (P. Meuleman), [Piet.Cools@UGent.be](mailto:Piet.Cools@UGent.be) (P. Cools).

<sup>1</sup> Shared senior authors with equal contribution.

time-efficient and more sustainable alternative. However, its use should be preceded by efficacy testing for each specific type of instrument.

## 1. Introduction

Correct reprocessing of reusable medical devices is an essential cornerstone of a good infection prevention and control program in healthcare facilities [1]. Reprocessing entails cleaning, disinfection and/or sterilization procedures to ensure the safe reuse of medical instruments [2]. This can be obtained through (the combination of) a variety of physical and chemical methods that include the use of detergents, oxidizing agents, aldehydes, radiation, and/or heat [3,4]. Spaulding's classification is a commonly used system to assess which procedure is required for adequate reprocessing [4]. This classification defines non-critical (in contact with intact skin), semi-critical (in contact with non-intact skin or mucosae), or critical instruments (in contact with blood or sterile tissues), with the latter posing the highest risk for infection [3].

Despite the many reprocessing guidelines and protocols, device-associated outbreaks have been increasingly reported [5–7] and several challenges and limitations remain. Critical devices (e.g. surgical instruments) and some semi-critical devices, such as heat-sensitive flexible endoscopes, undergo a largely automated process of sterilization or high-level disinfection (HLD) [3] but these procedures are often time-consuming. Many non- and semi-critical medical instruments still undergo (semi-) manual reprocessing, but these manual procedures are not standardized and therefore highly error-prone, particularly when it concerns devices with a complex design [8–11]. The use of chemical disinfectants is associated with a high environmental burden [12,13], and the risk of inducing or increasing microbial resistance to biocides and cross-resistance to antibiotics [14,15]. The recommended dwell time of disinfectants may also be compromised under time pressure [16]. Furthermore, corrosive effects on instruments have been reported as a result of heat sterilization or chemical disinfection [17,18].

Disinfection by ultraviolet (UV) C generated by mercury discharge lamps is well established in the context of environmental disinfection [19–21] and might provide an answer to some of these challenges. UVC inactivates microorganisms either by direct absorption or indirectly via reactive oxygen species, targeting their DNA, RNA, proteins, and lipids [22–24]. However, there are more energy-efficient alternatives available including UVC light-emitting diodes (LEDs). These LEDs offer various other advantages such as being more compact and eco-friendly as they are free of toxic mercury [25,26]. Nevertheless, UVC LEDs are at present not widely used which could be as a result of their lower irradiance efficiency as compared to mercury discharge lamps [26]. However, UVC LEDs remain highly effective in deactivating pathogens and the technology is continuously evolving, as forecasted by Haitz's law [27]. Ongoing research aims to further enhance their efficiency [28], indicating a promising future for UVC LEDs in various applications.

The implementation of UVC chambers for instrument disinfection in healthcare is still in an early stage due to several limitations that need to be considered such as the low quality of available research, the lack of standardization for UVC chambers, the low penetration depth of UVC, and the necessity for optimal exposure of the instrument without shadow formation [29]. UVC mercury chambers have been researched and shown to be effective for disinfection of electronic devices such as tablets [30,31] used in hospitals, and to some extent on medical devices including intravaginal probes [32–34], flexible fiberoptic laryngoscopes [35] and silicon dental impressions [36]. UVC LED has been investigated for stethoscope disinfection, revealing its potential for effective decontamination of these medical devices [37].

To the best of our knowledge, the efficacy and broad applicability of UVC LED chambers for instrument disinfection have not been thoroughly investigated, highlighting the need for further research to ensure their correct and optimal implementation. We aimed to evaluate the efficacy of a novel UVC LED chamber for disinfection of non- or semi-critical instruments, particularly those instruments for which current disinfection protocols exhibit limitations. In this study, we focused on how the shape and technical characteristics of the medical instruments influence the disinfection efficacy, using *Staphylococcus aureus* as a clinically relevant model pathogen.

## 2. Methods

### 2.1. Identification and selection of instruments

As an initial market survey, open interviews were held with potential end-users in health care (e.g. head nurses, dentists ...), which led to the selection of dental tools (hammer shaped needle, sawtooth needle, and chisel needle) as proof of concept for microbiological testing (Table 1). At 14 departments of the Ghent University Hospital, semi-structured interviews, and questionnaires (Supplementary Figure A1) were used to identify those instruments for which there is a high need for an alternative disinfection strategy. In total, 29 identified instruments were given a high, medium, or low score for six parameters (i.e. correct implementation of current protocol, impact of current protocol on lifespan of instrument, processing time of current protocol, use frequency, cost of instrument and risk of a healthcare-associated infection (HAI)) (Supplementary Tables A1 and A2) and plotted in two heatmaps displaying instrument clusters sharing similar characteristics (Supplementary Figure A2). Based on feasibility and accessibility, a subset of instruments were selected for microbiological testing, more specifically, a nasal/oral thermistor (sensor) (REF: P1 222, Pro-Tech, Downers Grove, Illinois, USA), a dental handpiece (REF: SYNEA WA-99 LT, W&H Dentalwerk, Bürmoos, Austria), a round ended orthodontic plier (REF: 4151-640, Masel Orthodontics, Carlsbad, California, USA), and a sharp ended orthodontic plier (REF: 678-106, Hu-Friedy, Chicago, Illinois, USA) (Table 1).

## 2.2. UVC LED device

The UVC LED disinfection device evaluated in this study was the ZAPARAY™ RAY-ONE prototype 0/102 (eLEDricity, Merelbeke, Belgium) and further designated in this manuscript as “UVC LED chamber”. The device consists of an enclosed chamber with a drawer made of a quartz bottom in which objects can be placed with a maximum size of 200 mm × 300 mm × 70 mm (width × depth × height). The chamber and drawer are lined with UVC reflective material. This reflective material, along with LEDs mounted at the top and bottom of the chamber that emit UVC radiation at 275 nm, ensure homogenous UVC exposure with an irradiance of approximately 0,395 mW/cm<sup>2</sup>.

