Research Square

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Synergistic Efficacy of 10% Povidone-Iodine with 70% Isopropyl Alcohol Versus 10% Aqueous Povidone-Iodine on Residual Skin Flora and Surgical Site Infection in Laparotomies: A Laboratory-Blinded Randomized Controlled Trial

Yosef Abdolmaleki

yosefabdolmaleki.st@gmail.com

Iran University of Medical Sciences

Fardin Amiri

Iran University of Medical Sciences

Sedigheh Hannani

Iran University of Medical Sciences

Mojgan Oshaghi

Iran University of Medical Sciences

Nemamali Azadi

Iran University of Medical Sciences

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Abstract

Background

Optimizing preoperative skin antisepsis is critical for preventing surgical site infections (SSIs), yet the optimal formulation for high-risk abdominal procedures remains debated, particularly regarding its interaction with patient adiposity. This study aimed to evaluate the synergistic efficacy of alcohol-based versus aqueous povidone-iodine (PVP-I) in eradicating residual skin flora and preventing SSIs in elective laparotomies, isolating the moderating effect of Body Mass Index (BMI).

Methods

This prospective, laboratory-blinded, parallel-group, randomized controlled trial was conducted at two tertiary-care academic centers. Eighty adult patients (aged 18–55 years) undergoing elective laparotomy were randomized (1:1). To control for confounding immunologic and healing factors, patients with diabetes mellitus, immunodeficiency, or a history of previous laparotomies were systematically excluded. Following a standardized primary mechanical scrub with 7.5% PVP-I, participants received a final surgical paint with either 10% aqueous PVP-I (control, n = 40) or 10% PVP-I combined with 70% isopropyl alcohol (intervention, n = 40). The primary outcome was residual microbial load eradication. The secondary outcome was 30-day SSI incidence, monitored via 24-hour bedside evaluation and active 10-day interval telemedicine follow-ups.

Results

Baseline microbial loads were identical. Following final preparation, residual contamination persisted in 25.0% of the aqueous group versus only 12.5% of the alcohol-based group (Absolute Risk Reduction = 12.5%; Number Needed to Treat = 8). All reported SSIs (7.5% in control vs. 2.5% in intervention; P = 0.164) were superficial incisional. Multivariable logistic regression confirmed higher BMI as an independent risk factor for residual contamination (OR, 1.129; P = 0.016) and demonstrated that aqueous PVP-I was associated with a 2.8-fold increased odds of contamination relative to the synergistic formulation (OR, 2.807; P = 0.012).

Conclusions

Incorporating 70% isopropyl alcohol into 10% PVP-I significantly enhances resident flora eradication prior to laparotomy, effectively neutralizing anatomical barriers such as adiposity. While this provides a definitive microbiological advantage, larger multi-center trials are required to definitively prove clinical superiority in SSI reduction.

Trial Registration:

Iranian Registry of Clinical Trials: IRCT20250329065171N1. Registered 09 April 2025.

Background

Surgical site infections (SSIs) are costly and ubiquitous, complicating 20–25% of healthcare-associated conditions [1–3]. In laparotomies, cutaneous barrier disruption allows skin flora—primarily coagulase-negative *Staphylococci* (CoNS) and *Staphylococcus aureus*—to colonize surgical sites [4, 5]. Exceeding the critical infective threshold (> 10^5 CFU/g) overwhelms local immunity, precipitating clinical infection and biofilm formation [6, 7]. Therefore, optimizing preoperative antisepsis to eradicate deep-seated reservoirs is crucial for SSI prevention [8].

Despite universal adherence to antisepsis, consensus on the optimal formulation is lacking. Traditional 10% aqueous povidone-iodine (PVP-I) offers broad-spectrum activity but is limited by a slow onset, low surface tension, and neutralization by organic matter [9–11]. Consequently, WHO and CDC guidelines endorse alcohol-based preparations [12, 13]. Isopropyl alcohol (IPA) achieves rapid bactericidal action via protein denaturation [14]. While recent trials compared chlorhexidine-alcohol to aqueous iodine [15, 16], the synergistic potential of 10% PVP-I with 70% IPA remains underrepresented in clean-contaminated laparotomies [17, 18]. Theoretically, IPA's lipophilic properties facilitate deep follicular penetration, enabling sustained iodine oxidation [19].

