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17 April 2024

Water disinfection by orifice-induced hydrodynamic cavitation[*]

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Abstract

Hydrodynamic Cavitation (HC) is considered as a promising water-disinfection technique. Due to the enormous complexity of the physical and chemical processes at play, research on HC reactors is usually carried out following an empirical approach. Surprisingly, past experimental studies have never been designed on dimensional-analysis principles, which makes it difficult to identify the key processes controlling the problem, isolate their effects and scale up the results from laboratory to full-scale scenarios.

The present paper overcomes this issue and applies the principles of dimensional analysis to identify the major non-dimensional parameters controlling disinfection efficacy in classical HC reactors, namely orifice plates. On the basis of this this analysis, it presents results from a new set of experiments, which were designed to isolate mainly the effects of the so-called cavitation number (σ_v). Experimental data confirm that the disinfection efficacy of orifice plates increases with decreasing σ_v. Finally, in order to discuss the significance of the results presented herein and frame the scope of future research, the present paper provides an overview of the drawbacks associated with dimensional analysis within the context of HC.

Keywords: hydrodynamic cavitation; water disinfection; E. Coli; dimensional

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analysis; orifice-plate reactor; review.

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1. Introduction

The lack of safe water in developing countries is affecting millions of people causing major sanitation and economic issues. Prohibitive costs and difficult access to chemicals (as well as qualified staff) [1, 2] prevent the implementation of water-disinfection technologies routinely adopted in developed countries. Such technologies also present shortcomings, the main one being associated with the fact that all chemicals used in the disinfection process may produce, under certain conditions, unhealthy and carcinogenic by-products (DBPs), such as trihalomethanes, haloacetic acids, bromate, and chlorite [3]. As a result, in the ongoing review of drinking water quality guidelines, the World Health Organization is updating risk assessments for dissolved chemicals, setting new stricter limits for DBPs [4]. From this picture it appears that there is a clear need to implement chemical-free water disinfection techniques, which must also be simple to use, robust and low-cost, especially to meet the demands of low-income countries.

In this context, techniques based on cavitation seem to be promising. Cavitation exploits the phenomenon of formation, growth, and collapse of vapour/gas bubbles triggered by pressure variations [5]. When the fluid experiences a critical pressure (i.e., lower than vapour pressure), the formation of cavities begins, and the maximum size of the cavities is typically reached under isothermal expansion. Subsequently, when higher pressure is recovered, bubbles undergo adiabatic collapse. Such a collapse leads to the formation of pressure-waves and micro-jets that instantly release a large amount of energy while generating intense normal and shear fluid stresses [6, 7, 8]. In the scientific literature, these severe conditions are considered as the main cause of cell membrane damage and consequently of microorganism death or inactivation [9, 10, 11]. Moreover, high temperature peaks promotes chemical reactions, such as the dissociation of water molecules into $^{\bullet}$OH radicals, which provide oxidizing power and increase the efficiency of disinfection [12].

Cavitation can be generated in two main ways: by ultrasonic waves travelling through the liquid (i.e., acoustic cavitation, AC), or by forcing the fluid through a constriction (i.e., hydrodynamic cavitation, HC) [13]. AC is energy demanding, works on batch and is effective only for fluid volumes in close proximity to the acoustic source. Thus, AC is deemed unsuitable for the treatment of large volumes of water [14, 15]. In the case of HC (which has been investigated considerably less than AC [16]), cavitation is tipically obtained by a pressure drop, e.g. generated by an orifice plate or a Venturi tube. In contrast to AC, HC is deemed as an energetically more efficient process [17, 18] and allows for the treatment of large volumes of moving water; so it is suitable for implementation in drinking- and waste-water treatment plants [19] as well as in the food and beverage processing [20, 21, 22] and chemical synthesis [23, 24, 25, 26].

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HC is induced by purely mechanical devices which can be used without the presence of qualified staff and is therefore suitable for use in developing countries. On the down side, HC is a more complex process than AC from the fluid-dynamics prospective. AC involves bubbles growing and collapsing in quiescent water, whereas HC commonly occurs in fast moving fluids whose dynamics responds to complex (and currently poorly-understood) non-linear interactions between bubbles and turbulence. As a consequence, the study of fluid dynamics within HC reactors for water treatment is still in its infancy and much more work is needed to identify governing parameters and quantifying their role in the game of disinfection.

Recently, many research-works have focused on demonstrating the effectiveness of HC as well as exploring the effects of different HC-reactor-geometries on disinfection efficiencies. Orifice plates [27, 14], Venturi tubes [28, 29], and rotor-stator reactors (e.g., high speed homogenizers) [30, 31] were the most investigated devices.. Other studies have focused on hybrid disinfection techniques (i.e., the combination of cavitation with chemical disinfectants) in order to reduce the amount of chemicals in the water treatment processes [32, 33, 34, 35]. This interest in the topic witnesses the great potential of HC for water disinfection [36]. However, the scientific literature on HC currently lacks of a sound methodological approach as well as sound theoretical grounds [37, 29]. In particular, due to its complexity, the study of HC for disinfection purposes has been commonly addressed using an empirical approach, although numerical studies have also been proposed (see, e.g., [38, 39, 40, 41]). However, to the best of our knowledge, none of the existing studies in the literature has based the experimental work on dimensional analysis. This clearly makes it difficult to: (i) identify all the relevant non-dimensional groups controlling the problem; (ii) isolate their effects on the observed disinfection efficiencies; and ultimately (iii) scale up from laboratory to full-scale HC reactors.

The objectives of the present paper are: (a) to identify, by means of dimensional analysis, the non-dimensional parameters controlling disinfection efficiencies in classical HC reactors such as orifice plates; (b) in light of this dimensional analysis, to provide a critical appraisal of the relevant literature (section 3) highlighting main results and knowledge gaps; (c) to present results from a systematic set of experiments where the effects of the so-called cavitation number (defined in the next section), were isolated and assessed. This parameter was chosen as the target of the present paper as it quantifies the intensity of cavitation and is therefore considered key for the design of HC reactors.

2. Dimensional analysis

When a problem is as complex as HC, it is convenient to first attempt to tackle it by adopting an empirical approach whose very first step should be dimensional analysis. Towards this end, let us consider the simple case of a HC reactor where cavitation is induced by orifice-plates only. This is convenient because: (i) the geometry of Venturi-tubes (i.e., the other commonly-employed HC reactor) is much more complex than orifices as it is associated with many

more influencing variables, which make the analysis significantly more convoluted; (ii) as Venturi tubes, orifice plates have been largely investigated in the literature and therefore the results of the present paper can be easily put into context; (iii) we present novel experiments involving orifice plates only.

Since most experimental studies deal with the case of HC reactors implemented in closed loop systems, we consider the case of a fixed volume of water V which goes through a HC reactor multiple times n_p. At these conditions it can be argued that the bacterial concentration C of a specific pathogen (measured in Colony Forming Units, CFU, per unit volume of water) depends on the following set of parameters:

$$C = f(C_0, \mu, \rho, \gamma_s, v_h, P_2 - P_v, n_p, L_i), \tag{1}$$

where C_0, is the initial pathogen concentration; μ, ρ and γ_s are the kinematic viscosity, the density and the surface tension of water, respectively; v_h is the mean fluid velocity at the downstream end of the constriction, P_2 is the absolute pressure recovered downstream of the orifice plate (see Figure 1), P_v is the absolute water-vapor pressure, L_i, in general terms, defines the set of variables characterizing the geometry of the reactor. In the simplest case of a circular orifice plate, which is the subject of the present paper, L_i includes: the characteristic diameter of the orifice (i.e,. the constriction) d, the diameter of the pipe upstream and downstream of the plate D, the orifice-plate thickness b and the number of orifices n.

As far as equation (1) is concerned, a few comments are in order: (i) as in many other Fluid Dynamics problems, Equation (1) does not include simple pressures but pressure-differences with respect to a reference value, which, due to the importance of bubble formation and collapse, is here identified as the water-vapor pressure; (ii) the effects of temperature are indirectly taken into account through parameters μ, ρ, γ_s and P_v; (iii) we did not consider the absolute water pressure upstream of the orifice plate (P_1) as this is a direct function of v_h and P_2 and is therefore redundant.

