An ingestible capsule for the photodynamic therapy of helicobacter pylori infection

An Ingestible Capsule for the Photodynamic Therapy of *Helicobacter pylori* infection

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***Abstract*— *Helicobacter pylori* (*H. pylori*) is a Gram-negative pathogen bacterium affecting the mucosa of the stomach and causing severe gastric diseases. *H. pylori* related infections are currently treated with pharmacological therapies, which are associated with increasing antibiotic resistance and consequent reduction of the efficacy down to 70-85%. Moreover, drugs have generally side effects that further affect the healthcare system in terms of additional financial and medical efforts. The aim of this work is to present an innovative device for the treatment of *H. pylori* infection, consisting of an ingestible lighting capsule performing photodynamic therapy by means of light at specific wavelengths. The proposed treatment is minimally invasive and the described system can be considered the first photodynamic swallowable device ever proposed. Preliminary experiments demonstrated that the capsule integrated with LED sources can provide the required lighting power to kill the bacterium with an efficiency up to about 96%.**

***Index Terms*— Helicobacter pylori, antibiotic resistance, therapeutic capsule, swallowable device, photodynamic therapy, LED**

I. Introduction

HELICOBACTER pylori (*H. pylori*) is a Gram-negative microaerophilic bacterium that colonizes the mucus layer of the stomach and duodenum [1]. The prevalence of the infection is more than the 50% of the world population, up to 90% in developing countries.

H. pylori can cause several diseases like as chronic gastritis, gastric and duodenal ulcers, gastric lymphoma, adenocarcinoma, and extragastric disorders such as idiopathic thrombocytopenic purpura, iron deficiency anemia and vitamin B12 deficiency. *H. pylori* is also considered as a class 1 carcinogen agent by the World Health Organization.

Currently, *H. pylori* infection is treated with a pharmacologic therapy consisting in a combination of a proton pump inhibitor with two or three antibiotics showing high failure rates: due to several side effects and antibiotic resistance, the efficacy of the pharmacologic therapy is reduced to 70-85% [1][3].

In order to overcome these limitations, the photodynamic therapy (PDT) approach has been explored worldwide for the treatment of this pathology. PDT was introduced at the beginning of the last century and originally used for tumor treatment. In traditional PDT, an external non-toxic dye, called photosensitizer, is injected or topically applied to the patient, being selectively accumulated in the target (i.e. a malignant tissue, a bacterium). After about 48-72 hours, the target is exposed to visible light. The interaction between the photosensitizer and the light, in the presence of oxygen, causes oxygen reactive species to be produced, thus inducing cell death [4]. PDT applications to kill pathogenic microorganisms are encountering the growing attention of clinicians, so that it has been proposed as a therapy for a large variety of localized infections. This revival of interest has largely been driven by the inexorable increase in drug resistance amongst many classes of pathogens. One of these bacteria, i.e. *H. pylori*, is known to be photo-labile without the exogenous assumption of photosensitizers [5]. *H. pylori*, in fact, naturally owns photoactive porphyrins, coproporphyrin and protoporphyrin IX, that are natural photosensitizers. Thus, *H. pylori* can be killed by exciting them with appropriate wavelengths, for example by means of a traditional gastroscope opportunely modified with optical fibers for light delivery [6] or a light-emitting inflatable balloon [7]. It is to be considered that all these solutions relied on the natural presence of oxygen in the treated areas and were associated with a good outcome in terms of killing efficacy [8]. However, these solutions have clear disadvantages, related to poor patient compliance due to their invasivity, to the high number of gastroscopic procedures to undergo, pain and risks of gastric perforation. To overcome these issues, wireless capsule endoscopy has been proposed in recent years [9], mainly as a diagnostic solution. Wireless capsule endoscopy is a non-invasive diagnostic method that reduces the level of discomfort and can be well tolerated by patients, with very few contraindications [10]. Although this technique was originally developed for the screening of the small bowel, it has been later applied to other gastrointestinal (GI) districts, such as the colon [11][12], esophagus and stomach [13][14][15]. One example of non-drug based therapeutic device has been proposed, with applications for the colon [16]. None of these devices can be applied to the therapy of *H. pylori*.

In this paper, the design and preliminary validation of a wireless ingestible capsule is presented as the first PDT

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swallowable device ever proposed. The design of the capsule and preliminary results are presented.

II. Materials and Method

From a technical viewpoint, it is essential to focus on the light emission module during the preliminary development phase before proceeding to the integration of the final device. In particular, the light emission module has to be dimensioned considering the required light intensity necessary to kill the bacterium. Afterwards, the module has been tested *in vitro* on the bacterium. Finally, the integration of the whole capsule has been performed, considering the efficiency of the battery with respect to the required performance in terms of capsule lifetime.