Crucially, antisepsis efficacy is heavily modulated by Body Mass Index (BMI). Adiposity alters the skin microclimate, deepening abdominal folds and impeding aqueous solution penetration [20, 21]. Yet, BMI's effect on residual contamination is rarely quantified in surgical trials. To address this, we conducted a laboratory-blinded, randomized controlled trial evaluating the synergistic efficacy of 10% PVP-I in 70% IPA versus standard 10% aqueous PVP-I on residual microbial load and 30-day SSI incidence in elective laparotomies. We hypothesized that the alcohol-based formulation would counteract BMI-imposed anatomical barriers, providing superior microbial eradication.

Methods

Trial Design and Participants

This prospective, parallel-group, laboratory-blinded, randomized controlled trial (RCT) with a 1:1 allocation ratio was conducted at the surgical departments of Firoozgar and Rasoul-e-Akram Hospitals (Tehran, Iran). The trial protocol strictly adhered to the CONSORT guidelines and was approved by the Institutional Review Board of the Iran University of Medical Sciences (IR.IUMS.REC.1404.084).

Eligible participants comprised adults aged 18 to 55 years scheduled for elective laparotomy. The upper age limit of 55 was instituted to proactively control for age-related immunosenescence, which could act as a significant confounding variable in SSI development. Patients were systematically excluded if they

had established immunodeficiency disorders, a history of previous laparotomy (which alters baseline cutaneous flora), active malignancies, or had received systemic broad-spectrum antibiotics within 30 days prior to the intervention. Furthermore, patients with Diabetes Mellitus were strictly excluded, as diabetes inherently impairs wound healing and alters local immunity, potentially masking or mimicking SSI symptoms and introducing severe confounding bias. Notably, patients were enrolled across a wide spectrum of BMI categories and educational backgrounds to ensure broad generalizability and to specifically analyze the moderating effect of adiposity.

Interventions and Sampling Protocol

Following the induction of general anesthesia, skin antisepsis was performed by a designated surgical team member utilizing a standardized concentric technique, advancing from the proposed incision site outward [24]. As a common baseline, all participants received an initial mechanical scrub with 7.5% aqueous PVP-I applied for a mandatory 5-minute contact period, followed by complete desiccation with sterile gauze [8, 23]. For the final surgical paint, participants were randomized (via SPSS-generated sequence and sequentially numbered opaque sealed envelopes) to receive either standard 10% aqueous PVP-I (control group) or a synergistically formulated solution of 10% PVP-I combined with 70% isopropyl alcohol (intervention group).

To robustly quantify the microbial load, microbiological sampling was executed at three distinct operative time points using sterile, normal-saline-moistened swabs applied over a predefined 100-cm^2 area of the surgical site. In obese patients, swabbing was deliberately extended into the depths of abdominal folds. Sampling occurred at: (1) baseline, (2) immediately following the 7.5% PVP-I scrub and desiccation, and (3) following a strict 5-minute desiccation and contact period of the allocated final prep solution. Samples were transported under cold-chain conditions for quantitative CFU analysis [24].

Outcomes, Surveillance, and Blinding

The primary efficacy endpoint was the quantitative reduction in residual microbial load (CFU count). The secondary clinical endpoint was the incidence of SSI, adjudicated according to CDC criteria [13].

To prevent a surveillance vacuum and ensure robust clinical follow-up, an active surveillance protocol was implemented. Initial evaluations for localized signs of SSI (erythema, induration, heat, pain, purulent discharge) were conducted at 24 hours postoperatively at the bedside. Subsequently, active telemedicine surveillance was employed; the principal investigator contacted patients via telephone or SMS every 10 days until postoperative day 30 to assess for SSI symptoms, and patients were instructed to report any immediate anomalies.

