Inhalation Formulation & Dosage of PVP Iodine for Respiratory Infections Treatment

Oron Zachar*

Abstract

The aftermath of the Covid-19 pandemic calls for a rethink of pharmaceutical options for treatment of viral respiratory infections. Respiratory infections start primarily at the surface epithelial cells. Being in permanent direct contact with the ambient air, the respiratory system tissue surface may be amenable to topical treatment approaches. We evaluate and define the method and options of translating effective wound care treatments into the realm of respiratory infections treatment and preventions. The goal is a broad-range, safe and cheap anti-viral respiratory infections medication, which can be made globally available for early-stage home use.

In a previous publication we discussed nanoparticles-based formulations. In this article we propose and evaluate a new mode of use of a well-established topical antiseptic – PVP Iodine (PVP-I). In contrast to all previous proposals of PVP-I disinfection nasal sprays or oral gargle, we present a deep and well controlled inhalation protocol to disinfect in one treatment modality both the bronchial tree, upper respiratory nasal and throat tissue simultaneously, using ultrasonic mesh nebulizers.

Molecular iodine is quickly absorbed from the lungs and bronchial tree into the blood, within about 10 minutes. Yet, iodine effectively disinfects most viruses within 1 minute, including SARS-CoV-2. Moreover, the slower to absorb PVP-I complex provides a continuously eluting reservoir that maintains the iodine concentration in the airways surface fluids for several hours. These and other factors unique to inhalation are discussed. We provide: **(i)** Dosage calculation and delivery protocols; **(ii)** Safety analysis based on guidelines, animal trials and WHO review reports; **(iii)** Evaluating published pilot clinical trials of SARS-CoV-2 with related nasal spray or oral gargle PVP-I treatments; **(iv)** Evaluating the potential use and modification of existing off-the-shelf market products to render our proposed treatment immediately available on a global scale.

Our recommended formulations comprise of PVP-I at concentrations 0.5% - 5% and pH between pH=6.5 to pH=7.5 (significantly higher than pH~3.5 of present market products). Inhalation dosage of just 0.5 mL may already be effective, which translates to aerosolizing 2 mL with a common nebulizer device.

PVP-I is globally available over the counter (OTC). The same is true of ultrasonic nebulizers. In practice, all the ingredients are already globally available to consumers. Hence, we hope health and regulatory authorities worldwide will invest the efforts to establish the validity of the proposed treatment. In the meantime, to prevent confusion or misuse by free-willing users and to facilitate clinical evaluation, we outline a prescription for the proper adaptation use of common commercially available market products.

In addition, we propose that the same formulations and protocols can be implemented prophylactically in hospital intensive care units (ICU) for the prevention of ventilator associated pneumonia (VAP).

Contents

Abstract	1
Introduction	2
Conceptual Approach	3
Pathogenesis of Respiratory Infections	4
Dosage Calculation Using Ultrasonic Nebulizers	5
Unique Advantages of PVP-I for Airway Disinfection	5
Analysis	5
PVP-Iodine: A Unique Disinfection Solution	5
In-Vitro PVP-I Tests of Respiratory Infections Pathogens including SARS-CoV-2	8
Clinical Trials with PVP-I	8
Inhalation safety of PVP-Iodine.....	10
Inhalation of PVP-I Clinical & Animal Testing.....	11
Targeted Dosage Deposition by Aerosol Inhalation	11
Conclusions: Proposed Formulation & Dosage of PVP-Iodine for Inhalation Treatment.....	14
Collecting the Threads.....	14
Using Present Market Products – An Immediate Realization Potential	15
Hospital Ventilator Associated Pneumonia (VAP) Prevention Application.....	16
References	16

Introduction

It doesn't need a pandemic to appreciate the significance of viral respiratory infections for public health. Viral respiratory tract infections are the most common illnesses worldwide, resulting in a wide range of severities, from the common cold to severe life-threatening respiratory tract infections. According to the World Health Organization (WHO), respiratory infectious diseases take first place in the ranking of the burden of disease measured by years lost through death or disability [1]. Nearly 25% of all deaths among children less than 5 years of age in developing countries are caused by acute respiratory infections (ARI). Any given year, the economic cost of ARI is significant. In the United States, consumers spend about \$2 billion per year for over-the-counter medications for ARI, and the total annual cost of ARI managed in outpatient settings is estimated to be more than \$10 billion [2]. In the USA alone, it is estimated that the cost to employers of patients with respiratory infections is more than \$100 Billion annually, including costs of medical treatment and time lost from work [3]. Somehow, this state of affairs seems to be assumed as immutable by both the wide public, national health authorities, and pharmaceutical research companies.

One of the technical problems, of developing pharmaceutical treatments for ARI, is that ARI is not one disease. Rhinoviruses are the most frequent agents and are mostly limited to the naso-pharyngeal tissue. Other common respiratory viruses include influenza, parainfluenza, respiratory syncytial virus (RSV), adenovirus, human metapneumovirus, human coronavirus, and enteroviruses, all of which have the capacity to spread deeper (see Figure-1) and induce a more severe illness. Hence, there is a need for a broad range antiviral drug to realize our goal.

The Covid-19 epidemic highlighted the factual status of lack of widely accessible, cheap and safe, broad spectrum anti-viral medication for respiratory infections. Ideally, we would like to have a medication that is so safe as to be permitted for sale OTC, similar to OTC pain and fever medications. The preferred clinical goal is to have a first-line treatment available for use by patients at home with the appearance of early symptoms of a respiratory infection, before it propagates deep into the lungs. At this stage, the infection is primarily limited to the upper regions of the bronchial tree and the extrathoracic regions (nasal and throat tissue).

The general effectiveness of povidone-iodine (PVP-I) as a microbicide that inactivate bacteria and viruses is extensively established and reviewed in the literature [4]. For respiratory infections, all previous disinfection aerosol proposals discuss nasal sprays or oral spray/gargles and have major deficits, including: (a) very limited reach to key respiratory tissues of interest – the deep nasal cavity, back of the throat, and the tracheal-bronchial tree; (b) very uneven and uncontrolled deposition on surface tissue; and (c) significant user dependent variance in the application process. All of these deficits are surmounted by our proposed implementation of ultrasonic nebulizers for the disinfection aerosol delivery. Yet, more specific issues of safety and irritability need to be analyzed in the context of such deep respiratory system deposition of a disinfection aerosol.

Figure-1: Many respiratory infections are quite localized to the upper respiratory system – extrathoracic airway (ET), tracheobronchial airway (TB). Most respiratory infections are initiated in the upper respiratory system, before propagating deeper in advanced stages of the infection.

Conceptual Approach

In the medical literature, there have been some discussions and even clinical explorations of respiratory infections treatment with topical application of disinfectants, particularly PVP-Iodine, limited to spray for nasal infections and/or oral gargles. Our proposed treatment formulation and associate use of ultrasonic nebulized aerosol inhalation delivery have multiple advantages:

- Reach into the bronchial tree and the even the deep lungs.
- Controlled dosage delivery.
- Controlled and predictable deposition in different locations of the respiratory system.
- Uniform dosage deposition over airway surface.
- Simultaneous treatment of the respiratory tract, throat, and nasal cavities.

- vi. Low dependence and low variance in the application of the human end-user.
- vii. Formulation optimized for anti-viral respiratory tract application (unlike prior formulations which were mostly left in the form for topical skin bacteria disinfection).