Relevant non-dimensional parameters can now be identified by application of the well-known Buckingham π theorem [42]. Towards this end ρ, v_h and d are chosen as the three repeating variables, which contain all the primary dimensions appearing in Equation (1), namely length [L], mass [M] and time [T] (CFU appearing in the definition of concentrations are dimensionless numbers and therefore cannot be accounted for as a primary dimension). Simple dimensional arguments lead to the following set of non-dimensional parameters:

$$Cd^3 = f_1\left(C_0 d^3, \frac{\rho v_h d}{\mu}, \frac{\rho v_h^2 d}{\gamma_s}, \frac{P_2 - P_v}{\rho v_h^2}, n_p, \overbrace{\frac{D}{d}, \frac{b}{d}, n}^{L^*}\right). \tag{2}$$

The dependent parameter on the left hand of Equation (2), can be combined with the first independent parameter to form a dimensionless bacterial

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concentration $\frac{C}{C_0}$, so that Equation (2) becomes:

$$\frac{C}{C_0} = f_2\left(C_0 d^3, \frac{\rho v_h d}{\mu}, \frac{\rho v_h^2 d}{\gamma_s}, \frac{P_2 - P_v}{\rho v_h^2}, n_p, \frac{D}{d}, \frac{b}{d}, n\right), \qquad (3)$$

where, C/C_0 is herein defined as a non-dimensional disinfection efficiency; $(\rho v_h d)/\mu$ is the Reynolds number of the jet forming at the downstream end of the orifice, which regulates turbulence and flow development within the HC reactors; $\rho v_h^2 d/\gamma_s$ is the so-called Weber number, which takes into account surface tension forces with respect to inertial forces and, presumably, strongly influences the behaviour of bubbles [43]; D/d and b/d are geometrical parameters that, together with the Reynolds number affect the flow characteristics of the orifice and hence the fluid stresses bacteria may be subjected to (bacteria are strongly sensitive to turbulence and fluid stresses, see e.g. [44]); $(P_2 - P_v)/(\rho v_h^2)$ is the so-called cavitation number, which quantifies the intensity of cavitation so that, for values above the one corresponding to the onset of supercavitation, the lower is its value the more intense is the formation and collapse of bubbles. It is worth mentioning that, in the current literature, the cavitation number σ_v is usually formulated adding a scaling factor 2, irrelevant for dimensional analysis, see Equation (4); $C_0 d^3$ is a dimensionless initial concentration, which, although arbitrarily defined, indicates that the effectiveness of a HC reactor might depend on initial conditions. In Equations (1), (2) and (3), f, f_1 and f_2 are functional relations between dependent and independent variables.

The next section provides an appraisal of the existing literature contextually to the dimensional analysis carried out above.

3. A critical appraisal of the literature

As hydrodynamic cavitation has attracted considerable research interest, the number of experiments available in the scientific literature is large and growing fast. In Table 1 we selected 12 works on the basis of the following criteria: (i) they all deal with HC induced by orifice plates or similar reactors such as nozzles or partially closed valves; (ii) they all provide sufficient experimental details; (iii) they all deal with disinfection of bacteria, except the work of Badve et al. [45] that used zooplankton, included for the sake of completeness. It is worth noting that *Escherichia Coli* is the most commonly adopted bacterium in these experiments as it is often present in naturally-contaminated water. Moreover, the microbiological quality of drinking water relies largely on examination of indicator bacteria such as coliforms, in particular *E. Coli*. For this reason, the procedures to measure its concentration is internationally regulated. In addition, *E. Coli* is simply cultivable in laboratory and is not particularly dangerous to handle during the experiments. For the sake of completeness and to provide an overall overview of the relevant literature, Table 1 provides information and parameters that were reported by the authors of each referenced paper and not only those already mentioned in the previous section. In order to interpret Table 1 the following definitions apply:

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- **reactor type** indicates the type of cavitating reactor used. OP refers to orifice plates; DynaJets®, DynaSwirl® and StratoJets® are patented reactors with a configuration comparable to an orifice plate; "valve" refers to as partially closed valve in which cavitation occurs; "pump" refers to experiments where the bacterial reduction solely due to the action of the pump was assessed;

- **configuration** is the geometry of the orifice plate used, e.g. 25×2 mm indicates a plate with 25 holes of 2 mm of diameter. Additional information indicate the shape of the holes: squared (S), rectangular (R), if not specified otherwise, circular holes were adopted. Orifice plates put in series are indicated with the "+" sign;

- **holes area** is the total area of the holes in the plate;

- α is the ratio of perimeter of the holes to their total area;

- β is the ratio of holes-area to cross-sectional area of pipe;

- **cavitation number**. This is considered one of the most important parameters to describe the intensity of cavitation. The literature, rather arbitrarily, introduced two types of cavitation numbers:

$$\sigma_v = \frac{P_2 - P_v}{1/2\,\rho\,v_h^2},\tag{4}$$

and,

$$\sigma_{v,\Delta P} = \frac{P_2 - P_v}{P_1 - P_2},\tag{5}$$

- **t** is the total duration of the treatment;

- **initial/final CFU** are the initial and final concentration of bacteria used in the disinfection experiments;

- **disinfection efficiency** is the bacteria concentration reduction δC on percentage or in logarithmic unit, e.g. $3\,log$ corresponds to a reduction in the bacterial concentration of three orders of magnitude.

Empty cells (-) in Table 1 indicate data not provided by the authors. Data with an asterisk were non directly provided by the referenced papers, but were derived by the authors of the present paper. Appendix A provides details about experimental methods and results provided by papers referenced in Table 1.

Table 1 witnesses the remarkable experimental efforts made by researchers to investigate the influence of the main variables involved in orifice-shaped reactors, e.g. the pressure drop, the velocity of the constricted flow, etc.. However, the dimensional analysis developed in the previous section highlights that the single dimensional variables are not the key information, but it is instead their suitable combination in dimensionless groups that is informative. The values of
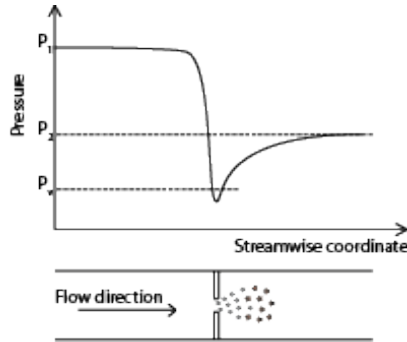
Figure 1: Upper panel reports the qualitative behaviours of the pressure along the centerline. Lower panel shows the formation and successive implosion of cavities.

those numbers therefore play the crucial role in determining reactor behavior and its effectiveness in inactivating bacteria. Aware of this fact, in Table 2 we report the dimensionless numbers used in the works reported in Table 1. In many of these studies, the experimental data necessary to calculate the dimensionless parameters were often not explicitly provided. Therefore, in Table 2, a qualitative comparison is made by simply reporting which non-dimensional parameters, among those of Equation 3, were left to vary ("×" symbol) and those that were kept constant ("✓" symbol) in a specific set of trials. Therefore, this table allows to asses whether the effects of one (or some) non-dimensional parameters were actually isolated.

Table 2 shows that past studies and experiments were designed to investigate/isolate the effects of dimensional, rather than non-dimensional parameters on disinfection efficiencies. The only non-dimensional group, whose effects were isolated (by three studies only [46, 12, 47]) is the one related to the initial concentration, which seems to be negatively correlated with the disinfection efficiency of orifice-based HC reactors. Therefore, while the available literature plays a very important role in identifying and quantifying the effectiveness of HC and different HC reactors, it does not allow to understand and explore the physical mechanisms underpinning the disinfection efficiencies observed in the experiments as these could be the effect of multiple variables and associated physical processes. The authors believe that, in order to progress in this research field, future experimental work should be designed and carried out using the dimensional analysis framework herein proposed or, if required, different versions of it.

Consistently with this idea, the remaining part of the paper is dedicated to the the presentation of a set of experiments that the authors have carried out in an orifice plate HC reactor to investigate mainly the effects of one of the aforementioned dimensionless parameter, namely, the cavitation number σ_v. This parameter is widely used to quantify the intensity of cavitation and is therefore commonly considered extremely important to characterize disinfection efficiencies. In fact, since bubbles implosion is often considered the key physical process

responsible for bacterial inactivation (although this hypothesis has recently been challenged, see [37, 29]), it is expected that disinfection efficiencies will be higher for lower σ_v. Experiments were also designed to further investigate the effects of initial bacterial concentration C_0 on disinfection efficiencies.