## 2.3. Contamination

*Staphylococcus aureus* (ATCC 25923) was aerobically cultured at 37 °C for 24 h on tryptic soy agar with 5 % sheep blood (TSA-5% SB) (BD, Franklin Lakes, New Jersey, USA). For each experiment, an inoculum suspension was freshly prepared by resuspending *S. aureus* colonies in 0.9 % sterile NaCl solution (saline). The optical density (OD) at 600 nm was measured using the Denovix DS-11 spectrophotometer (Denovix, Wilmington, USA) with the OD ranging between 1,0 and 1,7 to obtain a concentration of approximately 10<sup>9</sup> colony forming unit (CFU)/mL. As initial verification and reference, a 10 µL droplet of the inoculum suspension was pipetted onto the center of a sterile 60 × 15 mm polystyrene Petri dish and then immediately exposed to UVC. Dental tools, orthodontic pliers and handpieces were steam sterilized before the start of each experiment and thermistors were decontaminated with 70 % ethanol. Dental tools were contaminated by applying the inoculum through partial submersion in 7 mL of inoculum suspension in a 15 mL tube. Orthodontic pliers were partially submerged in 80 mL inoculum suspension in a 100 mL multipurpose beaker. Handpieces were contaminated through partial submersion of the instrument in 10 mL of inoculum suspension in a 50 mL tube. All instruments were incubated for 10 min at room temperature. Afterward, a tapping motion was applied to the instruments to remove the excess liquid by tapping the instrument four times against the border of its container, used during contamination. Thermistors were contaminated around the three sensor tips by rubbing a cotton swab that had been soaked with inoculum in a circular motion around the tips. To investigate whether rinsing before UVC exposure would improve the outcome of the disinfection procedure; dental tools, handpieces, and orthodontic pliers, were rinsed for 3 s under a jet of water as a simplified cleaning step. Pliers were held in an open orientation during rinsing for water to flow through the hinge. Afterward, instruments were dapped on tissue paper to remove excess liquid. All instruments were exposed to UVC immediately after the contamination procedure without allowing the instruments to dry up.

## 2.4. UVC disinfection

Petri dishes and medical devices were positioned directly in the center of the drawer, except for dental tools which were positioned in a fixture for stability. Orthodontic pliers were placed in an open orientation. Petri dishes and medical devices were subjected to a 5-min disinfection cycle which equated to a UVC dose of approximately 118 mJ/cm<sup>2</sup>. Contaminated Petri dishes and instruments that did not undergo UVC treatment were handled in parallel and served as an untreated control.

## 2.5. Assessment of log reduction factors

### 2.5.1. Bacterial recovery







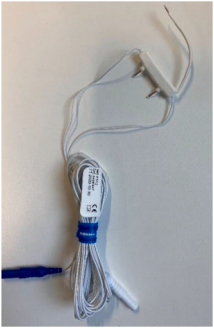
The 10 µL droplet in the Petri dish was collected by locally rinsing the area of the droplet with 1 mL of saline which was then added to 9 mL saline and resuspended. Afterward, it was locally rinsed for a second time with 1 mL originating from the already collected volume to apply additional friction for the collection of residual material on the surface of the Petri dish. Bacteria on dental tools were recovered by partial submersion in 7 mL saline in a 15 mL tube. Handpieces were placed in a 50 mL tube containing 10 mL saline. All tubes with objects inside were vortexed for 20 s. Orthodontic pliers were placed in an open orientation inside a 100 mL multipurpose beaker and rinsed five times with the same 10 mL saline. Thermistors were positioned in a fixture above a 60 × 15 mm polystyrene sterile Petri dish. Subsequently, the three sensor tips were each rinsed with 1 mL saline. Finally, all rinse fluids were collected in a 15 mL tube and vortexed.

### 2.5.2. Bacterial enumeration

The bacterial load was quantified by performing a tenfold serial dilution of the collected suspension. A droplet of 5 µL was pipetted onto TSA-5%SB for each dilution, and afterward, plates were slanted vertically at a nearly 90° angle to create a droplet line. Subsequently, plates were incubated at 37 °C for 18–24 h and colonies were manually counted. The dilution with the highest countable value within the range of 0–100 CFU was used to calculate the bacterial concentration of the original suspension. The log reduction was calculated as followed:

$$\log_{10} \text{reduction factor} = \log_{10} (\text{CFU/mL untreated}) - \log_{10} (\text{CFU/mL treated})$$

**Table 1**  
Medical devices.

dental tools			handpiece	orthodontic pliers		oral/nasal thermistor
hammer-shaped	sawtooth	chisel		round-ended	sharp-ended	
						

## 2.6. Confirmation of the absence of bacterial growth after UVC exposure

To confirm the lack of viable bacteria in the absence of growth with the droplet line technique, a 10  $\mu$ L droplet was spotted on a Petri dish and treated as previously described. The droplet was then collected by locally rinsing the area with 90  $\mu$ L saline. This total volume of 100  $\mu$ L was plated onto TSA-5%SB according to the spread plate technique. Similarly, thermistors and dental tools were prepared and handled as previously described and after bacterial recovery, the instruments were directly imprinted onto TSA-5%SB.

## 2.7. Statistical analysis

Statistical analysis was performed using GraphPad Prism version 8.0.1. Bar charts in all figures were depicted as mean  $\pm$  standard deviation with dots representing individual data points. Data was verified for normal distribution using the Shapiro Wilk test. A paired *t*-test was used when the data was normally distributed, otherwise, the Wilcoxon matched-pairs signed rank test was used. Statistical significance was expressed as \* ( $p < 0.05$ ), \*\* ( $p < 0.01$ ), \*\*\* ( $p < 0.001$ ), \*\*\*\* ( $p < 0.0001$ ), or ns (non-significant).