Pathogenesis of Respiratory Infections

Viral respiratory infections are initiated when virus particles are inhaled and infect primarily the respiratory mucosal surface epithelial cells [5]. It is common in medicine to regard the bronchial tree and the lungs as internal organs. Yet, topologically the respiratory system surface is in direct constant contact with the ambient air in the same way and level of contact as the nostrils. Therefore, at a clinical conceptual level, the respiratory system may be thought of as an external tissue surface amenable to topical treatment. Consequently, we proclaim that topical treatment substances, such as applicable to wound care infection treatment and prevention, might also be applicable to respiratory infections. The main issues to consider are of (i) safety and side effects specific to the respiratory system tissue; and (ii) the technical method of delivery of the required effective antimicrobial inhibitory concentration (IC) to the target inner regions of the respiratory system.

In the context of Covid-19, it is thought than one of the factors for the greater infectivity of SARS-CoV-2 over SARS-CoV-1 is its greater affinity (or bonding) for the upper respiratory tract at the initiation of the infection [6]. In the respiratory tract, peak SARS-CoV-2 load is observed at the time of symptom onset or in the first week of illness (see Figure-2), with subsequent decline thereafter (even as symptoms may get more severe, as the infections spread to deep regions of the lungs and further organs the body), which indicates the highest infectiousness potential just before or within the first five days of symptom onset [6]. Altogether, the pathogenesis of respiratory infections in general and the Covid-19 epidemics, in particular, highlights the need for early-stage home medication as key to (i) prevent both the interpersonal spread of infection to avoid future epidemics; and (ii) prevent the intrapersonal spread of infection from the original upper more benign upper respiratory system location to the more severe deep lungs and other body organs.

Figure-2: The SARS-CoV-2 viral load in the upper respiratory peaks in the first week of infection and declines significantly thereafter, even as symptoms become more severe. Viral culture from PCR positive upper respiratory tract samples has been rarely positive beyond 9 days of illness (adapted from [7]). Therefore, we estimate that the window of opportunity is essentially limited to the first week of infection and early symptoms, for focused upper respiratory system treatment to prevent progression and spread of the Covid-19 infection.

Dosage Calculation Using Ultrasonic Nebulizers

We summarize the calculation of the required quantity of aerosolized solution dosage (ASD) of solution to put in the nebulizer, needed in order to achieve a desired delivered solution dosage (DSD) to the bronchial tree (BT) target tissue and extrathoracic (ET) tissue (nasal+pharyngeal), with common ultrasonic nebulizer devices. Using common market ultrasonic nebulizers with 5 μm droplet size, only about 50% of aerosolized ASD quantity is inhaled, and only 25% of the inhaled quantity is deposited in the BT tissue and another 25% is deposited in the ET tissue. Therefore, in order to properly target the BT tissue, one needs to put in the nebulizer device a solution quantity ASD which is at least eight times (8X) the desired deposited solution dosage (DSD) to the bronchial tree (BT) target tissue, i.e.

$$\text{ASD} = 8 * \text{DSD} \quad (\text{Eq.1})$$

Next, we summarize the calculation of minimum required deposited solution dosage (DSD) to the bronchial tree (BT) target tissue, in order to achieve a desired target inhibitory concentration (IC) of the disinfectant active ingredient in the airway surface liquid (ASL) of the bronchial tree (BT). For healthy adults, the volume of ASL of the BT is about 1mL. If the aerosolized disinfection solution has an initial solution concentration (ISC), the required quantity (in mL) of deposited solution dosage (DSD) is

$$\text{DSD} = \text{IC}/(\text{ISC} - \text{IC}) \quad (\text{Eq.2})$$

For example, assume using a disinfection initial solution concentration $\text{ISC}=1\%$. For achieving at target $\text{IC}=0.5\%$ in the ASL, one needs to put in the nebulizer to aerosolize $\text{DSD}=1 \text{ mL}$ of the disinfection solution. For achieving at target $\text{IC}=0.2\%$ in the ASL, one needs to put in the nebulizer to aerosolize $\text{DSD}=0.25 \text{ mL}$ of the disinfection solution.

Advantages of PVP-I for Airway Disinfection

From the safety perspective, being already in medical use for sensitive tissue disinfection, including ophthalmic and nasal treatments, PVP-I safety profile is very promising. Little more needs to be done to formally establish PVP-I safety as aerosol inhalation medication. From the effectiveness perspective, a key advantage is due to an anomalous property of PVP-I. As illustrated in Figure-3, contrary to common intuition, under dilution of PVP-I 10% solution, the disinfecting free iodine concentration does not decrease, but actually *increases with dilution*, following a bell-shaped curve, reaching a maximum at approximately 0.1% strength PVP solution and only then decreasing with further dilution. Therefore, unlike with common antibiotics, we expect the treatment is not sensitive to the dilution factors of deposition in the target airway surface liquid (ASL) which may vary among individual patients or infection development stages. For example, starting with a significantly effective and well tolerated 2% PVP-I concentration, even a 5X dilution (to 0.4% PVP-I) upon deposition in the respiratory tissue surface liquid might not degrade much the disinfection potency.

Analysis

PVP-Iodine: A Unique Disinfection Solution

In more than 150 years of use for medical disinfection, including application to wounds, oral, nasal, and ophthalmic tissue, Iodine hasn't elicited bacterial or viral resistance. Iodine may have too many mechanisms of action for bacteria to adapt to [8]. Presently on the market there are three primary formulations of iodine: (a) A tincture, commonly at 2% concentration; (b) "Lugol's iodine", which has no alcohol, has twice the mass of potassium iodide as of elemental iodine; and (c) PVP-Iodine (PVP-I) water-based solutions, commonly at $\text{pH } 2.0 < \text{pH} < 5.5$. The focus of our analysis is on the potential use of PVP-I solutions. We shall argue that, for anti-viral respiratory infection purposes, it may be **beneficial to implement concentration around 2%, and delivered near neutral pH ($\text{pH} \sim 7$)**.

PVP-I Special Properties

Povidone-iodine (PVP-I) came into commercial use in 1955. It is on the World Health Organization's List of Essential Medicines. PVP-I is globally available for sale over the counter. PVP-I is an iodophor solution comprising a water-soluble complex of iodine and the solubilizing polymer carrier polyvinylpyrrolidone (PVP). In aqueous solution, a

dynamic equilibrium occurs between free iodine and the PVP-I complex. In terms of concentration labeling, a 10% w/v PVP-I solution, contains 1% w/v total titratable iodine. Yet, this is not the actively available iodine. The PVP-I complex functions as an eluting reservoir, releasing iodine to maintain a stable equilibrium concentration of free iodine. It is only the free iodine which is an active disinfectant, exhibiting a broad range of microbicidal activity against bacteria, fungi, protozoa, and viruses [9]. As the dissolved free iodine is used up when binding to other molecules, further iodine is released from the PVP-I complex, until the available iodine is exhausted. The PVP-I action as iodophor improves both solubility and stability while releasing the active iodine gradually from the polymer network over time. Therefore, its residual antimicrobial activity is maintained stable over an extended period, while side effects associated with iodine such as irritation and brown staining on mucous membranes are reduced. Remarkably, after all these years, its precise antimicrobial mechanism of action is still unknown, but it is believed that the active iodine species acts as an oxidizing agent.