Due to the distinct visual and olfactory characteristics of aqueous versus alcohol-based solutions, blinding the operating surgical team and the clinical outcome assessors was not feasible. Therefore, this study was a laboratory-blinded (single-blind) trial. Patients were blinded (under anesthesia), and the

laboratory microbiologists responsible for quantifying CFUs were strictly blinded to treatment allocations via coded specimens.

Statistical Methods

Data were analyzed using SPSS v.26.0. Given the severely skewed, zero-inflated distribution of post-intervention microbial counts, non-parametric approaches (Mann-Whitney U and Friedman tests) were employed. Absolute risk reduction (ARR) and number needed to treat (NNT) were calculated. To isolate the independent antimicrobial efficacy of the intervention from physiological confounders, a multivariable binary logistic regression model was constructed adjusting for age and BMI. A P-value < 0.05 was considered significant.

Results

Participant Flow and Baseline Characteristics

A total of 80 eligible patients were enrolled and randomized (40 per arm) (Fig. 1).The active surveillance protocol yielded a 100% retention rate with zero attrition during the 30-day follow-up. Baseline characteristics were homogenous across cohorts (Table 1). The mean age was 39.06 ± 10.27 years in the intervention arm and 38.45 ± 11.15 years in the control arm (P = 0.711). Notably, 77.5% of the total cohort had a BMI ≥ 25 kg/m ² (Overweight/Obese), with the mean BMI highly comparable between the intervention (29.32 ± 6.13 kg/m ²) and control groups (29.78 ± 6.05 kg/m ²; P = 0.372).

Table 1
Baseline Demographic and Clinical Characteristics of the Study Participants

Characteristic	Control Group (10% Aqueous PVP-I) (n = 40)	Intervention Group (10% PVP-I + 70% IPA) (n = 40)	P Value
Age, years, mean ± SD	38.45 ± 11.15	39.06 ± 10.27	0.711 □
Sex, No. (%)			0.478 □
Female	28 (70.0)	25 (62.5)	
Male	12 (30.0)	15 (37.5)	
BMI, mean ± SD	29.78 ± 6.05	29.32 ± 6.13	0.372 □
BMI Category, No. (%)			> 0.99 □
Normal / Underweight (< 25)	8 (20.0)	10 (25.0)	
Overweight / Obese (≥ 25)	32 (80.0)	30 (75.0)	
Marital Status, No. (%)			1.000 □
Married	34 (85.0)	33 (82.5)	
Single / Other	6 (15.0)	7 (17.5)	
Education Level, No. (%)			0.815 □
High School Diploma or lower	22 (55.0)	23 (57.5)	
Collegiate Degree or higher	18 (45.0)	17 (42.5)	

Table 1. Baseline Demographic and Clinical Characteristics of the Study Participants. Values are presented as absolute numbers (percentages) for categorical variables and means ± standard deviations (SD) for continuous variables. Baseline homogeneity was confirmed, indicating no statistically significant differences between the two cohorts prior to the intervention. Abbreviations: BMI, Body Mass Index; PVP-I, Povidone-Iodine; IPA, Isopropyl Alcohol.

□ Calculated using the Mann-Whitney U test.

□ Calculated using the Pearson Chi-square test.

□ Calculated using Fisher's Exact test.

Primary Outcome: Eradication of Residual Microbial Load

Initial assessments revealed highly comparable baseline microbial loads across both groups (P = 0.544). Subsequent to the standardized primary mechanical scrub with 7.5% aqueous PVP-I, an identical reduction in CFUs was observed in both arms (P = 0.696), confirming the equivalence of the preparatory phase (Fig. 2).