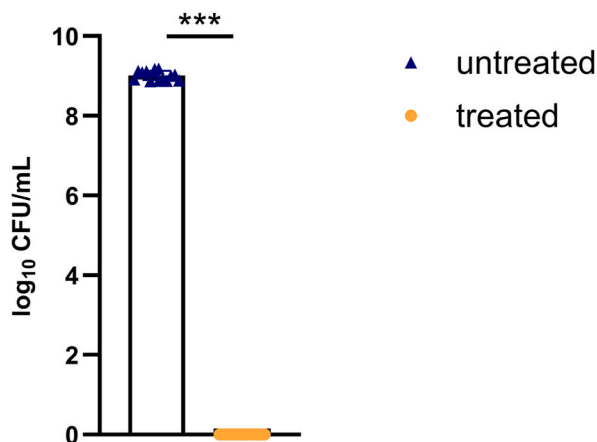
## 3. Results

### 3.1. Verification of the UVC LED chamber as a disinfection device

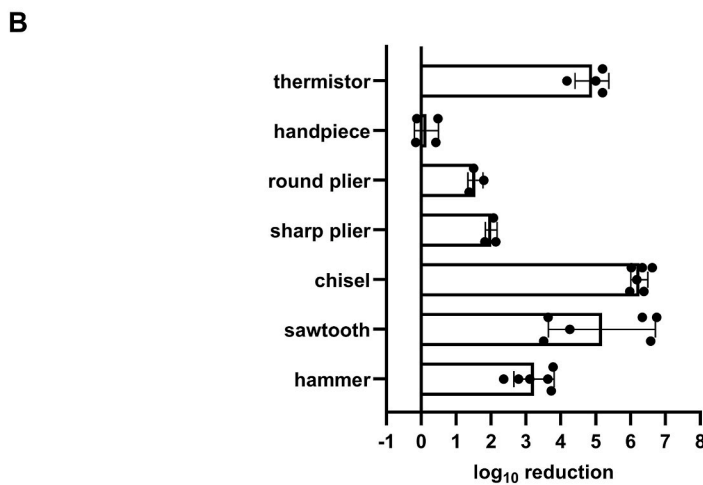
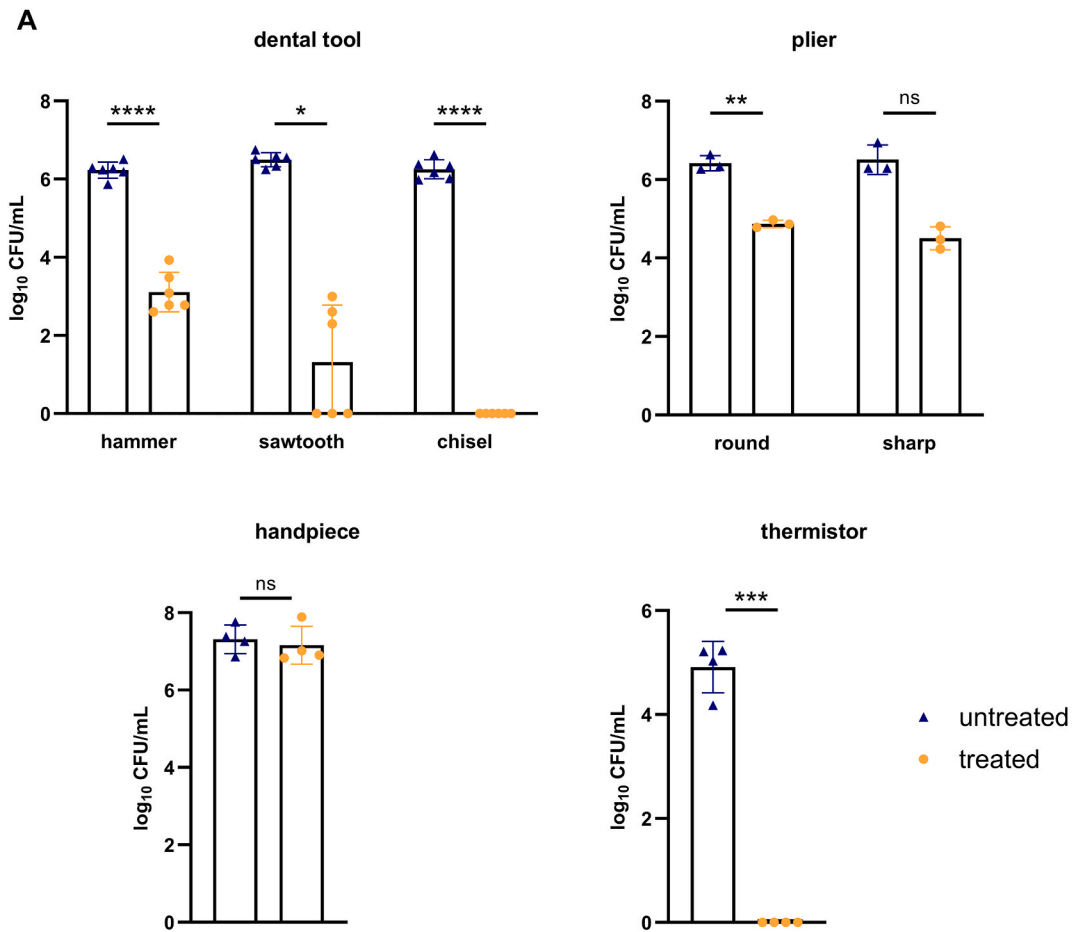
Before we initiated the evaluation of the device to disinfect medical instruments, we wanted to assess its usefulness in a disinfection experiment using Petri dishes as a simple surface. Collected suspensions from the untreated control ( $n = 14$ ) had an average bacterial concentration of  $1.02 \times 10^9$  CFU/mL (Fig. 1). Exposure of the droplet to a 5-min disinfection cycle resulted in a complete reduction below detectable levels across all experiments (Fig. 1), with an average  $\log_{10}$  reduction factor of 9. The absence of growth was confirmed when the treated droplet was spread-plated (Supplementary Figure A3).