PVP-I solutions have an important “concentration anomaly”. Over a certain range of PVP-I concentrations, the free iodine concentration actually increases in more diluted concentrations of povidone-iodine. This paradoxical effect follows a “bell curve” (see Figure-3 below): at low concentrations less than 0.05% lose their povidone-iodine complex characteristics and behave like aqueous iodine, the free iodine concentration rises until peaking at 0.1% PVP-I, then decreasing with further increase PVP-I concentration. Correspondingly, in vitro bactericidal efficacy of povidone-iodine has been shown to increase at more dilute concentrations peaking around 0.1% to 1%, with relatively faster killing rates. **An important beneficial consequence in our context of interest is that, unlike the common situation with disinfection and antibiotic substances, as the deposited PVP-I solution is diluted in the respiratory system airway surface liquid (ASL), the microbicidal effectiveness of the PVP-I may not necessarily be diminished.**

Figure-3: Free molecular iodine dependence on PVP-I solution concentration and pH (adapted from data presented in [10,11]).

For disinfection efficacy, it is not only the free iodine that is important. The iodine reservoir quantity, at higher PVP-I concentration results in longer duration of activity. For example, as the free iodine is used up, the iodine eluting reservoir of a 5% PVP-I is 5X larger than that of 1% PVP-I. Hence, particularly in *clinical dirty conditions*, the extended duration of activity of a 5% PVP-I solution may be more important to generate effective disinfection than the initial peak of iodine concentration upon application. We have found at least one clinical indication of such an effect, but more clinical research is needed for our proposed specific application to respiratory infections.

All of the presently available commercial PVP-I solutions sold on the market are with pH<5.5 (most actually at pH<3.5). Yet, as we argue in the following section, there may be preference for pH~7 for respiratory infections, not

only for better safety matching to the pH of the respiratory tissue, but also increased anti-viral potency in spite of the lower free iodine concentration.

The iodine mass portion of the PVP-I complex is about 1/10. i.e., a 1% concentration PVP-I contains 0.1% (i.e., 1 mg/ml = 1,000 mg/L) of Iodine. From Figure-3 we can see that for PVP-I solution at pH~7 the equilibrium concentration of free iodine is roughly 1 mg/L. Therefore, initially upon deposition, the PVP-I complex contains a reservoir of 1,000x the free iodine content, which can be eluted continuously to replace used up free iodine.

Optimizing the Antiviral Effectiveness of PVP-I

Iodine solution stability in water is sensitive to pH. A commonly neglected aspect is that the free iodine in the water solution is not present in one form. Dissolved iodine reacts with the water and can adopt a range of oxidation states, such that various iodine species exist in an aqueous environment: -1 to +5, e.g., -1 (iodide, I⁻); +1 (hypoiodous acid, HOI); +5 (iodate, IO₃⁻), of which only hydrated iodine (I₂), hypoiodous acid (HOI) and iodine cation (H₂OI⁺) possess anti-microbial activity [37]. Of key importance to us will be the balance between elemental I₂ and hypoiodous acid (HOI) content, which can be tuned by pH (see Table-1). In particular – **HIO is approximately 40 times more effective than elemental iodine I₂ against viruses** [12] – a fact that is largely neglected in the medical infections treatment literature and research data. The misguided common wisdom and consideration of USP requirements, in present market commercial products, seems to be that the target active ingredient is just the elemental I₂ content. Therefore, **low pH (pH<5.5) useful for high elemental I₂ content (see Table-1), is the present commercial market products standard. In contrast, we argue that a different standard, of around neutral pH~7, is preferred for an anti-viral medication formulation for respiratory infections** in order to, (a) not alter the respiratory tract natural pH, and (b) increase anti-viral potency with higher content of the HIO.

pH	Iodine		
	I ₂ (%)	HIO (%)	OI ⁻ (%)
5	99	1	0
6	90	10	0
7	52	48	0
8	12	88	0.005
9	-	-	-

Table-1: Effect of pH on the speciation of iodine (0.5% titratable iodine) [12]

Generally, elemental iodine is primarily effective against bacterial spores and protozoan cysts, whereas hypoiodous acid (HIO) is known to be an effective bactericide and virucide. For example, elemental iodine was found to be 2–3 times more effective against *Entamoeba histolytica* cysts than HIO, whereas HIO was found to be approximately 40 times more effective than elemental iodine against viruses [12].

Microbial group	Disinfection effectiveness of different iodine species
Bacteria (vegetative)	HIO > I ₂
Bacteria (spores)	I ₂ > HIO
Viruses	HIO > I ₂
Protozoa ^a	I ₂ > HIO

Figure-2: Trends in the disinfection power of different iodine species [12].

An important factor is the absorption time from the lungs into the blood stream. Elemental iodine is absorbed to the blood with a half-life of about 10 minutes. Therefore, low concentrations of PVP-I <0.1% would have an effective action time of about 10 minutes post inhalation. The absorption of larger molecules is mostly proportional to the

molecular size. PVP used for PVP-I solutions is on average of size 40 kDa. Drawing from information on inhaled albumin (68 kDa) and α 1-antitrypsin (45–51 kDa) having a half-life of multiple hours (since T_{max} in the blood is reached after more than 12 hours) [13], we can assume at least one hour of near original PVP concentration after the initial deposition. Therefore, moderate PVP-I concentration of 1% or more are expected to maintain the eluted free iodine concentration roughly stable for at least 1 hour post initial deposition.

In-Vitro PVP-I Tests of Respiratory Infections Pathogens including SARS-CoV-2

There are multiple reviews in the literature of in-vitro testing of the deactivation effectiveness of PVP-I on both viruses and bacteria. Many of the relevant respiratory infection pathogens show high susceptibility to PVP-I, with better than 4Log₁₀ (>4Log₁₀) reduction within 30 seconds [14]. Multiple in-vitro tests with SARS-CoV-2 have been recently published and reviewed [15]. For in-vitro test, it is important to distinguish clean from dirty conditions, where the dirty conditions include interfering substances. Already from bacterial tests, it appears the total reservoir of Iodine is important, leading to 1% PVP-I being significantly more effective than 0.1% under dirty conditions [16]. Recall, this is in contrast with the naïve expectation that the higher free Iodine content of 0.1% PVP-I would make it more effective. The general explanation in the literature is that more free iodine gets inactivated under dirty conditions, and the continuous eluting of iodine from the higher concentration PVP-I is making up for it by sustaining the concentration over a longer period of time.

There are several recently published SARS-CoV-2 in-vitro tests of PVP-I application. In one test, Four off-the-shelf products of the BETADINE® brand, at PVP-I concentrations w/v of 10%, 7.5%, 1.0% and 0.45%, were tested adapting the protocol from the EN14476 disinfectant testing methodology. All products demonstrated virucidal activity against SARS-CoV-2, corresponding to ≥ 4 log₁₀ reduction of virus titre, within 30 seconds of contact [18].

In another example, PVP-I was tested against *Klebsiella pneumoniae* and *Streptococcus pneumoniae* according to bactericidal suspension test EN13727 and against SARS-CoV, MERS-CoV, and influenza virus A subtype H1N1 according to virucidal suspension test EN14476. A PVP-I 7% gargle/mouthwash was diluted 1:30 with water to a concentration of 0.23% (the recommended concentration for home use in Japan) at room temperature under clean conditions and dirty conditions. The 0.23% concentration was effective for better than 4 Log₁₀ reduction factor within 30 seconds under dirty conditions on all tested bacteria and viruses [14].