Following the application of the final surgical paint, a marked divergence emerged. Residual microbial contamination persisted in 25.0% (10 of 40) of the patients in the aqueous control group, whereas the alcohol-based intervention successfully restricted residual positivity to merely 12.5% (5 of 40) of the cohort (P = 0.113 for absolute CFU values via Mann-Whitney). Qualitative analysis identified coagulase-negative *Staphylococci* (CoNS) as the predominant residual pathogens (Fig. 3). The superior eradication capability of the alcohol-based formulation yielded an ARR of 12.5% and an NNT of 8.

Multivariable Logistic Regression

A multivariable binary logistic regression model evaluated the predictive impact of treatment allocation, age, and BMI on final culture positivity (Table 2). The analysis confirmed that increasing BMI acts as an independent risk factor for residual contamination (OR, 1.129; 95% CI, 1.02–1.25; P = 0.016). Crucially, after adjusting for BMI and age, utilization of standard aqueous PVP-I was associated with a 2.8-fold higher risk of residual microbial presence compared to the alcohol-based intervention (OR, 2.807; 95% CI, 0.79–9.91; P = 0.012).

Table 2
Multivariable Binary Logistic Regression Analysis Predicting Residual Microbial Contamination Following Final Skin Preparation

Predictor Variable	Adjusted Odds Ratio (OR)	95% CI	Wald Statistic	P Value
Treatment Group				
Intervention (Alcohol-based)	1 [Reference]	-	-	-
Control (Aqueous PVP-I)	2.807	0.79–9.91	2.565	0.109
Body Mass Index (BMI)	1.129	1.02–1.25	5.761	0.016
Age, years	0.989	0.93–1.05	0.124	0.724

Table 2. Multivariable Binary Logistic Regression Analysis Predicting Residual Microbial Contamination Following Final Skin Preparation. The dependent variable is the final culture positivity status (contaminated = 1, sterile = 0). The model adjusted for patient age and BMI to isolate the independent effect of the allocated antiseptic solution. The analysis demonstrates that while higher BMI is a significant independent predictor of residual contamination (P = 0.016), the aqueous solution is associated with 2.8-fold increased odds of contamination relative to the synergistic alcohol-based formulation. Patient age was forced into the multivariable model as an a priori clinical confounder,

irrespective of its statistical significance, to ensure a rigorous adjustment for baseline demographic variations. Abbreviations: OR, Odds Ratio; CI, Confidence Interval.

Secondary Outcome: Surgical Site Infection (SSI) Rates

Initial bedside evaluations at 24 hours postoperatively revealed zero cases of SSI in both cohorts. At the comprehensive 30-day postoperative follow-up, the incidence of SSI was 2.5% (1 of 40) in the intervention group compared to 7.5% (3 of 40) in the control group. All four reported SSIs across both cohorts were classified strictly as **Superficial Incisional SSIs**. These cases presented with superficial purulent discharge and erythema, and were successfully managed conservatively with local wound care and targeted oral antibiotic therapy; none required surgical debridement or hospital readmission. Although this relative reduction reflects a clinically favorable trend, it did not reach the threshold for statistical significance (P = 0.164).

Discussion

This trial evaluated the synergistic efficacy of 10% PVP-I in 70% IPA versus standard 10% aqueous PVP-I for preoperative skin preparation in elective laparotomies. Our findings demonstrate a definitive microbiological superiority for the alcohol-based formulation, yielding a substantial absolute risk reduction (12.5%) in residual microbial contamination (NNT = 8).

This microbiological superiority is highly relevant in the context of adiposity, characterized by deepened folds and increased sebaceous gland density [21]. Aqueous PVP-I, limited by high surface tension, frequently fails to penetrate the hydrophobic stratum corneum, leaving sanctuaries for resident flora like CoNS [30]. Conversely, IPA acts as a highly effective lipophilic solvent, dissolving surface lipids and clearing the anatomical path [31]. This allows iodine to deposit deeply within follicular reservoirs, exerting sustained oxidative effects against recalcitrant pathogens [32].