### 3.2. Evaluation of the efficacy of the UVC LED chamber for disinfection of non-rinsed medical instruments

To evaluate the efficacy of the device for disinfection of medical instruments, various identified instruments were artificially contaminated with *S. aureus* and exposed to UVC. The bacterial load of the collected suspensions of the different dental tools and pliers was highly comparable for the untreated conditions, resulting in an average concentration of  $10^6$  CFU/mL (Fig. 2A). UVC exposure yielded significantly lower bacterial concentrations for all three dental tools, with a complete reduction for the chisel needle (Fig. 2A). Remarkably, for the sawtooth needle results varied from partial to complete reduction across different experiments. The average  $\log_{10}$  reduction factor was 3.23, 5.18 and 6.25 for the hammer-shaped needle, sawtooth needle, and chisel needle, respectively (Fig. 2B). Pliers showed a significant, but lower reduction of the bacterial load after UVC exposure with a  $\log_{10}$  reduction factor of 2 or less (Fig. 2B). UVC exposure of handpieces did not result in any bacterial reduction whereas thermistors showed the absence of growth after exposure, resulting in an average  $\log_{10}$  reduction factor of 4.90 (Fig. 2A and B). Additionally, the agar imprinting method confirmed the absence of bacterial growth on the UVC treated thermistors (Supplementary Fig. A4). However, a few colonies were observed for the treated chisel needle as compared to the dense growth of *S. aureus* for the untreated control (Supplementary Fig. A4).



**Fig. 1.** Bacterial load of a contaminated Petri dish without UVC exposure (untreated) and after 5 min of UVC exposure (treated). Log-transformed colony forming units (CFU)/mL of *S. aureus* under control and treatment conditions. Dots represent individual data points. Bar charts are shown as mean  $\pm$  standard deviation ( $n = 14$  treated and 14 untreated) (14 independent experiments), \*\*\* ( $p < 0.001$ ) (Wilcoxon matched-pairs signed rank test).



**Fig. 2.** Bacterial load and log reduction factors of non-rinsed medical instruments. Dots represent individual data points. Bar charts are shown as mean ± standard deviation (SD). (A) Log-transformed colony forming units (CFU)/mL of *S. aureus* under control and treatment conditions collected from dental tools (n = 6) (6 independent experiments), round and sharp ended orthodontic pliers (n = 3) (3 independent experiments), handpieces (n = 4) (2 independent experiments), and thermistor (n = 4) (2 independent experiments). \* (p < 0.05), \*\*p < 0.01, \*\*\* (p < 0.001), \*\*\*\* (p < 0.0001), ns (non-significant) (paired t-test except for data on sharp ended plier and sawtooth, Wilcoxon matched-pairs signed rank test) (B) Log<sub>10</sub> reduction factors for the different medical instruments.

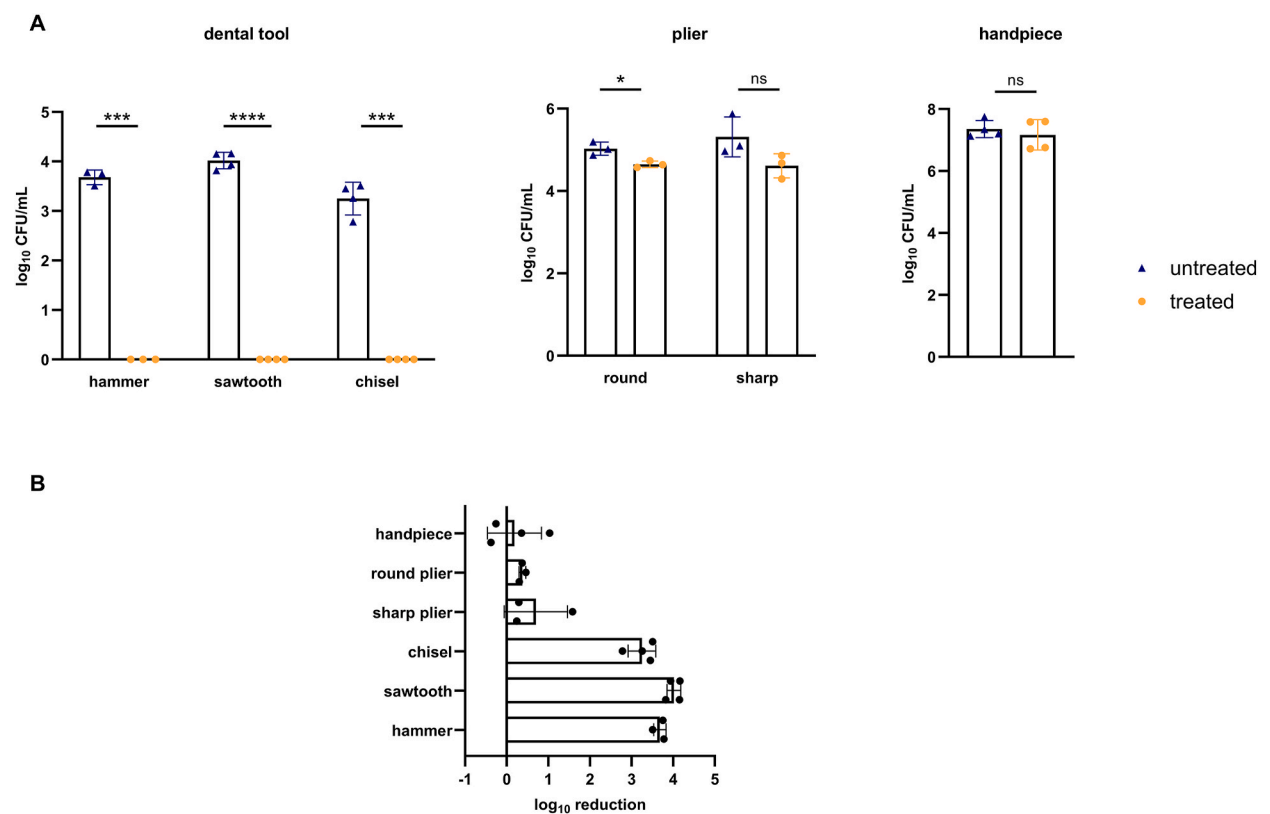
### 3.3. Evaluation of the efficacy of the UVC LED chamber for disinfection of rinsed medical instruments