A problem for our purpose with most standardized in-vitro tests, such as EN13727 for bacteria and EN14476 for viruses, is the limited focus of testing on the 1-minute mark. The standard 1-minute mark may be of direct relation to an expected clinical scenario of irrigation disinfection (as indeed is the case for ophthalmic treatment). But for our clinical case of interest, of a protected deposition followed by gradual absorption (with at least 10 minutes or even 60 minutes of residence time), there would be great interest and more relevance for having in-vitro test under dirty conditions which track the disinfection level over a period of at least 5-30 minutes. Such studies are remarkably rare, to the point of being anecdotal, in the published literature.

Oral gargle is very popular in Japan. Hence, in this context several unique studies of PVP-I were conducted by Japanese groups, which apparently are also less blindly fixated on the European ENXXXXX protocols. A study examined the effect of oral organic matter on in vitro short-time killing activity of PVP-I. Gargling samples collected from healthy volunteers served for PVP-I 0.23-0.47% testing. PVP-I deactivation of *Pseudomonas aeruginosa* within 60 seconds was not diminished in the presence of oral organic matter [17].

Clinical Trials with PVP-I

PVP-I is commonly used to sterilize the ocular surface prior to surgical procedures, and as treatment for adenoviral conjunctivitis. Yet the benefits of PVP-I have not been clearly documented in existing clinical management guidelines for ocular surface conditions [19]. In a pivotal clinical trial published in 1991, during an 11-month period, topical 5% PVP-I was used in over 3,000 patients to prepare the conjunctiva, in addition to customary prophylactic

antibiotics. A significantly lower incidence of culture-positive endophthalmitis ($P < 0.03$) was observed. Use of topical 5% PVP-I in the eye in over 3000 patient was not associated with any adverse reactions [20].

From the scientific research point of view, a major deficit of most published clinical trials is the focus on clinical outcomes. There is a scientific evidence gap between on the one hand (i) In-vitro test of disinfection quantification over a 1-minute duration in non-live media; and on the other hand (ii) clinical outcomes such as “incidence of endophthalmitis after intraocular surgery” with no clearly documented causal factors. In between these two extremes there should be interest in establishing decolonization evidence – for the direct disinfection effectiveness on a target live tissue under clinical conditions. For example, comparing the microbial load on the target tissue before/after the disinfection treatment application. We found very few such trials in the published literature.

3M company developed a unique PVP-I 5% gelling product pre-operative nasal and skin antiseptic. In a clinical trial, the 3M product sustained a greater than 2Log10 reduction in *S. Aureus* CFU from nasal swabs even 12h post treatment [21].

Figure-4: 3M Skin and Nasal Antiseptic reduction of *S. aureus* in the nares post-prep for subjects with baseline counts of at least 3.4 Log₁₀, [21].

A comparison of 5% vs. 1% povidone-iodine solutions in preoperative cataract surgery antisepsis was conducted in a prospective randomized double-blind study of 100 patients. The ocular surface of the eye was irrigated with PVP-I by dripping 2 ml of the solution from a syringe directly on to the eye for 1 minute. After a further minute a second swab was taken. Bacterial cultures showed a decrease in median colony forming units (CFU) pre-irrigation to post-irrigation drop of 96.7%, compared with a lesser reduction, drop of 40%, when using 1% PVP-I. The authors speculated that PVP-I 1%, although initially more bactericidal, has a lower reservoir of available iodine which is exhausted when the bacterial load is increased [22].

The bactericidal activities of PVP-I, chlorhexidine gluconate (CHG) and cetylpyridium chloride (CPC) gargles were compared. In vivo, with 6 subjects in each group, reduction rate in the oral bacterial count after gargling was measured. The mean reduction rate in bacterial count immediately after gargling was 99.4% for PVP-I, 59.7% for CHG and 97.0% for CPC [23], establishing the effectiveness of PVP-I in-vivo oral secretions environment.

A phase-II clinical trial of adenovirus eye infection treatment (~50 patients in each arm) evaluated the treatment effectiveness of 0.6% PVP-I. The proportion of adenoviral eradication by day 3 was 32% with PVP-I vs 8.7% with vehicle control [24]. We note that a similar trial, with 0.6% PVP-I, on bacterial eye infection did not show any effect. This is simply a manifestation of the fact that PVP-I is not a magic bullet, as is well documented by in-vitro testing, some viruses and bacteria take more than 60 minutes for significant deactivation by PVP-I. Yet this is not the case with any of the respiratory infections pathogens that we have reviewed [14].

In conclusion: (i) It appears PVP-I disinfection properties are effective also in a live tissue environment, including nasal tissue, in clinical settings; (ii) Most of the evidence is in the use of 5% PVP-I; (iii) There is good evidence for

anti-viral effectiveness of PVP-I intermediate concentrations greater than 0.5% (>0.5%) and up to 5% (<5%) in biologically relevant dirty conditions; (iv) The pH of PVP-I solutions is rarely, if ever, specified in the published clinical trials – manifesting the common ignorance about the importance of this parameter for the properties of PVP-I solutions. Yet, since test formulations are commonly derived from commercial products, or following USP guidelines, we assume all of them were with $2 < \text{pH} < 5.5$.

Covid-19 Clinical Trials of PVP-I

Presently there are multiple clinical trials underway around the world of PVP-I treatment or prophylaxis for Covid-19, all of them evaluating oral gargle or/and nasal spray methods of application. Some published pilot trials are already available. The outcome of these clinical trials is indicative of both (a) the realistic disinfection potential, and (b) the safety, of PVP-I on live respiratory system tissue similar to bronchial tree, nasal, and throat tissue.

In a clinical study from 2020, a total of 6,692 patients were evaluated for feasibility, usability and tolerability of the 0.5% PVP-I gargles and nasal drops. Tolerance was assessed in terms of altered taste, staining of teeth or nasal skin or irritation in the nose. None reported any serious reactions or adverse effects following use of 0.5% PVP-I [25]. This large population survey gives support to the safety, tolerability, and rareness of allergic reaction to respiratory tissue application of PVP-I at concentrations $\sim 0.5\%$.

A pilot, open labeled, randomized, parallel study compared the effect of 30 seconds, 3 times/day gargling using 1% commercial PVP-I and tap water on SARS-CoV-2 viral clearance among COVID-19 patients. Patients were instructed to gargle 10ml of Betadine® gargle/mouthwash for 30 seconds, three times per day for 7 days. Progress was monitored by day 4, 6 and 12 PCR (Ct value) from nasopharyngeal and oropharyngeal samples. The swabs were taken *before* the early morning gargle. Five confirmed Stage 1 COVID-19 patients were in each arm. Already at day 4 in 100% of patients for the PVP-I tested negative, compared with 40% for tap water gargle, and 20% control group [26].

In contrast, there was no observed effect on Covid-19 PCR test outcome in another recent pilot study, with 12 patients in each arm, also using a 1% PVP-I solution. Intervention consisted of 4 successive mouthwashes and gargles with 25 mL of 1% aqueous PVP-I, followed by one 2.5-mL nasal pulverization of the same solution into each nostril using an intranasal mucosal atomization device connected to a 5-mL syringe while sniffing. treatment sessions were conducted 4 times a day for 5 days [27].