4. Experimental methods

All the experiments were carried out in the Water Engineering Laboratory "Giorgio Bidone" at the Polytechnic of Turin (Italy) while bacteria preparation and sample analysis was performed at the Research Centre of SMAT, which is the Water Utility serving the city of Turin. The pilot plant used to induce cavitation is shown in the upper panel of Figure 2 and it consists in a closed loop pipe (stainless steel, 32 mm internal diameter) including a cylindrical holding thank of 35 l volume (300 x 500 mm). The water temperature was controlled by two chiller-units connected to a cooling coil placed inside the thank. A centrifugal multistage pump (Lowara 3SV-11, 2900 rpm, 1 kW) was used to recirculate the water and an electromagnetic flow meter (Endress Hauser PROline Promag 10) was employed to monitor the flowrate. Two manometers, named M1 and M2 (lower-left panel of Figure 2) were used to monitor P_1 and P_2, respectively. A ball-valve was used to control P_2 and a transparent control section made of glass (lower-right panel of Figure 2) was used to observe the occurrence of cavitation. The cavitation unit was mounted between two flanges and was made of a stainless steel-plate of 16 mm thickness (lower-left panel of Figure 2), where 4 holes of 2.5 mm diameter were drilled and arranged in a diamond pattern. Each test consisted in the treatment of 21 l of Milli-Q® water contaminated by *E. Coli* bacteria at different concentrations.

A reference sample was taken at the beginning of each test, after contaminated water was mixed within the whole hydraulic circuit for 10 minutes at very low flow-rates that induced no cavitation. Successive samples were taken at different times during each test. Each sample (300 ml), was then stored in sterile plastic bottles that were kept at a constant temperature of 4 °C for a period of maximum 24 hours. The samples were then brought to SMAT labs for microbiological analysis to reconstruct the variation of the bacterial concentration C with time during each experiment. After each experiment, the entire hydraulic circuit was sterilized by injecting 2 ml of sodium hypochlorite and then rinsed three times. At the end of the procedure a sample was taken to verify the absence of either chlorine- or bacteria-residuals to make sure that following experiments were carried out at identical "circuit" conditions.

E. Coli was chosen as the reference bacterium for this study since it allows a comparison with the works presented so far in the literature. *E. Coli* (ATCC 8739, IELAB) was propagated on Chromogenic Coliform Agar (Oxoid) overnight at 37 °C. Colonies were resuspended in Maximum Recovery Diluent (Oxoid) and live bacteria concentration was measured through absolute ATP quantification by Dendridiag SW reagents (GLBiocontrol) following the manufacturer's instructions. The desired amount of bacteria was then transferred into 1 l of Milli-Q® water and further diluted to a final volume of 21 liters of

Table 1: Experimental studies about water disinfection by orifice plate reactors or similar devices. Data marked with * were not directly provided by the referenced papers, but derived by the authors of the present paper. (see text for the meaning of each column)

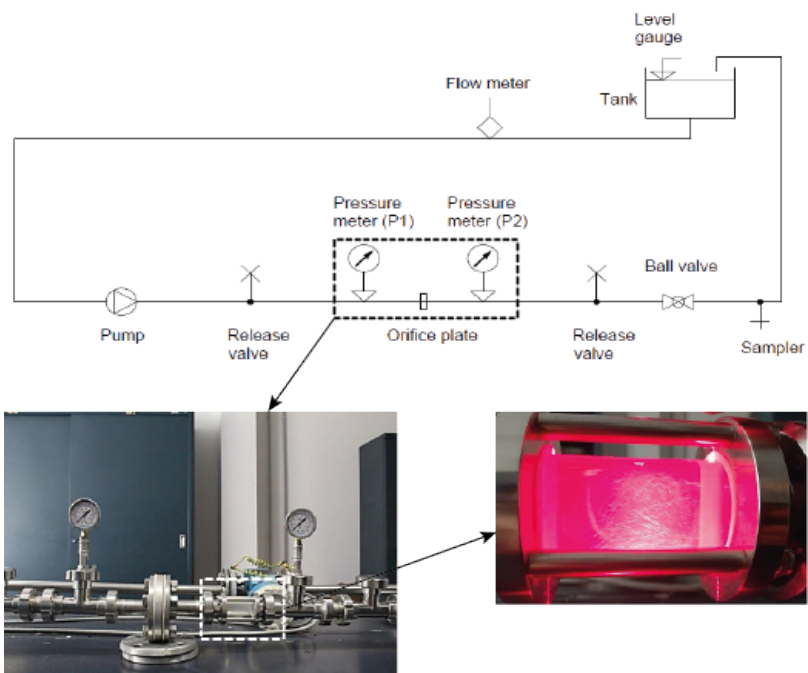| Authors (year)	Reactor type	Bacteria	Configuration	Holes area [m^2]	α [mm^{-1}]	β [$-$]	Q [l/s]	v_h [m/s]	P_1 [bar]	P_2 [bar]	σ_v	$\sigma_{v,\Delta P}$	V [l]	t [min]	Initial CFU [CFU/ml]	Final CFU [CFU/ml]	Disinfection efficiency
Jyoti et al. (2001) [48]	Valve	Coliform	Valve	-	-	-	-	-	1.72	0*	-	0.57*	75	15	4580	3180 ± 10	30% ± 0.22%
				-	-	-	-	-	3.44	0*	-	0.28*	75	15	4280	3780 ± 20	11% ± 0.22%
				-	-	-	-	-	5.17	0*	-	0.19*	75	15	3940	3020 ± 10	23% ± 0.22%
				-	-	-	-	-	5.17	0*	-	0.19*	75	60	-	-	44%
Kalumuck et al. (2003) [46]	DynaJets®	E. Coli	DynaJets®	-	-	-	-	4.13 - 5.17	4.13 - 5.17	-	-	0.24* - 0.19*	1.5	40	10^8 - 10^9	10^5 - 10^6	3 log
				-	-	-	-	-	10.3	0	-	0.09*	1.5	120	10^8 - 10^9	10^4 - 10^5	5 log
				-	-	-	-	-	**10.3**	0	-	0.09*	1.5	30	10^7	10^2	5 log
Balasundaran et al. (2006) [49]	OP	E. Coli	25x2 mm	7.85E-05	2.00	0.14	2.99	47	15	0	0.13	-	20	-	-	-	-
			1x12 mm	1.13E-04	0.33	0.20	3.86	34	13.8	0	0.17	-	20	-	-	-	-
			1x15 mm	1.77E-04	0.27	0.32	5.28	30	10	0	0.22	-	20	-	-	-	-
			1x17 mm	2.27E-04	0.24	0.41	5.57	25	4	0	0.32	-	20	-	-	-	-
			1x19 mm	2.84E-04	0.21	0.51	5.61	20	31	0	0.49	-	20	-	-	-	-
			1x22 mm	3.80E-04	0.18	0.68	5.51	15	0	0	0.92	-	20	-	-	-	-
Balasundaran et al. (2011) [27]	OP	E. Coli	40x2 mm	1.26E-04	2.00	-	4.58	36.4	-	0	0.14	-	20	-	-	-	-
			32x(2x2) mm (S)	1.28E-04	2.00	-	4.16	32.5	-	0	0.19	-	20	-	-	-	-
			16x(2x4) mm (R)	1.28E-04	1.50	-	4.24	33.1	-	0	0.18	-	20	-	-	-	-
			25x2 mm	7.85E-05	2.00	-	2.68	34.1	-	0	0.17	-	20	-	-	-	-
			5x5 mm	9.82E-05	0.80	-	3.26	33.2	-	0	0.18	-	20	-	-	-	-
			1x14 mm	1.54E-04	0.30	-	5.07	32.9	-	0	0.18	-	20	-	-	-	-
Azuma et al. (2007) [50]	OP	E. Coli	1x0.10+1x0.16	-	-	-	-	355.7	900	-	0.154	-	-	-	1.4×10^{11}	-	100% in 3 passes
		B. Subtilis	1x0.10+1x0.23	-	-	-	-	353.7	800	-	0.037	-	-	-	-	-	100% in 5 passes
		B. Halodurnas	1x0.10+1x0.23	-	-	-	-	353.7	800	-	0.037	-	-	-	-	-	100% in 6 passes
		P. Putida	1x0.10+1x0.23	-	-	-	-	353.7	800	-	0.037	-	-	-	-	-	100% in 4 passes
		E. Coli	1x0.10+1x0.23	-	-	-	-	353.7	800	-	0.037	-	-	-	-	-	100% in 3 passes
Sawant et al. (2008) [45]	OP	Zooplankton	25% open	7.85E-05	0.40	-	0.80	10	3.7	-	5.13	-	50	120	10^7	-	79% in 1 pass
	OP		50% open	1.56E-04	0.29	-	1.70	11	3.2	-	3.94	-	50	120	10^7	-	75% in 1 pass
	OP		75% open	2.35E-04	0.12	-	1.30	6	3.1	-	14.68	-	50	120	10^7	-	82% in 1 pass
	Valve		20% open	5.73E-06	-	-	1.00	16	3.4	-	1.93	-	50	120	10^2	-	57% in 1 pass
	Valve		40% open	2.73E-05	-	-	1.90	15	3.2	-	2.02	-	50	120	10^3	-	33% in 1 pass
	Pump		-	-	-	-	2.80	-	2.9	-	-	-	50	120	10^4	-	28% in 1 pass
Arrojo et al. (2008) [12]	OP	E. Coli	1x5 mm	1.96E-05	0.80	-	1.11	57	-	-	-	-	50	120	10^7	-	-
			6x2 mm	1.88E-05	2.00	-	1.11	59	-	1	0.12*	-	50	120	-	-	-
			25x1 mm	1.96E-05	4	-	1.11	57	-	1.5	0.16*	-	50	120	-	-	-
			25x1 mm	1.96E-05	4	-	1.11	57	-	2	0.19*	-	50	120	-	-	100%
			25x1 mm	1.96E-05	4	-	1.11	57	-	2	0.19*	-	50	120	-	-	85%
			25x1 mm	1.96E-05	4	-	1.11	57	-	2	0.19*	-	50	120	-	-	70%
Loraine et al. (2012) [47]		E. Coli	1x3.23 mm	8.19E-06	2.00	0.40	-	-	2.1	-	-	0.47*	2	60	10^6	10^1	5 log
			8x1.14 mm	8.17E-05	2.00	0.40	-	-	16.5	-	-	0.06*	1.8	120	10^6	10^1	5 log
	DynaSwirl®		72x0.38 mm	8.17E-06	2.00	0.40	-	-	5.2	-	-	0.19*	2	240	10^6	10	6 log
			1x4.5 mm	1.59E-05	4.00	0.02	-	-	3.45	-	0.33	0.28*	-	270	10^7	-	4 log
			1x4.5 mm	1.59E-05	2.00	0.40	-	-	2.1	-	0.5	0.47*	2	60	10^7	-	7 log
	DynaSwirl®	Klebsiella	1x3.23 mm	8.04E-06	1.33	0.09	-	-	1	-	1	0.98*	-	120	10^7	-	4 log
			-	8.19E-06	-	-	-	-	2.1	-	0.5	**0.47**	2	210	10^3 - 10^5 - 10^7 - 10^9	10^2	7 log
	StratoJet®	P. Aeruginosa	8x1.14 mm	8.17E-06	-	-	-	-	16.5	-	-	0.06*	2	60	10^7	10^2	5 log
	StratoJet®	P. Syringae	8x1.14 mm	8.17E-06	-	-	-	-	16.5	-	-	0.06*	1.8	90	$\sim 10^8$	$\sim 10^5$	3 log
			-	-	-	-	-	-	16.5	-	-	-	1.8	20	$\sim 10^7$	$\sim 10^1$	6 log
Wang et al. (2015) [35]	OP	?	33x2 mm	1.04E-04	2.00	0.40	-	-	3.5	-	-	0.28*	25	60	10^3	10^3	57.30%
			33x2 mm	1.04E-04	2.00	0.40	-	-	4.0	-	-	0.24*	25	60	10^3	10^3	60%
			33x2 mm	1.04E-04	2.00	0.40	-	-	4.5	-	-	0.22*	25	60	10^3	10^3	67.30%
			33x1 mm	2.59E-05	4.00	0.02	-	-	4.5	-	-	0.22*	25	60	10^3	10^3	67%
			33x2 mm	1.04E-04	2.00	0.40	-	-	4.5	-	-	0.22*	25	60	10^3	10^3	63%
			20x2 mm	6.28E-05	2.00	0.05	-	-	4.5	-	-	0.22*	25	60	10^3	10^3	58%
			17x3 mm	1.20E-04	1.33	0.09	-	-	4.5	-	-	0.22*	25	60	10^3	10^3	40%
Badve et al. (2015) [51]	OP	E. Coli	1x2 mm	3.14E-06	2.71	-	-	-	3	-	0.62	-	4	-	10^7	-	15%
Filho et al. (2015) [52]	Nozzle	E. Coli	1x2 mm	3.14E-06	2.71	-	0.48	87.8	80	-	-	-	40	30	10^5	10^3	98.30%
			1x2 mm	3.14E-06	2.71	-	0.48	109.8	100	-	-	-	40	30	10^5	10^2	99.96%
			1x2 mm	3.14E-06	2.71	-	0.48	131.8	120	-	-	-	40	30	10^5	10^1	100%
Liu et al. (2016) [53]	OP	E. Coli	49x1 mm	3.85E-05	-	0.048	0.56	-	2.5	0	0.92*	0.40*	20	120	1.60×10^3	-	99%