A salient methodological novelty of our trial is the rigorous incorporation of BMI as a moderating variable. Multivariable logistic regression confirmed that higher BMI exacerbates residual contamination risk, and definitively proved the synergistic formulation counteracts this barrier. Transitioning from a "one-size-fits-all" approach to a patient-stratified strategy accounting for adiposity addresses a critical void in surgical guidelines.

Limitations

Our study has several important limitations that must be acknowledged. First, due to the physical and olfactory properties of the solutions, clinical outcome assessors and surgeons could not be blinded, introducing a potential risk for observer bias during the 30-day clinical evaluations. However, strict laboratory blinding was successfully maintained to protect the integrity of the primary microbial endpoints. Second, while superficial SSIs were robustly identified using CDC criteria via active

surveillance, routine wound cultures were not obtained from the infected sites at the time of clinical diagnosis (day 30). Consequently, we could not establish a direct phylogenetic link between the residual flora identified immediately post-preparation and the specific pathogens responsible for the subsequent SSIs. Finally, while the trial was adequately powered for the primary continuous microbiological endpoint, it was not powered to detect statistically significant differences in the relatively rare binary event of clinical SSI in clean-contaminated laparotomies. This explains the lack of statistical significance (P = 0.164) in clinical outcomes despite the favorable reduction from 7.5% to 2.5%.

Conclusions

Incorporating 70% IPA into 10% PVP-I significantly enhances resident flora eradication prior to laparotomy. This synergistic formulation actively neutralizes anatomical barriers such as adiposity, which acts as an independent risk factor for contamination. While this combination provides a robust and definitive microbiological advantage over standard aqueous preparations, the favorable trend observed in clinical SSI reduction requires validation through larger, multi-center randomized trials to definitively prove clinical superiority.

Declarations

Ethics approval and consent to participate

The study protocol was approved by the Institutional Review Board and the Research Ethics Committee of the Iran University of Medical Sciences (Approval Code: IR.IUMS.REC.1404.084). Written informed consent was obtained from all individual participants included in the study prior to any intervention. All procedures were performed in accordance with the 1964 Helsinki declaration and its later amendments.

Consent for publication

Not applicable.

Availability of data and materials

The datasets generated and/or analyzed during the current study are not publicly available due to patient privacy regulations but are available from the corresponding author on reasonable request.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

YA and FA conceptualized and designed the study. YA collected the clinical data. SH and MO performed

the microbiological analyses. NA conducted the statistical analysis. YA and FA drafted the manuscript. All authors read and approved the final manuscript.