Inhalation safety of PVP-Iodine

Iodine is an essential nutrient, with well-established mechanisms of absorption and clearance. Allergic reactions to Iodine are rare. An inhaled dosage of 1 mL of a 1% concentration PVP-I contains 0.1% Iodine (i.e., 1 mg/ml = 1,000 mg/L) of Iodine. The PVP carrier component safety profiling has been extensively documented in the literature. National regulatory bodies have concluded that PVP usage is safe as both a food-grade additive and also as a pharmaceutical excipient. Absorption studies revealed that there is no or very limited absorption of PVP in humans. Extensive toxicity studies confirmed that the PVP is a biologically inert substance. PVP is found to be non-toxic, non-irritant, non-sensitizer. PVP was found to be safe for oral and topical applications in any manner, including ocular. PVP-I is routinely used in ophthalmology for both treatment and prophylaxis of eye infections [28].

In a study of 23 adult patients in Japan, with chronic respiratory diseases showing repeated infections, patients gargled PVP-I, 4 times/day over periods lasting from several months up to over 2 years. Episodes of infections with *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *H. influenzae* were reduced by about 50%. More importantly, from the safety point of view this clinical study indicates remarkable tolerance of oral and throat soft tissue to daily repeated and extended duration topical exposure to PVP-I contact in respiratory infected patients [29].

Absorption into the Body

The recommended daily intake of iodine by the WHO is 0.15 mg, with acceptable daily intake of PVP up to 50 mg/kg body weight [30]. When healthy patients were given more than 100 times the recommended intake of iodine (i.e., 15 mg) daily for 38 days, there were no deleterious effects [31]. Therefore, even 15 times per day applications of our proposed treatment are established to be safe as far as absorption to the body is concerned. Hence, there remains to consider the organ specific safety considerations for the respiratory tract tissue.

pH Balance

The pH of barrier organs is quite variable. A healthy lung has neutral pH~7 [30], while in cystic fibrosis the lung pH is acidic. Reduced airways surface liquid pH facilitates respiratory bacterial infection. Increasing a commercial PVP-I product from an original pH~4 to a desired pH~7 with phosphate buffer decreased ocular irritancy of the antiseptic [30]. Present commercial PVP-I products comprise a pH buffer which stabilizes their pH to $2 < \text{pH} < 5$, most commonly around pH~3. Consequently, even a 1/10 dilution with water does not change much the pH level [32]. pH values below 7.0 decrease ciliary beating in vitro (50% at pH = 5.5 and almost 100% at pH = 3.5) [33]. Hence, we speculate that when sensitivity of ciliated cells to PVP-I solutions was found in various tests or clinical trials, it was due to the pH of the solution and not the iodine (as far as we know, this suggestion is not stated in the literature). Therefore, if starting with a commercial PVP-I product solution, it is highly preferred to adjust the pH to pH~7.

Inhalation of PVP-I Clinical & Animal Testing

To our knowledge, deep inhalation testing (i.e., into the bronchial tree) was thus far explored only with iodine vapor inhalation (not droplet aerosol, which is our intended treatment). Rapid absorption of iodine vapor following inhalation is supported by studies in rats, mice, dogs, sheep and monkeys, with a half-life of 10 min [12]. Similarly, human volunteers exposed to radioactive elemental iodine vapor by inhalation showed clearance with a half-life of 10 minutes, with the iodine being removed by mucociliary clearance to the gastrointestinal (GI) tract [12].

As previously noted, there is a large body of human clinical research and experience with sinonasal and throat application of PVP-I, and the associated safety considerations have been recently reviewed [31]. For example, 1% PVP-I sinus rinses were very well-tolerated similar to saline irrigations, when used twice daily for 30 days, for post-surgery treatment with acute exacerbations of chronic rhinosinusitis and gram-positive bacteria on culture [34].

Targeted Dosage Deposition by Aerosol Inhalation

Aerosol droplets deposition distribution is dependent upon the droplet size (See Figure-5). Ultrasonic nebulizers generate a well-defined stream of fixed sized droplets. Common market products have droplets around 5 μm , but this can be easily manufactured to have a different value anywhere between 3 μm - 10 μm . Nasal inhalation (with a mask) would result with most aerosol deposited in the nasal cavity. Therefore, for the treatment of any non-nasal section of the respiratory system, oral inhalation is recommended. Moreover, since typically about 25% of inhaled aerosol gets exhaled, exhalation through the nose can deposit significant aerosol in the nasal cavity target even when inhalation is done orally.

As illustrated in Figure-5, by oral inhalation, maximum delivery to the deep lungs occurs with aerosol droplets of size ~3 μm , getting about 30% deposited in the deep lungs (alveoli) tissue. Maximum delivery to the upper bronchial tree (BT generations 0-15) occurs droplets of size ~6 μm , getting about 30% deposited in the TB tissue, with similar 30% deposited in the extrathoracic (ET) tissue and about 15% in the deep lungs. If aerosol of 10 μm droplets is used, about 60% of deposition will occur in the upper extrathoracic (ET) nasal and throat tissue, while only 20% is deposited in the trachea-bronchial (TB) tree and almost nothing reaches the deep lungs tissue. Hence, if needed, the deep lungs can be protected from the solution inhalation simply by choice of the nebulizer droplet size. At droplets size of ~5 μm , a nearly even depositions of 25% is delivered to each of bronchial tree (BT), extrathoracic (ET), deep lungs (alveoli), and in exhalation also to the nasal tissue. Therefore, the available common market mesh nebulizer

products, having droplets size of about $5\mu\text{m}$ are quite suitable for our purpose. Hence, **for dosage calculations, e.g., of delivery to a target bronchial tree (BT) tissue, we shall assume 25% deposition efficiency.**

Figure-5. Primary structures of the respiratory system and associated nanoparticle deposition fractions when breathing at rest. Adapted from [35].

Distribution of Dosage in the Bronchial Tree

The bronchial tree is a branched tubing system. Antimicrobial drugs exhibit concentration-dependent efficacy. Ensuring an appropriate inhibitory concentration (IC) of these drugs in the relevant body fluid is important for obtaining the desired therapeutic action. For inhaled antimicrobials, the relevant body fluid for drug concentration purposes is the airway surface liquid (ASL) [36], also referred to as epithelial lining fluid (ELF). The delivered dose is deposited onto and diluted by the ASL. We argue that dosage planning, correction, and controlled verification can all be achieved by examining the easily accessible trachea and/or primary bronchi (first generation, G1). Based on deposition models (see Fig. 6), the trachea can serve as a good concentration estimator (to within a predictable factor) and effective lower bound for the local delivery concentration in all 10 first generations of the bronchial tree. Therefore, tracheal concentration is an effective, well-defined, and measurable representative of target IC.

Figure-6: Local concentration of inhaled aerosol in different generations of the bronchial tree, where the trachea is generation 0. Adapted from [36].

Dosage Quantification for Achieving Target Disinfection Inhibitory Concentration (IC)

We focus attention on the bronchial tree (BT) as the key new target tissue for disinfection presented in this article. The total surface area of the bronchial tree is about 1 m^2 ($10,000 \text{ cm}^2$). Using the surface area and mucosal thickness data from Table-4 below, we estimate **the combined mucosal volume in the top half of the bronchial tree to be about 1 mL in a healthy adult**. In contrast, the total surface area of the pulmonary alveoli is about 100 m^2 (*i.e.*, $1,000,000 \text{ cm}^2$), with mucosal thickness of about $0.07 \text{ }\mu\text{m}$, resulting in a total ASL volume of 7–10 mL. Therefore, disinfection of the upper bronchial tree at early stages, before the infection has spread deep into the lungs, is the most reasonable and realistic target treatment.