To evaluate whether rinsing improves the overall disinfection procedure for those instruments where complete reduction was not obtained, a short rinsing step was included before UVC treatment. Rinsed untreated controls showed on average 2.7 and 1.3 log<sub>10</sub> lower bacterial loads (CFU/mL) for dental tools and pliers, respectively, compared to unrinsed counterparts (Fig. 3A and 2A). Rinsing followed by UVC exposure resulted in undetectable growth for the hammer shaped needle, sawtooth needle, and chisel needle with an average log<sub>10</sub> reduction factor of 3.68, 4.02, and 3.25 respectively, resulting in an overall average log<sub>10</sub> reduction factor of 3.65 (Fig. 3B). However, no rinsing effect was observed for pliers and handpieces for the treatment conditions (Fig. 3A and 2A). This resulted in an average log<sub>10</sub> reduction factor of less than 1 for pliers and no reduction for handpieces (Fig. 3B). Instrument imprinted plates of the rinsed dental tools showed a few colonies for all treated objects as compared to untreated objects (Supplementary Figure A5).

## 4. Discussion

In this study, we evaluated the performance of the novel ZAPARAY™ UVC LED chamber for the disinfection of artificially contaminated simple surfaces (i.e. Petri dishes) as well as of complex medical instruments. First, we have shown that the device was able to induce complete reduction (9 log<sub>10</sub> reduction) of *S. aureus* on a standard Petri dish that has a smooth, flat, non-porous surface without any cavities.

The results obtained on medical devices varied considerably depending on the instrument. In literature, research has been described where instruments were artificially contaminated and disinfected with UVC [38,39]. However, these studies employed limited contamination through several inoculated sites of 1 cm<sup>2</sup> across the device, including a blood pressure gauge and N95 respirators. Post-exposure, these areas were sampled with a pre-moistened cotton swab and placed in a solution for bacterial recovery, or in the case of respirators, the areas were cut out. Another study evenly distributed a small inoculum onto the membranes of stethoscopes which were placed onto plate count agar for bacterial recovery [37]. These approaches are unfeasible to demonstrate the efficacy of a UVC chamber for medical devices that are more complex in shape or technical design. Therefore, this study employed a different



**Fig. 3.** Bacterial load and log reduction factors of rinsed medical instruments. Dots represent individual data points. Bar charts are shown as mean ± standard deviation (SD). (A) Log-transformed colony forming units (CFU/mL) of *S. aureus* under control and treatment conditions collected from dental tools (n = 3 or 4) (4 independent experiments), round and sharp ended orthodontic pliers (n = 3) (3 independent experiments), handpieces (n = 4) (2 independent experiments). \* (p < 0.05), \*\* (p < 0.01), \*\*\* (p < 0.001), ns (non-significant) (paired t-test except for data on handpieces, Wilcoxon matched-pairs signed rank test) (B) Log<sub>10</sub> reduction factors for the different medical instruments.

methodology through partial submersion of the devices (with exception of the thermistors), thereby enabling a more accurate representation of natural contamination. Studies using UVC mercury chambers for disinfection of devices naturally contaminated during routine use, have been described for some medical devices such as electronic devices [30,31], intravaginal probes [32–34] and flexible fiberoptic laryngoscopes [35]. This approach could result in a more thorough understanding of the effectiveness as well as the practical implementation of the UVC chamber in daily practice as compared to artificial contamination in laboratory setting. However, the complexity of this study design may render on-site studies for each potential instrument of interest impractical and may not always be necessary depending on the result obtained on an artificially contaminated instrument.

The instruments subjected to UVC LED disinfection in our study were semi-critical devices that require at least HLD [3]. For this reason, the devices (with exception of the thermistors) were contaminated with a high inoculum, such that upon disinfection a  $\log_{10}$  reduction factor of 6 could be demonstrated, which is the endpoint for HLD recommended by the Food and Drug Administration [40]. Using different types of dental tools, all having the same rod-like structure but with variable ends, we demonstrated that the efficacy of the UVC LED chamber for disinfection of medical instruments was influenced by their technical and/or structural characteristics as reduction levels varied from a 3  $\log_{10}$  to 6  $\log_{10}$  reduction for the non-rinsed dental tools. For instance, UVC disinfection of the chisel needle, the dental tool with the lowest complexity, consistently resulted in undetectable growth, averaging a reduction of 6.25  $\log_{10}$  with the droplet line technique. In contrast, the sawtooth needle and hammer shaped needle demonstrated an average reduction of 5.18  $\log_{10}$  and 3.23  $\log_{10}$ , respectively. The conflicting results of the sawtooth needle could potentially be explained by the different volumes of inoculum remaining between the ridges after excess droplet removal. Rinsing for 3 s under a stream of water as a simplified cleaning process reduced the bacterial load by 2.7  $\log_{10}$  and improved the final outcome of the disinfection procedure for all dental tools. Bacterial growth remained undetected with the droplet line technique for all dental tools. However, a few colonies were still observed when the rinsed and treated instruments were imprinted onto agar. Thorough cleaning before HLD or sterilization is required to ensure the subsequent technique is effective at inactivating microorganisms [3]. The incorporation of a more thorough cleaning method may further improve the outcome, and when combined with UVC LED, may offer an effective disinfection alternative.