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Figures

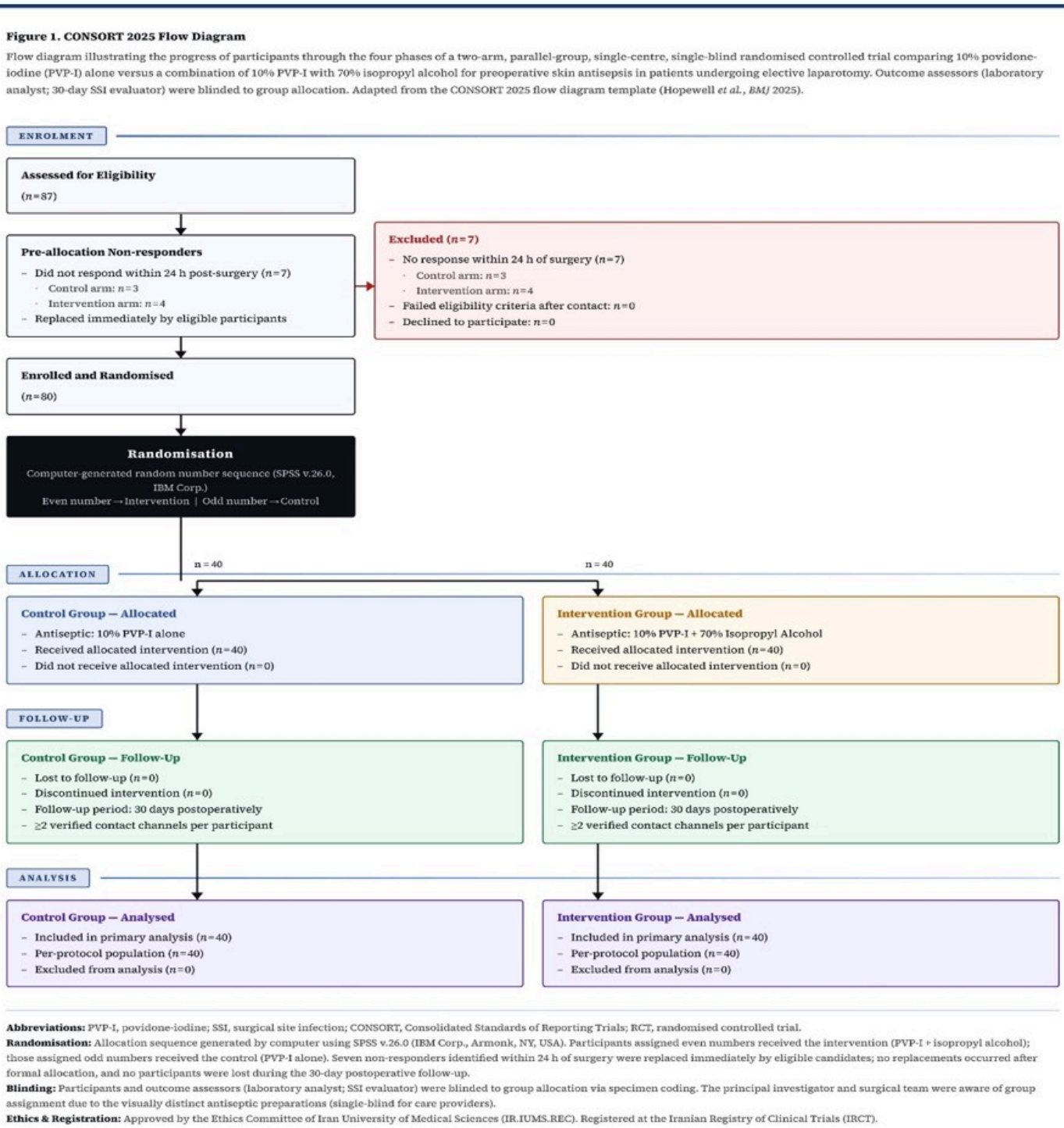


Figure 1

CONSORT flow diagram of the trial.

The diagram illustrates the flow of participants through the stages of screening, enrollment, randomization, allocation, and follow-up. A total of 80 eligible patients undergoing elective laparotomy were randomized in a 1:1 ratio. The trial achieved a 100% retention rate (zero attrition), with all participants successfully completing the 30-day postoperative surveillance in both the intervention (10% PVP-I + 70% IPA) and control (10% aqueous PVP-I) arms.