In addition, since the ASL volume of the pulmonary (PL) fluid is about 10X the volume ASL of the trachea-bronchial (BT), where for $5\text{ }\mu\text{m}$ aerosol both PL and TB deposition is about equal fraction of 25% of the inhaled aerosol, we can infer from it that the **concentration of delivered disinfection solution to the pulmonary tissue is about 1/10 the concentration of delivered disinfection solution to the tracheo-bronchial tissue**.

We conclude that, for calculating the inhaled disinfection deposition onto the bronchial tree ASL, about 0.25mL of aerosol liquid gets deposited in the bronchial tree (BT) ASL from each 1 mL of inhaled aerosol (assuming 25% deposition fraction for typical $5 \text{ }\mu\text{m}$ aerosol). Since the BT ASL volume is itself 1mL, it follows that the **disinfection liquid is diluted upon deposition**, as illustrated for example in Table-3.

Table-3. Dilution of inhaled PVP-I 1% solution aerosol upon deposition onto the ASL in the bronchial tree (BT) and resulting effective concentration of PVP-I in the BT ASL. 25% deposition fraction in the BT is assumed for typical $5\text{ }\mu\text{m}$ aerosol.

Inhaled solution [ml]	BT deposited solution dosage (DSD) [ml]	Resulting dilution factor in the ASL of the bronchial tree (BT)	Resulting PVP-I concentration in the ASL from an original initial solution concentration ISC=1% aerosol
1 ml	0.25 ml	20%	0.2%
4 ml	1 ml	50%	0.5%

It is important to take into consideration that the dilution will be larger in an inflammation state with excess bronchi secretions. Therefore, it is highly advantageous to have a disinfection liquid like the PVP-I with a wide range of effective inhibitory concentrations and safety.

Table-4. Surface area and mucosal thickness in parts of the bronchial tree (Adapted from [36]). Generation $k = 0$ is the trachea; $th_{k, PLC}$ is the thickness at generation k of the periciliary layer.

Generation (k) ²	$th_{k, PLC}$ ³ (μm)	Airway surface area (cm^2)
0	6.0	71
1+2	5.2	72
3+4+5	4.2	178
6+7+8	3.5	374
9+10+11	3.1	871
12+13+14	2.8	1631

Breathing Cycle Losses in Aerosol Delivery

Inhalation represents less than half of the duration of a full breathing cycle. Hence, when aerosols are delivered continuously, only about a half of the aerosolized dose is considered to have been delivered effectively. This would be the common situation for home users utilizing standard commercial medicament aerosol devices available for purchase in pharmacies. Even if we assume that a portion of the aerosol cloud remains in the oral cavity in between inhalation, we need to **assume at least about 50% loss of continuously aerosolized dosage** by common market ultrasonic nebulizers. By contrast, using a breath-actuated nebulizer (also available commercially, but presently much more expensive) would correspondingly avoid wastage of disinfection solution during the approximately two thirds of the breathing cycle that is not inhalation.

Conclusions: Proposed Formulation & Dosage of PVP-Iodine for Inhalation Treatment

We culminate the analysis with a specific proposal of optimal formulations and delivery protocols for a treatment of viral respiratory infections. First, we collect the threads of knowledge from our discussion thus far. Then we integrate the conclusions into a specific quantitative frame of a treatment protocol. In addition, we provide a tentative evaluation and guidance for the potential repurposing use of widely available existing market products to realize our proposal, making it possible for off-label use by practicing doctors in any place in the world where such use is permitted.

Collecting the Threads

Goals: A first line, home use, broad range, cheap and widely available antiviral respiratory infections medication, introducing to the market

- i. An acute treatment of viral respiratory infections (such as caused by Influenza, SARS-CoV, and Rhinovirus). We are not interested here in chronic use, nor in regular prophylaxis of viral infections.
- ii. A unified controlled treatment targeting both the trachea-bronchial tree and nasopharyngeal tissue.
- iii. A widely accessible first-line treatment for home use, cheap, and potentially suitable for OTC sale.

Safety: PVP-I solutions at concentrations 0.5% - 5% and buffered to pH~7 can be assumed to have high safety and tolerance level for deposition in the respiratory tract, because

- i. Absorption to the body is no risk at the quantities of relevance. Iodine is an OTC dietary supplement at quantities much higher than what our inhalation treatment protocol calls for. PVP is largely recognized as a mostly inert substance that is cleared from the body without being cumulated in any body tissue.
- ii. PVP-I act as both an eluting reservoir and a sink, maintaining a balanced equilibrium. i.e., the concentration of free iodine in the solution and in the airway surface liquid is stabilized and cannot reach some uncontrol excess.
- iii. Iodine vapor inhalation in work environment safety standards recommendations in USA and EU indicate high tolerance to deep lungs iodine inhalation in general.
- iv. PVP-I solutions at concentrations 0.5% - 5% have been widely used in application to sensitive pre/post-surgical tissue or viral infected tissue, for disinfection of nasal, throat, and ocular treatment. Therefore, we assume that such concentrations are safe also for short term respiratory tract tissue contact.
- v. pH~7 would be significantly more compatible with the airway surface liquid (ASL) and tissue pH. Therefore, direct use of present market products (all with pH<5.5) is not advised (but can be properly adapted as we elaborate below).

Efficacy: PVP-I solutions at a concentration ~1% and buffered to pH~7 can be assumed to have high virucidal effectiveness within about 10 minutes of active disinfection time after deposition in the respiratory tract, since

- i. Tests of PVP-I in concentrations between 0.5% to 5% in clinical settings on human nasal, oral, and ocular tissue showed significant active disinfection properties on both viruses and bacteria. Significant disinfection occurred within 1 minute in multiple test cases.
- ii. At pH~7 there is about 50% of the free iodine in the form of hypoiodous acid (HIO) which is about 40x more virucidal than elemental iodine (I_2).
- iii. Elemental iodine showed clearance from the lungs with a half-life of 10 minutes. Therefore, low concentrations of <0.1% PVP-I would have an effective ~10min of action. Yet, with PVP-I complex molecular weight of ~40K Dalton, solutions at concentrations >0.5% would reside and likely maintain free iodine concentration in the airway surface liquid for at least 60 min and possibly multiple hours (a prediction which can and should be verified clinically). Indeed, one test (by 3M company) with a 5% PVP-I solution showed a disinfection effectiveness >2Log₁₀ lasting for over 6 hours post application.

Dosage calculation: Only around 1/8 of the dosage quantity nebulized continuously by common market nebulizing devices gets deposited into the trachea-bronchial tree and diluted into the airway surface liquid (ASL), because

- i. There is loss of about 50% from the dosage quantity nebulized continuously by common market nebulizing devices, because inhalation is not more than 50% of the breathing cycle.
- ii. At droplets size of ~5 μ m (as for common commercial ultrasonic mesh nebulizers), a nearly even depositions of 25% is delivered to each of bronchial tree (BT), Extrathoracic (ET), the deep lungs (alveoli), and in exhalation also to the naso-pharyngeal tissue.
- iii. The upper trachea-bronchial tree ASL volume is about 1 mL.