UVC was unable to decontaminate handpieces, both in the rinsed and non-rinsed conditions. These instruments are complex in nature due to technical characteristics and therefore require a multistep decontamination process [41,42]. This complexity likely prevented UVC from accessing the internal part of the device, leading to the absence of bacterial reduction. However, this does not negate the possibility of the use of a UVC chamber as an alternative disinfection step for the exterior of the device prior to lubrication and HLD or sterilization, as the exterior is commonly cleaned/disinfected through a manual procedure involving a disinfectant cloth [43]. A similar trend was observed with the orthodontic pliers, where only partial reduction (2  $\log_{10}$  or less) was obtained as the contaminating bacteria inside the hinge were likely not accessible to UVC. Implementation of an adequate disinfection alternative would be beneficial as these instruments frequently experience corrosion through heat sterilization, the commonly used technique in practice [18]. We were able to show a complete reduction of the bacterial load on an oral/nasal thermistor, demonstrating an average reduction of 4.90  $\log_{10}$ , which was further confirmed using the instrument imprinting procedure. Currently, these instruments are either single-use or reusable provided they are manually disinfected. A UVC disinfection chamber may offer the possibility for reprocessing single-use thermistors, on condition that it complies with guidelines and regulations which could contribute to more sustainable practices [44,45]. Although the reuse of single-use devices is performed worldwide, it remains a matter of controversy and is unauthorized in several countries [3,46,47]. In addition, the error-prone manual disinfection of reusable thermistors could be replaced by a standardized process utilizing UVC chambers, if preceded by a manual cleaning step.

Standardization of UV disinfection is still under development whereby a variety of other standards are currently being used, such as the EN14885 standard developed for chemical disinfection [29]. However, the recent British standard BS8628:2022 [48] specifically developed for validation of UV(C) disinfection systems is increasingly used and entails laboratory tests conducted on stainless steel disc carriers. Here, we demonstrated that the disinfection efficacy on a simple surface (i.e. a standard Petri dish or stainless-steel disc) does not necessarily reflect the disinfection performance on a contaminated instrument. This highlights the importance of assessing the efficacy on real-life instruments and the standards used to validate UVC chambers should consider this.

Here, we evaluated the efficacy of a UVC LED chamber for disinfection of medical instruments, focusing on how the shape and technical characteristics of these instruments influence the disinfection efficacy. While the study was limited to the clinically relevant pathogen *S. aureus* in the absence of organic soling, our results on medical devices, combined with existing literature on bacterial species [49–51] suggest that variations in the design of instruments have a greater impact on the reduction efficacy than variations between bacterial species. However, future research should include a broader variety of microorganisms such as yeasts, molds, and viruses to provide more insight into the required dose for effective decontamination of medical devices. Additionally, only a limited subset of instruments was examined, while an extensive list of unexplored devices remains. These instruments could be investigated in follow-up research. The use of UVC as an alternative disinfection strategy for medical devices should be preceded by efficacy testing for each candidate device to ensure the safe use of UVC chambers in medical settings, mitigating the risk of acquiring a nosocomial infection.

## 5. Conclusions

This study evaluated the efficacy of UVC LED for disinfection of a range of medical instruments that have not been previously assessed. Using *S. aureus* as the test organism, we demonstrated the high disinfection capacity of the novel ZAPARAY™ UVC LED chamber on a simple surface, such as a Petri dish which is smooth, flat, non-porous, and without cavity. However, varying results obtained from medical instruments indicate that instrument shape and technical characteristics may substantially influence the efficacy of UVC disinfection. These factors should be considered, along with the initial level of contamination after routine use, cleaning



steps, and the desired level of disinfection (non-critical vs. semi-critical devices). Our findings highlight the potential of UVC chambers as a standardized substitute for specific (manual) disinfection procedures, but they may not be suitable for all instruments. Given the variability in efficacy, it is essential that their use is preceded by efficacy testing for each specific type of instrument. In this manner, the study provides novel insights into the broad applicability of UVC LED chambers and offers a benchmark for future research in exploring the potential of UVC LED.

## Funding

This work was supported by the Flanders Innovation and Entrepreneurship (VLAIO) [Grant numbers HBC.2022.0304 and HBC.2021.0849]. The funding source was not involved in any aspects of the study design, the execution, the analysis, the writing, or the decision to submit for publication.

## Data availability statement

Data will be made available on request.

## CRediT authorship contribution statement

**Hannah Siwe:** Writing – review & editing, Writing – original draft, Methodology, Investigation, Formal analysis. **Annelies Aerssens:** Methodology, Investigation, Formal analysis. **Mieke V. Flour:** Methodology, Investigation, Formal analysis, Conceptualization. **Silke Ternest:** Methodology. **Leen Van Simaey:** Methodology. **Duncan Verstraeten:** Resources, Conceptualization. **Alain F. Kalmar:** Resources. **Isabel Leroux-Roels:** Writing – review & editing, Supervision, Resources, Investigation, Formal analysis, Conceptualization. **Philip Meuleman:** Writing – review & editing, Supervision, Resources, Investigation, Formal analysis, Conceptualization. **Piet Cools:** Writing – review & editing, Supervision, Resources, Investigation, Formal analysis, Conceptualization.

## Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: H.S. is an employee of eLEDricity, M.V.F. was an employee and shareholder of eLEDricity until halfway through the study. D.V. and A.F.K. are shareholders of eLEDricity but were not involved in the analysis or interpretation of the data. All other authors declare they have nothing to disclose.

## Acknowledgements

We thank the staff of the departments at the Ghent University Hospital, and the private practice end-users for their contribution to the study.

## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.heliyon.2024.e37281>.

## References

- [1] World Health Organization, Global report on infection prevention and control, Retrieved 21 November 2023, from, <https://www.who.int/publications/i/item/9789240051164>, 2022.
- [2] World Health Organization, "Minimum Requirements for Infection Prevention and Control programmes.", 2019.
- [3] Center for Disease Control and Prevention, Guideline for disinfection and sterilization in healthcare facilities, Retrieved 20 November 2023, from, <https://www.cdc.gov/infectioncontrol/pdf/guidelines/disinfection-guidelines-H.pdf>, 2008, May 2019.
- [4] G. McDonnell, P. Burke, Disinfection: is it time to reconsider Spaulding? *J. Hosp. Infect.* 78 (3) (2011) 163–170.
- [5] P.M. Southworth, Infections and exposures: reported incidents associated with unsuccessful decontamination of reusable surgical instruments, *J. Hosp. Infect.* 88 (3) (2014) 127–131.
- [6] W.A. Rutala, D.J. Weber, Reprocessing semicritical items: outbreaks and current issues, *Am. J. Infect. Control* 47 (2019) A79–A89.
- [7] A.M. Laheij, et al., Healthcare-associated viral and bacterial infections in dentistry, *J. Oral Microbiol.* 4 (2012).
- [8] M.J. Alfa, et al., Manual methods are suboptimal compared with automated methods for cleaning of single-use biopsy forceps, *Infect. Control Hosp. Epidemiol.* 27 (8) (2006) 841–846.
- [9] N. Kenters, et al., Infectious diseases linked to cross-contamination of flexible endoscopes, *Endosc. Int. Open* 3 (4) (2015) E259–E265.
- [10] J.T. Walker, et al., Cleanability of dental instruments – implications of residual protein and risks from Creutzfeldt-Jakob disease, *Br. Dent. J.* 203 (7) (2007) 395–401.
- [11] D.L. Buescher, et al., Disinfection of transvaginal ultrasound probes in a clinical setting: comparative performance of automated and manual reprocessing methods, *Ultrasound Obstet. Gynecol.* 47 (5) (2016) 646–651.
- [12] N. Musee, et al., Occurrence, effects, and ecological risks of chemicals in sanitizers and disinfectants: a review, *Environmental Chemistry and Ecotoxicology* 5 (2023) 62–78.