Figure 2: Experimental set-up: upper image shows the schematic representation of the experimental set-up, lower left image shows the orifice plate and pressure measurements points, lower right image shows the transparent test section illuminated by red laser light during disinfection experiments.

Table 2: Dimensional analysis of the works presented in Table 1. ✓: parameters kept constant in all the tests. ×: parameters varied between the tests.

Authors (year)	$C_0 d^3$	$\frac{\rho v_h d}{\mu}$	$\frac{\rho v_h^2 d}{\gamma_s}$	$\frac{D}{d}$	$\frac{b}{d}$	$\frac{P_2 - P_v}{\rho v_h^2}$	n	n_p
Jyoti et al. (2001) [48]	×	×	×	×	×	×	×	×
Kalumuck et al. (2003) (a) [46]	×	×	×	✓	✓	×	×	✓
Kalumuck et al. (2003) (b) [46]	×	✓	✓	✓	✓	✓	✓	✓
Balasundaran et al. (2006) [49]	×	×	×	×	×	×	×	×
Balasundaran et al. (2011) [27]	×	×	×	×	×	×	×	×
Azuma et al. (2007) [50]	×	×	×	×	×	×	×	×
Sawant et al. (2008) [45]	×	×	×	×	×	×	×	×
Arrojo et al. (2008) (c) [12]	×	×	×	×	×	×	×	✓
Arrojo et al. (2008) (d) [12]	×	✓	✓	✓	✓	✓	✓	✓
Loraine et al. (2012) (e) [47]	×	×	×	✓	✓	×	×	✓
Loraine et al. (2012) (f) [47]	×	✓	✓	✓	✓	✓	✓	✓
Wang et al. (2015) (g) [35]	×	×	×	✓	✓	×	×	×
Wang et al. (2015) (h) [35]	×	×	×	×	×	×	×	✓
Badve et al. (2015) [51]	✓	✓	✓	✓	✓	✓	✓	✓
Filho et al (2015) [52]	×	×	×	✓	✓	×	×	×
Liu et al. (2016) [53]	✓	✓	✓	✓	✓	✓	✓	✓
Our results	✓	✓	✓	✓	✓	×	✓	✓
(numerical value)	✓	154 900	65 900	12.8	6.4	×	4	410

Milli-Q® water while filling the tank at the inlet of the circuit to reach the desired concentration. The starting bacteria concentration of each experiment was confirmed by Colilert Quanti-Tray 2000 assay (IDEXX). *E. Coli* concentration at the different time points was determined by Colilert Quanti-Tray 2000 assay (IDEXX) according to standard procedures [54].

Three groups of experiments were performed to analyze the effect of different cavitation numbers σ_v on the disinfection efficiency. As expressed in Equation (4), assuming constant temperature conditions (and hence constant values of fluid properties such as P_v, γ_s, ρ and μ), the variables involved in the computation of σ_v are the recovery pressure P_2 and the orifice fluid velocity v_h. The former was directly measured, whereas the latter was estimated simply

Table 3: Hydraulic and geometric characteristics of the orifice plate reactor.