Therefore, by way of example, we outline two possible protocols, one based on a 5% solution and the other based on a 1% solution concentration of PVP-I.

Example Protocol-1: Nebulizing 2 mL, 5% concentration PVP-I for inhalation with a continuous nebulizer. 0.25 mL (1/8 of 2mL) gets deposited onto the trachea-bronchial tree (TB), getting diluted into a preexisting 1mL of ASL, resulting with a 1% PVP-I concentration in the ASL of the trachea-bronchial tree.

Example Protocol-2: Nebulizing 4 mL, 1% concentration PVP-I for inhalation with a continuous nebulizer. 0.5 mL (1/8 of 4mL) gets deposited onto the trachea-bronchial tree (TB), getting diluted into a preexisting 1mL of ASL, resulting with a 0.33% PVP-I concentration in the ASL of the trachea-bronchial tree.

Using Present Market Products – An Immediate Realization Potential

There is availability on the market of commercial PVP-I solutions at concentrations of 5%, 1% and also some 0.5%, most of which are labeled for use as “oral gargle”. These can be used without any dilution. Yet, the most widely available PVP-I solutions are 10% and 7.5% PVP-I solutions, which can be routinely found for OTC purchase in pharmacies, drug stores, and online stores. These can be diluted 1/2 with distilled water. All are buffered at pH<5.5, mostly around pH~3.0. The key preparation step is to adjust the pH of the available market product to 6.5<pH<7.5 by using a pH buffer. Popular and very accessible pH buffers include: (i) Potassium phosphate, (ii) Baking Soda.

For demonstration, we used a commercial PVP-I 0.5% Betadine Sore Throat Gargle. We measured its pH=3.0, being stabilized by citric acid. To modify the pH level, we used a pH=7.0 (at 25 degrees Celsius) commercial phosphate buffer (by Rocker). At a 1/10 mixing (adding 1cc of the pH=7.0 buffer to the Betadine original solution), the PVP-I solution was restabilized at a reasonable pH=6.5, up from the original pH=3.0.

The recommended dosages for inhalation are (i) 2mL from a medium concentration solution (2.5%-5%); and (ii) 4mL from a low concentration solution (0.5%-2%), as explained in the two examples which we provided above. The inhalation is best performed using an ultrasonic mesh nebulizer device, which are commonly available for purchase

OTC in pharmacies and online purchase (see illustration of typical devices market in Figure-7). We recommend performing oral inhalation, with exhalation to be done via the nose. In such a method of administration, we expect the aerosol deposition to be distributed to all the respiratory organs, with roughly 25% deposited in the oral/throat region, 25% in the trachea-bronchial tree, 25% in the deep lungs, and 25% in the nasal cavity.

Figure-7: Ultrasonic mesh nebulizers with accessories for nasal inhalation and oral inhalation. It is important to have air inlets on the sides of the oral tube.

Hospital Ventilator Associated Pneumonia (VAP) Prevention Application

For bacterial infections, particularly in the context of preventing hospital intensive care VAP, the same formulations are expected to be applicable. For ventilated patients, an additional risk-reduction benefit of PVP-I inhalation treatment is the possibility of suppression of biofilm formation inside the endotracheal or tracheostomy tube. We here propose that such prophylactic inhalation treatment should become a new standard of care from day one of intubation for all ICU patients.

*Yamor Technologies Ltd.

Funding: This research received no external funding.

Acknowledgments: We thank Scott Hirsch for drawing our attention to recent publications about PVP-I nasal spray clinical trials with SARS-CoV-2.

Conflicts of Interest: Yamor Technologies intends to promote a drug development program based on the framework presented in this article.

References

1. Avendaño Carvajal L., Perret Pérez C. (2020) Epidemiology of Respiratory Infections. In: Bertrand P., Sánchez I. (eds) Pediatric Respiratory Diseases. Springer, Cham. https://doi.org/10.1007/978-3-030-26961-6_28.
2. Eugene D. Shapiro, Epidemiology of acute respiratory infections, Seminars in Pediatric Infectious Diseases, Volume 9, Issue 1, 1998, Pages 31-36, ISSN 1045-1870, [https://doi.org/10.1016/S1045-1870\(98\)80048-4](https://doi.org/10.1016/S1045-1870(98)80048-4).
3. Birnbaum HG, Morley M, Greenberg PE, Colice GL. Economic burden of respiratory infections in an employed population. Chest. 2002 Aug;122(2):603-11. <https://doi.org/10.1378/chest.122.2.603>.
4. Sauerbrei, A. Bactericidal and virucidal activity of ethanol and povidone-iodine. MicrobiologyOpen. 2020; 9:e1097. <https://doi.org/10.1002/mbo3.1097>.
5. Kanta Subbarao, Siddhartha Mahanty, Respiratory Virus Infections: Understanding COVID-19, Immunity, Volume 52, Issue 6, 2020, Pages 905-909, ISSN 1074-7613, <https://doi.org/10.1016/j.immuni.2020.05.004>.
6. Cevik M, Kuppalli K, Kindrachuk J, Peiris M. Virology, transmission, and pathogenesis of SARS-CoV-2 BMJ 2020; 371 :m3862, <https://doi.org/10.1136/bmj.m3862>.