- [13] K. Romero Starke, et al., Are healthcare workers at an increased risk for obstructive respiratory diseases due to cleaning and disinfection agents? A systematic review and meta-analysis, *Int J Environ Res Public Health* 18 (10) (2021).
- [14] J.-Y. Maillard, Resistance of bacteria to biocides, *Microbiol. Spectr.* 6 (2) (2018), <https://doi.org/10.1128/microbiolspec.arba-0006-2017>.
- [15] A.D. Russell, Bacterial resistance to disinfectants: present knowledge and future problems, *J. Hosp. Infect.* 43 (1999) S57–S68.
- [16] B.F. Leas, et al., AHRQ comparative effectiveness technical briefs, in: *Environmental Cleaning for the Prevention of Healthcare-Associated Infections*, Agency for Healthcare Research and Quality (US), Rockville (MD), 2015.
- [17] M. Dioguardi, et al., Management of instrument sterilization workflow in endodontics: a systematic review and meta-analysis, *Int J Dent* 2020 (2020) 5824369.
- [18] A. Wichelhaus, et al., Corrosion of orthodontic pliers using different sterilization procedures, *J. Orofac. Orthop./Fortschritte der Kieferorthopädie* 65 (6) (2004) 501–511.
- [19] N.G. Reed, The history of ultraviolet germicidal irradiation for air disinfection, *Publ. Health Rep.* 125 (1) (2010) 15–27.
- [20] W.A. Hijnen, E.F. Beerendonk, G.J. Medema, Inactivation credit of UV radiation for viruses, bacteria and protozoan (oo)cysts in water: a review, *Water Res.* 40 (1) (2006) 3–22.
- [21] J. Henderson, et al., Field study of early implementation of UV sources and their relative effectiveness for public health and safety, *J. Occup. Environ. Hyg.* 19 (9) (2022) 524–537.
- [22] S.E. Beck, et al., Comparison of UV-induced inactivation and RNA damage in MS2 phage across the germicidal UV spectrum, *Appl. Environ. Microbiol.* 82 (5) (2015) 1468–1474.
- [23] C.A. Juan, et al., The chemistry of reactive oxygen species (ROS) revisited: outlining their role in biological macromolecules (DNA, lipids and proteins) and induced pathologies, *Int. J. Mol. Sci.* 22 (9) (2021).
- [24] A. Rodger, UV absorbance spectroscopy of biological macromolecules, in: G.C.K. Roberts (Ed.), *Encyclopedia of Biophysics*, Springer Berlin Heidelberg, Berlin, Heidelberg, 2013, pp. 2714–2718.
- [25] L.T. Budnik, L. Casteleyn, Mercury pollution in modern times and its socio-medical consequences, *Sci. Total Environ.* 654 (2019) 720–734.
- [26] T. Nicolau, et al., A comprehensive analysis of the UVC LEDs' applications and decontamination capability, *Materials* 15 (8) (2022).
- [27] *Nature Photon*, **Haitz's law**, from, <https://doi.org/10.1038/nphoton.2006.78>, 2007.
- [28] T. Bhattarai, A. Ebong, M.Y.A. Raja, A review of light-emitting diodes and ultraviolet light-emitting diodes and their applications, *Photonics* 11 (6) (2024) 491.
- [29] N. Demeersseman, et al., Shedding a light on ultraviolet-C technologies in the hospital environment, *J. Hosp. Infect.* 132 (2023) 85–92.
- [30] E.M. Allen, et al., The effectiveness of germicidal wipes and ultraviolet irradiation in reducing bacterial loads on electronic tablet devices used to obtain patient information in orthopaedic clinics: evaluation of tablet cleaning methods, *J. Hosp. Infect.* 105 (2) (2020) 200–204.
- [31] M. Resendiz, et al., Comparative effectiveness of rapid-cycle ultraviolet decontamination to chemical decontamination on high-touch communication devices, *Am. J. Infect. Control* 47 (9) (2019) 1135–1139.
- [32] M. Pichon, et al., Decontamination of intravaginal probes infected by human papillomavirus (HPV) using UV-C decontamination system, *J. Clin. Med.* 8 (11) (2019).
- [33] C. Kyriacou, et al., Time-effectiveness and convenience of transvaginal ultrasound probe disinfection using ultraviolet vs chlorine dioxide multistep wipe system: prospective survey study, *Ultrasound Obstet. Gynecol.* 60 (1) (2022) 132–138.
- [34] G. Kac, et al., Evaluation of a new disinfection procedure for ultrasound probes using ultraviolet light, *J. Hosp. Infect.* 65 (2) (2007) 163–168.
- [35] U.C. Ezeh, et al., The effectiveness of ultraviolet smart D60 in reducing contamination of flexible fiberoptic laryngoscopes, *Laryngoscope* 133 (12) (2023) 3512–3519.
- [36] J. Wezgowiec, et al., Evaluation of antimicrobial efficacy of UVC radiation, gaseous ozone, and liquid chemicals used for disinfection of silicone dental impression materials, *Materials* 15 (7) (2022).
- [37] G. Messina, et al., A new UV-LED device for automatic disinfection of stethoscope membranes, *Am. J. Infect. Control* 43 (10) (2015) e61–e66.
- [38] G. Moore, et al., Use of UV-C radiation to disinfect non-critical patient care items: a laboratory assessment of the Nanoclave Cabinet, *BMC Infect. Dis.* 12 (2012) 174.
- [39] J.L. Cadnum, et al., Effectiveness of ultraviolet-C light and a high-level disinfection cabinet for decontamination of N95 respirators, *Pathog Immun* 5 (1) (2020) 52–67.
- [40] Food and Drug Administration, Medical washers and medical washer-disinfectors - class II special controls guidance document for the medical device industry and FDA review staff, Retrieved 17 November 2023, from, <https://www.fda.gov/medical-devices/guidance-documents-medical-devices-and-radiation-emitting-products/medical-washers-and-medical-washer-disinfectors-class-ii-special-controls-guidance-document-medical>, 2002.
- [41] G. Smith, A. Smith, Microbial contamination of used dental handpieces, *Am. J. Infect. Control* 42 (9) (2014) 1019–1021.
- [42] E.C. Deasy, et al., Effective cleaning and decontamination of the internal air and water channels, heads and head-gears of multiple contra-angle dental handpieces using an enzymatic detergent and automated washer-disinfection in a dental hospital setting, *J. Hosp. Infect.* 128 (2022) 80–88.
- [43] G.W.G. Smith, et al., Survey of the decontamination and maintenance of dental handpieces in general dental practice, *Br. Dent. J.* 207 (4) (2009). E7–E7.
- [44] Food and Drug Administration, Enforcement priorities for single-use devices reprocessed by third parties and hospitals, Retrieved 25 November 2023, from, <https://amdr.org/wp-content/uploads/2021/03/1168-.pdf>, 2000.
- [45] A. Molero, et al., Sustainability in healthcare: perspectives and reflections regarding laboratory medicine, *Ann Lab Med* 41 (2) (2021) 139–144.
- [46] European Commission, National rules on reprocessing of single-use devices, Retrieved 25 November 2023, from, [https://health.ec.europa.eu/medical-devices-topics-interest/reprocessing-medical-devices/national-rules-reprocessing-single-use-devices\\_en](https://health.ec.europa.eu/medical-devices-topics-interest/reprocessing-medical-devices/national-rules-reprocessing-single-use-devices_en), 2022.
- [47] W. Popp, et al., What is the use? An international look at reuse of single-use medical devices, *Int. J. Hyg Environ. Health* 213 (4) (2010) 302–307.
- [48] 8628 Bs, Disinfection using ultraviolet radiation. Methods for quantitative testing of automated ultraviolet disinfection activities by direct illumination. Determination of Bactericidal, Mycobactericidal, Sporicidal, Yeasticidal, Fungicidal, Virucidal and Phagocidal Activities. 2022, British Standard Institution, 2022.
- [49] H. Schöbel, et al., Antimicrobial efficacy and inactivation kinetics of a novel LED-based UV-irradiation technology, *J. Hosp. Infect.* 135 (2023) 11–17.
- [50] D.-K. Kim, D.-H. Kang, J.L. Schottel, UVC LED irradiation effectively inactivates aerosolized viruses, bacteria, and fungi in a chamber-type air disinfection system, *Appl. Environ. Microbiol.* 84 (17) (2018).
- [51] D.-K. Kim, S.-J. Kim, D.-H. Kang, Bactericidal effect of 266 to 279nm wavelength UVC-LEDs for inactivation of Gram positive and Gram negative foodborne pathogenic bacteria and yeasts, *Food Res. Int.* 97 (2017) 280–287.