σ_v [-]	Configuration	Holes area [m^2]	Q [l/s]	v_h [m/s]	P_1 [bar]	P_2 [bar]	V [l]	t [min]
0.20	4x2.5 mm	1.96E-05	0.6	30.5	7.5	0	21	30 - 360
0.40	4x2.5 mm	1.96E-05	0.6	30.5	7.5	1	21	30 - 120
0.65	4x2.5 mm	1.96E-05	0.6	30.5	7.5	2	21	30 - 240

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as the ratio between the flow rate and the holes area (see also the discussion section for more details on the definition of v_h and its shortcomings).

The downstream recovery pressure (or back-pressure) P_2 was varied by means of the ball-valve (see Figure 2) in order to vary σ_v. As shown in Table 3, the other parameters (orifice velocity and flow rate) were kept constant and so were all the non-dimensional parameters identified in Equation (3).

In the first group of experiments the configuration characterized by $\sigma_v = 0.20$ was studied. Seven tests with initial concentration C_0 between 10^2 $CFU/100$ ml and 10^5 $CFU/100$ ml were carried out. The duration of the experiments varied between 120 and 360 minutes, which correspond to a number of passages $n_p \sim$ 205 and 620, respectively. Samples were taken every 30 minutes.

The second group of experiments was performed at $\sigma_v = 0.40$. Six experiments with initial concentration between 10^2 $CFU/100$ ml and 10^4 $CFU/100$ ml were performed. The total duration of the tests was 120 minutes ($n_p \sim 205$) and samples were taken every 30 minutes.

In the last group of experiments, the configuration with $\sigma_v = 0.65$ was studied. Three tests of 240 minutes ($n_p \sim 410$) with initial concentrations between 10^3 $CFU/100$ ml and 10^6 $CFU/100$ ml were performed. Samples were taken at 60, 120, 180 and 240 minutes.

Two control experiments were performed by removing the orifice plate to investigate the effects of the pump on disinfection efficiencies. In those scenarios the flow rate was higher due to the absence of the orifice plate. The initial concentration was 10^2 $CFU/100$ ml and the tests lasted for 120 minutes, corresponding to $\sim$ 360 passes (the number of passes in this case is higher due to the higher flow rate). Samples were taken every 30 minutes. The bacterial concentration remained constant for the entire duration of the experiment.

During all the orifice-plate experiments, and the control experiments without the orifice-plate reactor, the water-temperature was controlled by means of two chiller units. It is finally pointed out that for all hydrodynamic configurations, the ball-valve was always working in a non cavitating regime and, therefore, it never played any role in the game of disinfection.

5. Results

Figure 3 shows C/C_0 vs n_p curves for each individual trial. In order to avoid overcrowding of the figure, the 95% confidence intervals (as estimated from the Quanti-Tray/2000 method [54]) associated with each experimental data-point, are reported in Table 5 in Appendix B. Figure 3 indicates that the orifice plate employed in the experiments caused a reduction in bacterial concentration in all the experimental configurations investigated. Confidence intervals associated with each measurement (see Table 5) are quite large and make it difficult to identify statistically-significant trends. However, it seems that, contrary to what reported in the previous literature [12, 46], the initial concentration value C_0 of bacteria (or its dimensionless counterpart $C_0 d^3$) have no clear effect on the non-dimensional disinfection efficiencies at all the cavitation numbers investigated. Moreover, contrary to what reported in the literature [12, 47], the

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C/C_0 vs n_p curves do not show any obvious initial plateau (or quasi-stationary phase), which is commonly interpreted as a colony fragmentation, rather than an effective disinfection phase. However, it should be noted that the concentrations of bacteria used herein (much lower than those used by [46, 47]) are unlikely to generate colonies and therefore this could be the reason underpinning the observed results.

Since no clear effects of the initial concentration were observed, average C/C_0 vs n_p curves were computed from each group of experiments corresponding to each cavitation number (i.e., each curve is the average of the curves shown in panels 3a - 3c) and are reported in Figure 4a. In this Figure the shaded error bars represent the standard deviations of concentration obtained from each experiment group. As previously predicted, Figure 4a shows that the average C/C_0 vs n_p curves drop faster for lower values of the cavitation numbers σ_v. This is in agreement with the idea that a more intense cavitation (i.e., a lower σ_v) promotes a more efficient disinfection.

The series of mean disinfection values were then fitted by the exponential law $C/C_0 = \exp(-r \cdot n_p)$ as shown in Figure 4b, in order to obtain the bacterial reduction rate, r, typical of each cavitation number. Aiming to a fair comparison, the same number of sampling values were considered for all cavitation numbers. The rates obtained are reported in Table 4 and confirm that at lower cavitation numbers correspond higher disinfection rates. The R-square values shown in Table 4 witnesses goodness of data fitting.

Table 4: Bacterial reduction rates r and coefficients of determination R^2 corresponding to the exponential fitting of the average disinfection curves shown in Figure 4b
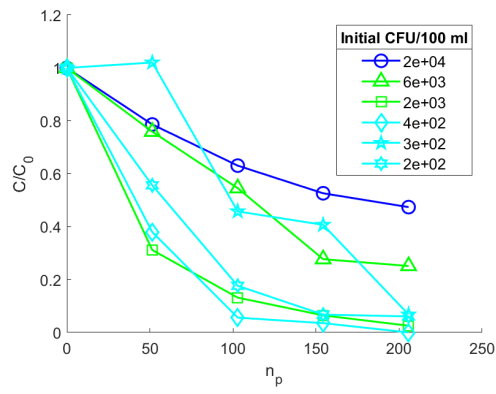
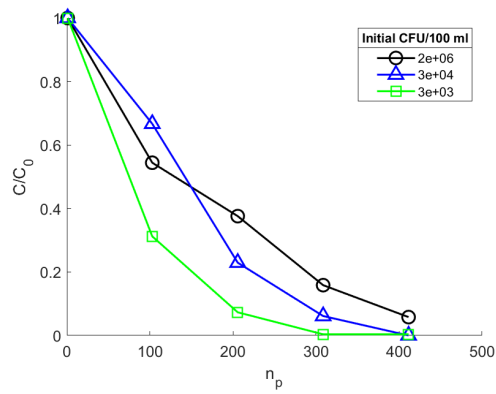| Cavitation number	r($\cdot 10^3$)	R^2
$\sigma_v = 0.2$	10.5	0.980
$\sigma_v = 0.4$	9.56	0.993
$\sigma_v = 0.6$	7.10	0.997

6. Discussion

It is now important to point out that dimensional analysis represents a valid starting point for the design of experiments and for the development of empirical formulae, but it is certainly not free from drawbacks, which are now discussed to clarify the significance of the results presented herein and frame the scope of future research-works. A key problem of dimensional analysis is associated with the fact that it is not always straightforward to rigorously take into consideration all the factors influencing a problem, often because it is difficult to associate such factors with well-defined and measurable variables. For example, in the case of orifice-plates, the onset of cavitation (i.e., the critical number of σ_v below which cavitation occurs), can be very sensitive to fine experimental-conditions. This means that if no-control on these details is possible, the cavitation number may
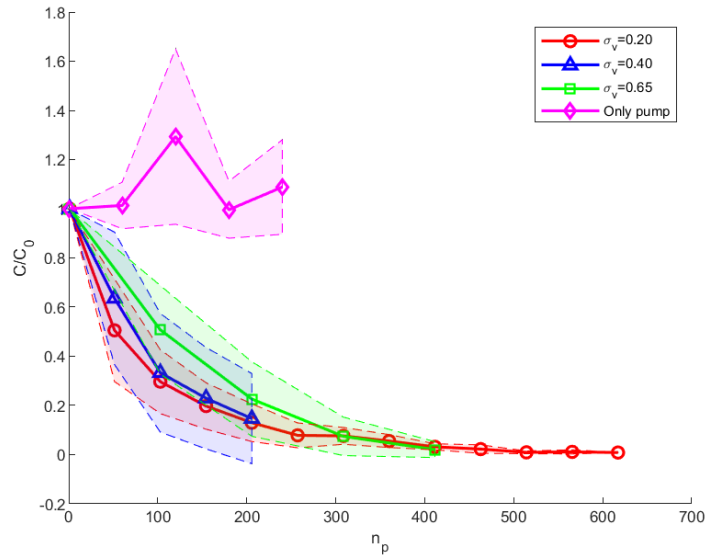
13
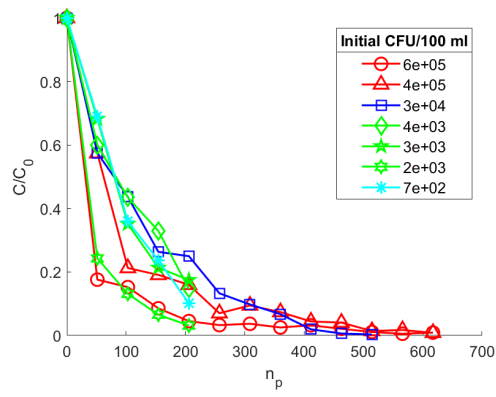
(a) $\sigma = 0.20$
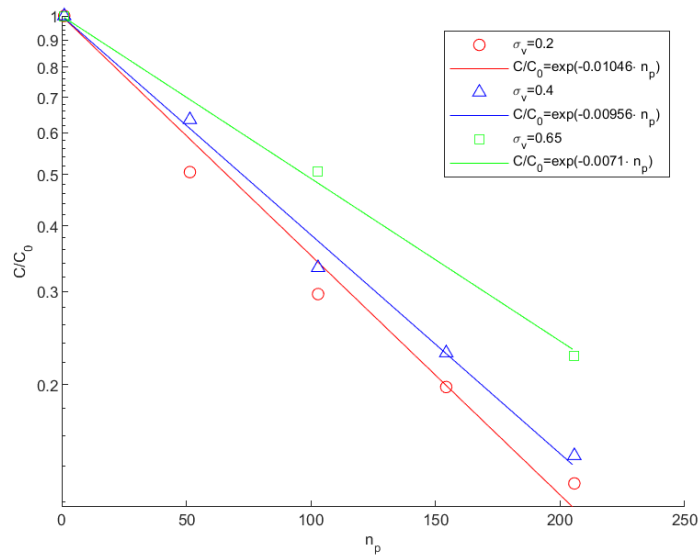
(b) $\sigma = 0.40$