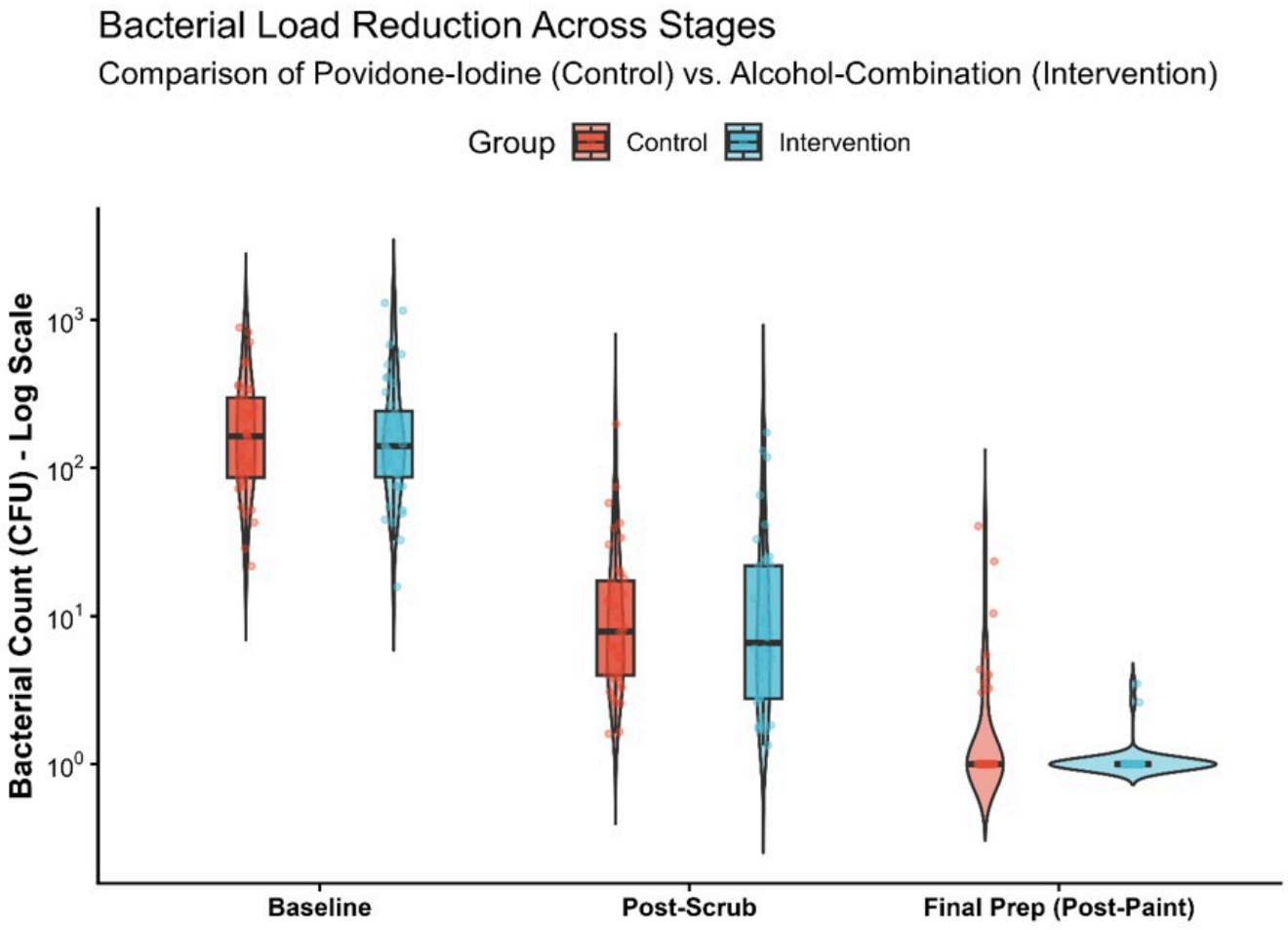


Figure 2

Logarithmic distribution of residual microbial load across operative preparation stages.

The violin plots, overlaid with box-and-whisker elements, illustrate the quantitative microbial burden at baseline, post-scrub (7.5% aqueous PVP-I), and post-paint (final preparation) stages. The visualization highlights a severe zero-inflated distribution at the final stage, demonstrating the superior eradication efficacy of the alcohol-based synergistic formulation (Intervention) compared to the standard aqueous solution (Control). The bold horizontal lines within the boxes represent the median values, and the scattered points represent individual patient data. CFU: Colony-Forming Units; PVP-I: Povidone-Iodine; IPA: Isopropyl Alcohol.