7. Cevik M, Kuppalli K, Kindrachuk J, Peiris M. Virology, transmission, and pathogenesis of SARS-CoV-2 BMJ 2020; 371 :m3862, <https://doi.org/10.1136/bmj.m3862> .
8. Joseph Capriotti, MD, Aron Shapiro and Lauren Lilyestrom, Iodine: An Elemental Force Against Infection, Reviews of Ophthalmology, 1 MAY 2009, <https://www.reviewofophthalmology.com/article/iodine-an-elemental-force-against-infection> .
9. Lepelletier D, Maillard JY, Pozzetto B, Simon A. 2020. Povidone iodine: properties, mechanisms of action, and role in infection control and *Staphylococcus aureus* decolonization. Antimicrob Agents Chemother 64:e00682-20, <https://doi.org/10.1128/AAC.00682-20>
10. Wada, H, Nojima, Y, Ogawa, S, et al. Relationship between virucidal efficacy and freeiodine concentration of povidone-iodine in buffer solution. Biocontrol Sci. 2016;21(1):21-27, <https://doi.org/10.4265/bio.21.21>
11. Zamora JL. Chemical and microbiologic characteristics and toxicity of povidone-iodine solutions. Am J Surg. 1986 Mar;151(3):400-6. [https://doi.org/10.1016/0002-9610\(86\)90477-0](https://doi.org/10.1016/0002-9610(86)90477-0) .
12. WHO (2018). Alternative drinking-water disinfectants - Bromine, iodine and silver. World Health Organization, Geneva, Switzerland, ISBN 978-92-4-151369-2, https://www.who.int/water_sanitation_health/publications/alternative-disinfectants/en/ .
13. Labiris, N. R., & Dolovich, M. B. (2003). Pulmonary drug delivery. Part I: physiological factors affecting therapeutic effectiveness of aerosolized medications. British journal of clinical pharmacology, 56(6), 588–599. <https://doi.org/10.1046/j.1365-2125.2003.01892.x> .
14. Eggers, M., Koburger-Janssen, T., Eickmann, M., & Zorn, J. (2018). In Vitro Bactericidal and Virucidal Efficacy of Povidone-Iodine Gargle/Mouthwash Against Respiratory and Oral Tract Pathogens. Infectious diseases and therapy, 7(2), 249–259. <https://doi.org/10.1007/s40121-018-0200-7> .
15. Yan, C.H. and Bleier, B.S. (2020), Prophylactic and therapeutic topical povidone-iodine in coronavirus disease 2019 (COVID-19): What is the evidence?. Int Forum Allergy Rhinol., 10: 1271-1273. <https://doi.org/10.1002/alr.22735> .
16. Sylvie Salvatico, Catherine Feuillolay, Y. Mas, F. Verrière, Christine Roques. Bactericidal activity of 3 cutaneous/mucosal antiseptic solutions in the presence of interfering substances: improvement of the NF EN 13727 European Standard?. Médecine et Maladies Infectieuses, Elsevier Masson, 2015, 45 (3), pp.89-94. [ff10.1016/j.medmal.2015.01.006ff. fhal-02134801](https://hal.archives-ouvertes.fr/hal-02134801). <https://hal.archives-ouvertes.fr/hal-02134801> .
17. Yoneyama A, Shimizu M, Tabata M, Yashiro J, Takata T, Hikida M. In vitro short-time killing activity of povidone-iodine (Isodine Gargle) in the presence of oral organic matter. Dermatology. 2006;212 Suppl 1:103-8. <https://doi.org/10.1159/000089207> .
18. Centers for Disease Control and Prevention. Guidance for dental settings: interim infection prevention and control guidance for dental settings during the COVID-19 response: centers for disease control and prevention. 2020. <https://www.cdc.gov/coronavirus/2019-ncov/hcp/dental-settings.html> .
19. Edington M, Ramaesh K, Lockington D Virucidal benefits of povidone-iodine use on the ocular surface: a reviewBMJ Open Ophthalmology 2020;5:e000509. <https://doi.org/10.1136/bmjophth-2020-000509> .
20. Speaker MG, Menikoff JA. Prophylaxis of endophthalmitis with topical povidone-iodine. Ophthalmology. 1991 Dec;98(12):1769-75. [https://doi.org/10.1016/s0161-6420\(91\)32052-9](https://doi.org/10.1016/s0161-6420(91)32052-9) .
21. 3M Infection Prevention Division, "Safety & Efficacy Information 3M Skin and Nasal Antiseptic", <https://multimedia.3m.com/mws/media/7167880/3m-skin-and-nasal-antiseptic-safety-and-efficacy-brochure.pdf> .
22. Ferguson, A. W., Scott, J. A., McGavigan, J., Elton, R. A., McLean, J., Schmidt, U., Kelkar, R., & Dhillon, B. (2003). Comparison of 5% povidone-iodine solution against 1% povidone-iodine solution in preoperative cataract surgery antisepsis: a prospective randomised double blind study. The British journal of ophthalmology, 87(2), 163–167. <https://doi.org/10.1136/bjo.87.2.163> .
23. Shiraishi T, Nakagawa Y. Evaluation of the bactericidal activity of povidone-iodine and commercially available gargle preparations. Dermatology. 2002;204 Suppl 1:37-41. <https://doi.org/10.1159/000057723> .

24. Pepose JS, Ahuja A, Liu W, Narvekar A, Haque R. Randomized, Controlled, Phase 2 Trial of Povidone-Iodine/Dexamethasone Ophthalmic Suspension for Treatment of Adenoviral Conjunctivitis. *Am J Ophthalmol*. 2018 Oct;194:7-15. <https://doi.org/10.1016/j.ajo.2018.05.012>.
25. Khan, M. M., & Parab, S. R. (2021). Tolerability and usability of 0.5% PVP-I gargles and nasal drops in 6692 patients: Observational study. *American journal of otolaryngology*, 42(2), 102880. <https://doi.org/10.1016/j.amjoto.2020.102880>.
26. Nurul Azmawati Mohamed, Nizam Baharom, Wan Shahida Wan Sulaiman, Zetti Zainol Rashid, Wong Kon Ken, et. al., Early Viral Clearance among COVID-19 Patients When Gargling with Povidone-Iodine and Essential Oils: A Clinical Trial, *International Medical Journal* Vol. 27, No. 6, pp. 651 - 654, December 2020.
27. Guenezan J, Garcia M, Strasters D, et al. Povidone Iodine Mouthwash, Gargle, and Nasal Spray to Reduce Nasopharyngeal Viral Load in Patients With COVID-19: A Randomized Clinical Trial. *JAMA Otolaryngol Head Neck Surg*. 2021;147(4):400–401. <https://doi.org/10.1001/jamaoto.2020.5490>
28. Grzybowski A, Kanclerz P, Myers WG. The use of povidone-iodine in ophthalmology. *Curr Opin Ophthalmol*. 2018 Jan;29(1):19-32. <https://doi.org/10.1097/ICU.0000000000000437>.
29. Nagatake T, Ahmed K, Oishi K. Prevention of respiratory infections by povidone-iodine gargle. *Dermatology*. 2002;204 Suppl 1:32-6. <https://doi.org/10.1159/000057722>.
30. Mallesh Kurakula, G.S.N. Koteswara Rao, Pharmaceutical assessment of polyvinylpyrrolidone (PVP): As excipient from conventional to controlled delivery systems with a spotlight on COVID-19 inhibition, *Journal of Drug Delivery Science and Technology*, Volume 60, 2020, 102046, ISSN 1773-2247, <https://doi.org/10.1016/j.jddst.2020.102046>.
31. Frank S, Capriotti J, Brown SM, Tessema B. Povidone-Iodine Use in Sinonasal and Oral Cavities: A Review of Safety in the COVID-19 Era. *Ear, Nose & Throat Journal*. 2020;99(9):586-593. <https://doi.org/10.1177/0145561320932318>.
32. Berkelman, R. L., Holland, B. W., & Anderson, R. L. (1982). Increased bactericidal activity of dilute preparations of povidone-iodine solutions. *Journal of clinical microbiology*, 15(4), 635–639. <https://doi.org/10.1128/jcm.15.4.635-639.1982>.
33. Zajac, M., Dreano, E., Edwards, A., Planelles, G., & Sermet-Gaudelus, I. (2021). Airway Surface Liquid pH Regulation in Airway Epithelium Current Understandings and Gaps in Knowledge. *International journal of molecular sciences*, 22(7), 3384. <https://doi.org/10.3390/ijms22073384>
34. Lee VS, Pottinger PS, Davis GE. Tolerability and effectiveness of povidone-iodine or mupirocin versus saline sinus irrigations for chronic rhinosinusitis. *Am J Otolaryngol*. 2020 Sep-Oct;41(5):102604. <https://doi.org/10.1016/j.amjoto.2020.102604>.
35. Cheng YS. Mechanisms of pharmaceutical aerosol deposition in the respiratory tract. *AAPS PharmSciTech* [Internet]. 15(3), 630–640 (2014). Available from: <https://link.springer.com/article/10.1208/s12249-014-0092-0>.
36. Hasan MA, Lange CF. Estimating in vivo airway surface liquid concentration in trials of inhaled antibiotics. *J. Aerosol Med*. 20(3), 282–293 (2007). <https://doi.org/10.1089/jam.2007.0603>.
37. Cooper RA. Iodine revisited. *Int Wound J*. 2007 Jun;4(2):124-37. <https://doi.org/10.1111/j.1742-481X.2007.00314.x>.