(c) $\sigma = 0.65$

Figure 3: Disinfection efficiency of the orifice-plate reactor at different cavitation numbers. Each color represents a different order of magnitude of *E. Coli* initial concentration (C_0).

(a)

(b)

Figure 4: Average behavior of the disinfection curves at different cavitation number. In the panel (a), the shaded regions correspond to the standard deviation. In the panel (b), the exponential fitting are shown.

not represent an objective parameter to quantify consistently the intensity of cavitation among different experiments. In particular, the onset of cavitation may depend on fine geometrical details of the orifice (e.g. small manufacturing defects such as irregular edges of the inlet or artificial roughness due to milling), upstream flow conditions (i.e. velocity statistics, turbulence length-scales and the flow-structure in general) and the chemical properties of water (including the concentration of nuclei) [43]. These are all factors that are difficult to identify with a parameter (or a set of parameters), yet, they can have a measurable effect on disinfection efficiency. In order to circumvent this issue, the experiments presented herein were carried out using always the same hydraulic circuit (which presumably maintained similar flow conditions upstream of the HC reactor), the same orifice-plate (i.e., no changes in the slightest details of the orifice-geometry) and ultra-pure water (which, from the point of view of water-chemistry, should guarantee similar initial conditions). However, it is not always straightforward, especially in applications, to have such controlled conditions, therefore caution should be used when either comparing results from experiments carried out in different facilities or when extending laboratory results to field applications.

Another key issue is that it is not easy to perfectly isolate the effect of individual non-dimensional parameters, often because technical limitations prevent to control or monitor the actual value of some dimensional parameters. For example, the experiments presented herein were designed to isolate the effects of the cavitation number σ_v as, for each series of trials, the other non-dimensional parameters listed in Equation (3) were assumed to be constant. A key hypothesis underpinning this argument is that v_h, could be estimated from continuity principles, as the ratio between the flow rate and the holes area. This is representative of the velocity at the downstream end of the holes in the case of non-cavitating flows. When cavitation occurs, it is well known that, due to the pressure drop caused by flow separation at the orifice inlet, a cloud of water-vapor forms, meaning that the flow exiting from the orifice is multiphase with an average density and velocity, which are very difficult to measure/control and are clearly dependent on the cavitation number [55, 56]. Therefore, strictly speaking, besides σ_v, the non-dimensional parameters containing v_h (i.e. the Reynolds and the Weber number) probably varied a little among different tests pertaining to the same group (i.e. the same value of σ_v). Whether such variations can have significant effects on the disinfection efficacy remains an open question. One of the difficulties in providing an answer to this question and, more generally, in the use of empirical approaches, is that dimensional analysis is only a tool to find links between dimensional variables but hardly gives any hint to understand the processes controlling the problem of interest, which is a key prerequisite for the interpretation of experimental data. Moreover, this lack of understanding makes it difficult to quantify the effects of non-dimensional parameters other than through blind data-fitting, whose validity is often limited to the dataset it is applied to.

Within this context, the authors claim that, one of the tightest bottlenecks for the development of efficient HC reactors is the complete lack of understanding of what, from a purely mechanical point of view, kills bacteria. This is

because, in HC reactors, besides imploding bubbles, many other processes are triggered, which could be harmful to microorganisms. For example, Dular and co-workers [57, 29], argue (and provide good evidence) that fast and abrupt pressure differences are much more effective than imploding bubbles in killing pathogens in water. Moreover, there is quite a substantial literature demonstrating that turbulence can induce fluid stresses that can be lethal to microorganisms [44]. Until it will not be possible to quantify the sensitivity of microorganisms to fluid shear and normal stresses (and to the non-dimensional parameters that control the magnitude of such stresses), it will be extremely difficult to design and optimize HC reactors or other mechanically-based means of water disinfection.

7. Conclusions

The interest in the use of HC as a water-disinfection technique has grown fast in the recent years, both from an academic and an industrial point of view. The studies available from the literature have proved that HC is a very promising and flexible technique which can be used alone or in series with other methods (e.g., chlorination). However, robust and reliable design tools that allow to go from the laboratory to full scale applications are, to the best of the authors' knowledge, not available yet. This is clearly caused by the fact that cavitating flows are poorly understood, and hence difficult to model, as they involve turbulent multiphase flows occurring in complex geometries, which leave little hope to theoretical or computational modeling approaches.

As a result of this complexity, the vast majority of the literature approaches the problem from an empirical point of view. Empirically-derived design-relations can be very effective but must be determined from a large number of experiments, which must be designed and carried out on the basis of a rigorous dimensional analysis. While dimensional analysis is customarily adopted to tackle an enormous amount of engineering problems within the remit of Fluid Mechanics, it has surprisingly never been adopted within the field of HC and this represents a major shortcoming the present paper attempts to address. In particular, by application of dimensional analysis and the Buckingham-π theorem, we have derived Equation (3), which provides a set of non-dimensional parameters governing the simple problem of disinfection via HC triggered by circular orifice plates.

On the basis of this set of parameters, a number of experiments were designed and carried out to investigate the effects of the cavitation number and the dimensionless initial concentration on disinfection efficiencies. Results from these experiments indicate that C/C_0 vs n_p curves are not influenced by the initial concentration whereas, although heavily masked by experimental uncertainty, the effects of σ_v seem to be present. This points towards confirming the significant role played by the formation and implosion of bubbles in the game of disinfection and provides a first step towards the development of effective empirical formulae for the design of HC reactors.

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However, as discussed in the previous section, the development of effective empirical formulae cannot be left to an arid coupling between experiments and dimensional analysis but must be supported by a sound understanding of the physical processes controlling disinfection in HC reactors. In particular, the authors recommend that future research efforts should be directed towards fundamental studies aiming at understanding the effects of fluid stresses on microorganisms.

Acknowledgments

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8. Appendix A

Jyoti and Pandit [48] explored the microbicidal effectiveness of various cavitating reactors for naturally-contaminated bore well water. They made a comparative analysis of different disinfection techniques, including ultrasonication (AC), high-speed homogenisation (HC), high-pressure homogenisation (HC) and a cavitating valve (HC). In ultrasonication and high-speed/pressure homogenisation they treated a small water volume (1 l). For the case of the cavitating valve, they treated 75 l of bore well water at three different pump discharge pressures (P_1) of 1.72, 3.44 and 5.17 bar, obtaining an increase in the disinfection efficiency when the pump discharge pressure increased. They observed that HC was, energetically, the most efficient technique, resulting in maximum bacteria concentration drops of 44% at $P_1 = 5.17$ bar. The authors provided confidence intervals of the results estimated via repeated trials but failed to provide details about the geometry of the valve and the cavitation numbers reached during the experiments.