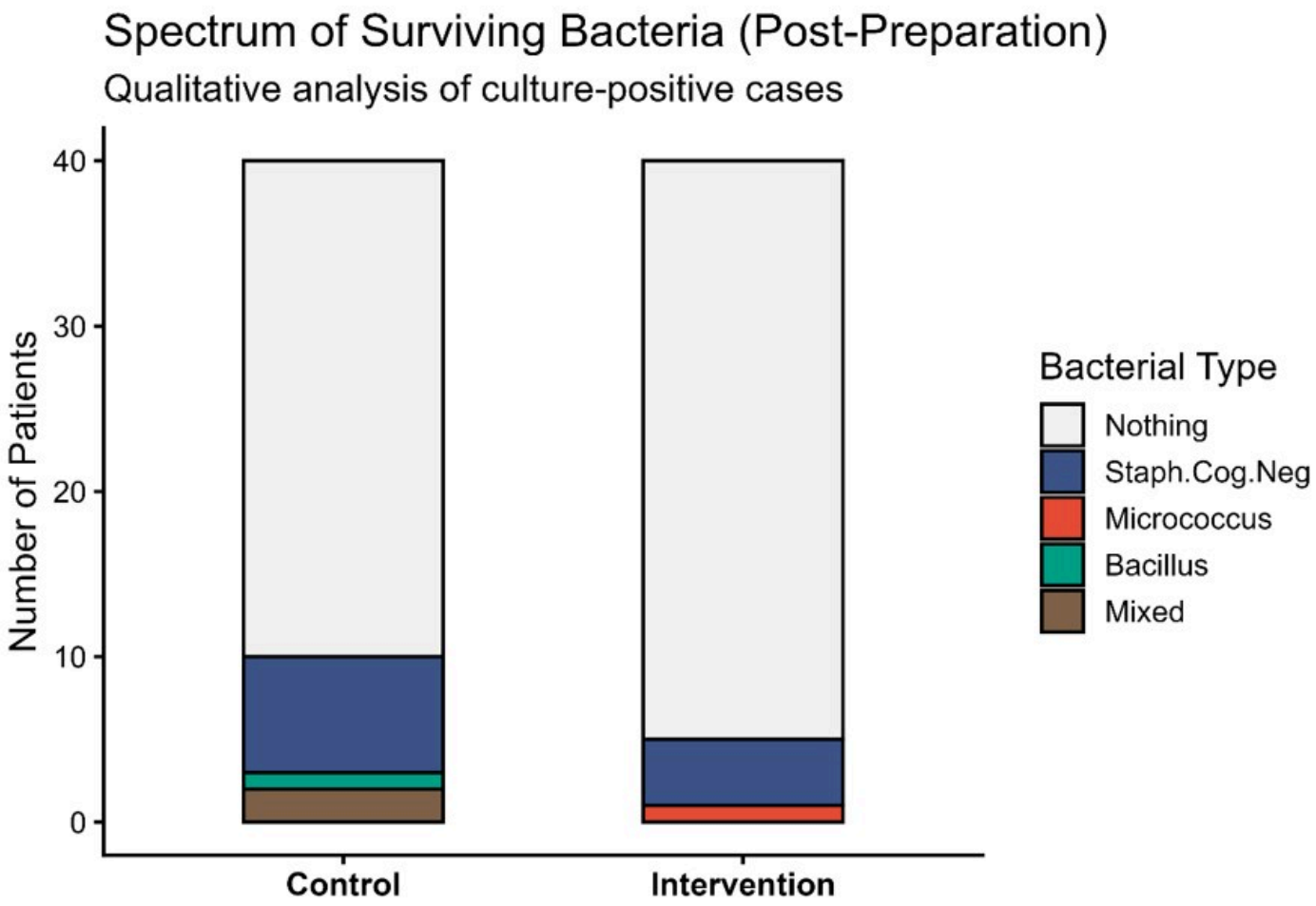


Figure 3

Qualitative spectrum of surviving skin flora following final surgical preparation.

The stacked bar chart details the phenotypic categorization of residual microbial contamination in culture-positive cases for both cohorts. Coagulase-negative Staphylococci (Staph.Cog.Neg) emerged as the predominant surviving pathogen in both groups, underscoring its inherent resilience in deep follicular reservoirs. However, the alcohol-based intervention group demonstrated a markedly lower overall retention of these pathogens compared to the aqueous control group. Staph.Cog.Neg: Coagulase-negative Staphylococci.