Kalumuck et al. [46] used the DynaJets® cavitating device to investigate the effects of cavitation on a small volume of 1.5 liters of high concentrated solution of $E.Coli$ ($5 \times 10^8 - 2 \times 10^9$ CFU/ml). Four experiments were conducted in a pressure ranges of P_1 between 4.13 and 5.17 bar and a single experiment at 10.3 bar, but no information on the associated cavitation number were provided. In the run performed at 10.3 bar, they achieved up to $5\,log_{10}$ reduction in the concentration of $E.\ Coli$ in 30 minutes, while the experiments executed in the pressure range between 4.13 and 5.17 bar shown a $3\,log_{10}$ reduction in the first $20 - 40$ minutes. Three more experiments were performed at moderate initial concentration of $E.\ Coli$ (10^7 CFU/ml). In this case, they obtained a $3\,log_{10}$ and $5\,log_{10}$ reduction in bacteria concentration at 20 and 30 minutes, respectively. They also reported a bacterial reduction of $0.6\,log_{10}$ attributed exclusively to the pump. No data are provided about the reactors' geometry.

Balasundaram and Harrison. [49] investigated the $E.\ Coli$ cell damage due to hydrodinamic cavitation, by analysing the periplasmic and cytoplasmic proteins released from the cell wall destruction. A wide range of cavitation numbers σ_v between 0.13 and 0.92 was investigated and the maximum extent of proteins release was found at $\sigma_v = 0.17$. They also investigated the influence of cell growth rate, finding a lower resistance to cavitation of cells grown at a higher growth rate. In a later work [27] they presented the influence of the geometry and the number of orifices on selective release of periplasmic proteins. Configurations with circular, squared and rectangular orifices were studied. For the same holes-area, the release of total soluble proteins was similar, however the plate with circular holes allowed for a greater release of acid phosphatase. They also studied the influence of the flow rate on the release of acid phosphatase after 1000 passes, finding higher percentage of release for higher flow rates. The best configuration was the one with the higher number of circular holes, were the flow rate was maximum. Unfortunately, in this study no information about initial concentration and bacterial survival rate was provided.

Azuma et al. [50] proposed a high pressure cavitating device with two cav-

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itating orifices in series and a plunger pump capable of discharging pressures up to 1050 bar. The cavitation numbers (σ_v) used in the study varied between 0.037 and 0.487, while the upstream nozzle velocity varied between 176 m/s and 384 m/s. No information about the downstream nozzle velocity and cavitation number were provided. In the second phase of the experiments they compared sterilization rate among Gram-positive (*Bacillus Subtilis*, *Bacillus Halodurans*) and Gram-negative (*Escherichia Coli*, *Pseudomonas Putida*) bacteria. The disinfection mechanisms suggested in this work are the high shear stresses reached in the orifice and the shock waves generated by bubbles' collapses. They achieved a complete disinfection of a mixture of water and *E. Coli* in three successive treatments at $\sigma_v = 0.154$. The experiments comparing Gram-positive and Gram-negative bacteria resistance to cavitation showed that Gram-positive bacteria are stronger than Gram-negative bacteria under the two conditions studied, namely $\sigma_v = 0.104$ and $\sigma_v = 0.037$. This behavior was ascribed to the more resistant cell-wall of Gram-positive bacteria.

Sawant et al. [45] studied the effect of a single orifice plate on the disinfection of the zooplankton in sea water. In all the experiments just once pass through the cavitation device was made. The test loop was composed of a centrifugal pump, a valve and a single orifice-plate positioned in sequence. During the experiments, they isolated the effects of the cavitating valve, the orifice plate and the pump, individually. The maximum percentage of disinfection due to the pump and the valve was 57% while almost 28% of the zooplankton was killed by the pump alone. The maximum percentage of killing achieved with the orifice plate (and the valve fully open) was 82%, related to a cavitation number (σ_v) equal to 14.68. Similar values of disinfection efficiencies were obtained in spite of wide differences in cavitation numbers tested. This behavior was explained as an effect of the weak cell wall of zooplankton.

Arrojo et al. [12] compared the disinfection efficiency of different orifice plates and Venturi tubes, varying the numbers of holes, the discharge pressure and the initial concentration of *E. Coli*. For an initial concentration of 10^7 CFU/ml, they found a higher disinfection efficiency for the configuration with the highest number of holes with the smallest diameter. The experiments performed with orifice plates showed a first stage where the CFU number increased. This lag-phase lasted for about 30 minutes and the authors explain this behavior as an effect of bacteria-agglomerates fragmentation. from the comparison between the orifice plate and the Venturi-tube they found that, in order to develop the same number of cavitating events, orifices plates need a higher discharge pressure (P_1) than Venturi tubes. They also point out that cavitation achieved with orifice plates is resulting in more violent cavity collapses due to the sudden pressure recovery. Acting on in initial concentration in the interval 10^3 - 10^5 CFU/ml, they found that, for orifice plates, the higher is the initial concentrantion the lower is the disinfection efficiency while Venturi-tubes showed no correlation between disinfection efficiency (C/C_0) and initial *E. Coli* concentration. In this study the cavitation number for the various trials is not specified.

Loraine et al. [47] compared different types of cavitating devices, including

the so-called DynaJets®, orifice plates, the so-called StratoJet® and a single orifice DynaSwirl®, all with the same total holes' area. The first group of disinfection experiments aimed at comparing the disinfection efficiency associated with different types of gram-negative bacteria. The first test was performed with a single orifice DynaSwirl® cavitating jet operating at 2.1 bar. The initial concentration was 10^7 CFU/ml with a test batch volume of 2 litres. Both Klebsiella Pneumoniae and $E.\ Coli$ underwent a $5\,log_{10}$ reduction in 60 minutes, corresponding to a 99.99% removal. A similar experiment with an 8-orifice StratoJet® operating at 16.5 bar and a batch volume of 1.8 l was used to compare disinfection efficiency for $E.\ Coli$, $Pseudomonas\ Syringae$ and $Pseudomonas$ $Aeruginosa$. This test showed approximately half efficiency in $E.\ Coli$ disinfection ($5\,log_{10}$ reduction in 120 minuts). Nearly $3\,log_{10}$ decrease in $P.\ Aeruginosa$ concentration was observed in 90 min, while $P.\ Syringae$ concentrations showed a $6\,log_{10}$ reduction in 20 min. These differences in disinfection efficiencies were ascribed to the degree of cross-linking in the peptidoglycan layer of the cell walls. However, when the results are presented as a function of the number of passes through each reactor, the differences in removal efficiency of $E.\ Coli$ between the single orifice DynaSwirl® and the 8-orifice StratoJet® were relatively small.

These authors investigated the DynaSwirl® at operating pressure drops ($P_1 - P_2$) of 3.45, 2.1 and 1 bar, corresponding to cavitation numbers (σ_v) of 0.33, 0.5 and 1, respectively. The best disinfection efficiency was found for $P_1 - P_2 = 2.1$. At this pressure drop the authors investigated disinfection efficiencies for $E.\ Coli$ (gram negative) and $B.\ Subtilis$ (gram positive). $B.$ $Subtilis$ concentrations were reduced by $4.5\,log_{10}$, while $E.\ Coli$ concentrations were reduced by more than $7\,log_{10}$. This experiment confirms that the thick cell wall of gram-positive bacteria is more resistant to cavitation then the thin cell wall of gram-negative species. A sensitivity analysis was carried out by varying the initial $E.\ Coli$ concentration between 10^3 and 10^9 CFU/ml. General trends showed a slow initial reduction in the concentration followed by a higher reduction rate until the concentration fell below 100 CFU/ml. The initial lag period, where the bacterial concentration remained approximately constant, lasted longer for higher concentrations, while during the rapid reduction phase the disinfection efficiencies were comparable for all cases. Standard deviation of the bacteria concentrations were calculated from the duplicates of the CFU/ml measurements, but no information about the number of trials were provided.

Wang et al. [35] evaluated the effectiveness of hydrodynamic cavitation on bore well water disinfection. They compared the effect of HC alone with a hybrid system whereby HC was combined with the use of sodium hypochlorite and chlorine dioxide. All the hybrid experiments showed an increase in disinfection efficiency. This study also investigates the effects of the reactor geometry (i.e. by varying the number and diameter of holes) and of the inlet pressure (P_1), but no information on the investigated cavitation numbers were provided. All the experiments were carried out using relatively low concentration of $E.\ Coli$ ($2500 - 3000$ CFU/ml). It was observed that the higher the inlet pressure (i.e. P_1) the higher the disinfection efficiency. Furthermore it was observed that for a given constriction area, more holes of smaller diameter lead to improved

disinfection efficiencies. In this study, confidence intervals on the measured concentration are not provided.

Badve et al. [51] investigated HC within the context of microbial disinfection of ships ballast water. The initial concentration of microbes for all the experiments was around 10^7 CFU/ml. They compared orifice plates and Venturi tubes limiting the number of passes through the devices to 50. Results show that Venturi tubes work better than single orifice plates. No precise information about the cavitation numbers of the various configurations were provided.

Filho et al. [52] used a high pressure cavitating jet apparatus to inactivate *E. Coli* in artificially - and natural - contaminated water. For the former, they achieved a disinfection efficiency up to 90% in 15 minutes at 100 bar. After 30 minutes, the inactivation rate reached 98.30, 99.96 and 100% at pressure of, 80, 100 and 120 bar, respectively. No information about the cavitation number characterizing the system was found. For naturally-contaminated water (i.e., for concentrations of *E.Coli* around $10 - 100$ CFU/ml) the disinfection efficiency was independent of the jet pressure. After 30 minutes, inactivation rates of 98.89 and 97.31% were reached for discharge pressures of 100 and 50 bar, respectively. Also in this work, confidence intervals on the measured concentration are not provided.

Liu et al. [53] used a multi-orifice plate made of 49 holes of 1 mm diameter for the disinfection of *E.Coli*. A single reactor geometry was studied with an initial concentration of bacteria equal to $1.6 \times 10^5 CFU/100$ ml. This device reached a disinfection efficiency of 98% in 60 minutes. The authors did not provide information regarding the cavitation number characterizing the system studied as well as they did not indicated the number of trials and the confidence intervals on the measured concentration.

9. Appendix B

Table 5: Most Probable Number (MPN) of the CFU values in the single disinfection experiment (run) plotted in Figure 3, with upper and lower limit of the 95% confidence interval [54].

σ_v	run	t (min)	n_p	MPN/100 ml	Lower limit	Upper limit
0.2	1	0	0	579400	379100	847200
0.2	1	30	51	101900	72700	140400
0.2	1	60	103	88200	62900	120200
0.2	1	90	154	49500	34400	69300
0.2	1	120	206	26200	16600	39700
0.2	1	150	257	18900	11300	30400
0.2	1	180	309	21300	12700	32600
0.2	1	210	360	14600	8200	24600
0.2	1	240	411	18500	11000	29200
0.2	1	270	463	12200	6800	21400
0.2	1	300	514	6300	2900	13700

Table 5: Most Probable Number (MPN) of the CFU values in the single disinfection experiment (run) plotted in Figure 3, with upper and lower limit of the 95% confidence interval [54].

σ_v	run	t (min)	n_p	MPN/100 ml	Lower limit	Upper limit
0.2	1	330	566	3100	700	8900
0.2	1	360	617	5200	1800	10800
0.2	2	0	0	365400	231900	555500
0.2	2	30	51	209800	145500	301100
0.2	2	60	103	77600	55300	104500
0.2	2	90	154	69700	49700	95300
0.2	2	120	206	58300	40500	80600
0.2	2	150	257	25600	15700	38400
0.2	2	180	309	34500	23300	50100
0.2	2	210	360	26900	17100	39800
0.2	2	240	411	16100	12400	32300
0.2	2	270	463	14800	8500	25100
0.2	2	300	514	5100	1700	10600
0.2	2	330	566	6300	2900	13700
0.2	2	360	617	3000	700	7400
0.2	3	0	0	32550	20660	49810
0.2	3	30	51	18720	12610	28100
0.2	3	60	103	14210	10130	19680
0.2	3	90	154	8570	6110	11720
0.2	3	120	206	8130	5790	11140
0.2	3	150	257	4320	2910	6140
0.2	3	180	309	3180	2080	4640
0.2	3	210	360	2180	1340	3390
0.2	3	240	411	630	290	1370
0.2	3	270	463	200	30	710
0.2	3	300	514	100	10	550
0.2	4	0	0	4884	3100	7215
0.2	4	30	51	2481	1623	3719
0.2	4	60	103	2143	1402	3209
0.2	4	90	154	1658	1149	2380
0.2	4	120	206	767	546	1062
0.2	5	0	0	3076	1953	4712
0.2	5	30	51	2098	1455	3011
0.2	5	60	103	1081	770	1472
0.2	5	90	154	657	468	892
0.2	5	120	206	537	383	740
0.2	6	0	0	1664	1154	2340
0.2	6	30	51	404	273	574

Table 5: Most Probable Number (MPN) of the CFU values in the single disinfection experiment (run) plotted in Figure 3, with upper and lower limit of the 95% confidence interval [54].

σ_v	run	t (min)	n_p	MPN/100 ml	Lower limit	Upper limit
0.2	6	60	103	218	134	339
0.2	6	90	154	109	56	195
0.2	6	120	206	52	23	119
0.2	7	0	0	727	476	1049
0.2	7	30	51	501.2	357	688
0.2	7	60	103	261.3	171	399
0.2	7	90	154	172	116	261
0.2	7	120	206	73.8	53	100
0.4	8	0	0	17220	11940	24500
0.4	8	30	51	13540	9650	18400
0.4	8	60	103	10860	7740	15000
0.4	8	90	154	9060	6460	12410
0.4	8	120	206	8160	5820	11030
0.4	9	0	0	5810	4140	7950
0.4	9	30	51	4410	3060	6250
0.4	9	60	103	3170	2070	4660
0.4	9	90	154	1610	930	2680
0.4	9	120	206	1460	820	2460
0.4	10	0	0	2142	1527	2944
0.4	10	30	51	987	723	1337
0.4	10	60	103	441	306	625
0.4	10	90	154	189	113	304
0.4	10	120	206	75	36	149
0.4	11	0	0	410.6	260.6	618.9
0.4	11	30	51	148.3	123.1	177
0.4	11	60	103	21.8	13.4	33.1
0.4	11	90	154	6.3	2.5	12.7
0.4	11	120	206	0	0	0
0.4	12	0	0	295	188	440
0.4	12	30	51	301	197	442
0.4	12	60	103	135	78	234
0.4	12	90	154	120	60	203
0.4	12	120	206	20	3	71
0.4	13	0	0	166.4	115.4	234
0.4	13	30	51	90.8	66.5	123.1
0.4	13	60	103	28.8	18.3	42.7
0.4	13	90	154	11	5.7	20.1
0.4	13	120	206	9.8	4.7	18.4

Table 5: Most Probable Number (MPN) of the CFU values in the single disinfection experiment (run) plotted in Figure 3, with upper and lower limit of the 95% confidence interval [54].

σ_v	run	t (min)	n_p	MPN/100 ml	Lower limit	Upper limit
0.65	14	0	0	1732900	1167700	2709500
0.65	14	60	103	1046200	705000	1509000
0.65	14	120	206	727000	475700	1048900
0.65	14	180	309	290900	190400	446100
0.65	14	240	411	151500	108000	207800
0.65	15	0	0	32700	19000	44400
0.65	15	60	103	21800	13400	33900
0.65	15	120	206	7500	3600	14900
0.65	15	180	309	2000	300	7100
0.65	15	240	411	0	0	370
0.65	16	0	0	2755	1857	4168
0.65	16	60	103	860	613	1155
0.65	16	120	206	201	124	318
0.65	16	180	309	10	1	55
0.65	16	240	411	10	1	55
No Plate	17	0	0	307.6	195.3	471.2
No Plate	17	30	60	344.8	218.9	520.7
No Plate	17	60	120	461.1	292.7	687.9
No Plate	17	90	180	344.8	218.9	520.7
No Plate	17	120	240	344.8	218.9	520.7
No Plate	18	0	0	209.8	145.5	301.1
No Plate	18	30	60	204.6	137.9	306.9
No Plate	18	60	120	185	131.9	256.3
No Plate	18	90	180	204.6	137.9	306.9
No Plate	18	120	240	185	131.9	256.3
No Plate	19	0	0	3448	2189	5207
No Plate	19	10	20	3654	2319	5555
No Plate	19	20	40	4884	3100	7215
No Plate	19	30	60	3255	2066	4981
No Plate	19	60	120	5172	3384	7636
No Plate	19	90	180	3076	1953	4712
No Plate	19	120	240	4352	2762	6500