A. Light emission module

In order to design an efficient and reliable emission module, i.e. the light emitters to incorporate into the capsule, the source parameters have been evaluated in terms of minimal energy required to perform an effective therapeutic action. As a first step, we focussed on *in vitro* irradiation conditions only. One of the most important parameters to analyse is the irradiance E (W/cm^2), corresponding to the light energy received by a surface (in our case: the *H. pylori* cultured area) per unit time and surface. In photobiology and photophysics, the integral of irradiance over the treatment time is often referred to as "energy dose" or simply "dose" (D), and depends on the radiant flux Φ_{watt} (W) of the considered source, on the irradiated surface S and irradiation time T by the formula:

$$D = \frac{1}{S}\int_0^T \Phi_{watt}(t)\cdot dt \qquad (1)$$

In literature [5] it has been demonstrated that the most effective wavelength range to kill the bacterium *in vitro* is 375-425 nm (blue), with an irradiation density of ~9-16 J/cm^2 (*H. pylori* reduction factor ~10^3-10^6 respectively). In the range 625-675 nm (red), the same value of 16 J/cm^2 is accompanied by a *H. pylori* reduction factor of about 20.

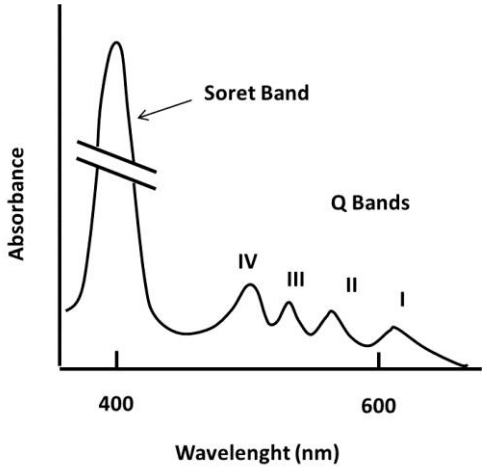

Figure 1 Typical porphyrine absorption spectrum (adapted from [18])

These dose values correspond to about 5-8.9 mW/cm^2 released in 30 minutes for red and blue respectively [5], being 30 minutes the typical gastric emptying time corresponding to the mean capsule permanence in the stomach.

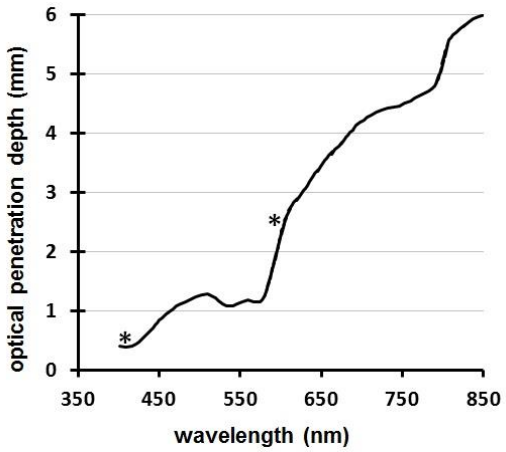

Figure 2. Penetration depth vs wavelength in human mucosa tissue (adapted from [19]). Red and blue wavelengths to which *H. pylori* is more sensitive are indicated by asterisks (405 and 625nm respectively).

These results reflect the structure of the typical absorption spectrum of the natural photosensitizers of *H. pylori* (Fig. 1), and must be coupled with the knowledge of the different light absorption by tissues. In fact, in view of *ex-vivo* and *in-vivo* experiments, the optical properties of the stomach wall may determine a shift in the best irradiation wavelengths. In particular, it is worth noting that red wavelengths are more penetrating in the human mucosa layer respect to blue ones, even if these last ones are more effectively absorbed by the photosensitizer. Embedding red and blue LED sources in the same devices will then improve the overall efficacy against *H. pylori*, considering that the bacterium is mainly located under a mucosal layer.

For all these reasons, low current LEDs at different wavelengths (λ) have been used, with nominal peaks at 375 and 460 nm (Nichia corp., Japan), 405 and 625 nm (Vishay Intertechnology, United States). In all cases, spectrum are broader than the nominal ones and cover the effective wavelenghts for killing *H. pylori*.

III. Experimental Tests

A. On bench experiments

In order to establish a proper timing for *in vitro H.pylori* irradiation necessary for eradication, LED emission spectrum, radiant flux and irradiance have been measured. The emission spectrum was acquired by a portable spectrometer (Avantes, The Netherlands). The radiant flux Φ_{watt} of the single chosen LED, i.e. the emitted power over the whole emission solid angle, has been evaluated by means of an integrating sphere (3A-IS-V1, Ophir, United States). The integrating sphere is a black body model which integrates the light emitted by the LED in all directions onto an embedded photodiode. Each LED has been powered with a battery (3.7 V, 30 mAh). The experimental setup for the measurement of the radiant flux is

shown in Fig. 3. LEDs are placed on the integrating sphere to measure the emission on the photodiode.

Fig. 3. Experimental setup for the measurement of the radiant flux. Left: integrating sphere where the LED is placed for testing. Right: power meter.

Each measurement has been repeated three times per LED, in order to improve statistical significance. The results of these measurements are reported in Table I and compared with the radiant flux calculated from the LED datasheets. For both 625 and 460 nm LEDs, these indicated the LED luminous intensity I_v (cd) as a mean typical value over all emission directions. Starting from I_v, the luminous flux Φ_v in lumen has been calculated by the formula:

$$\Phi_v = 2\pi\langle I_v\rangle \qquad (2)$$

considering a total emission solid angle of 2π and a linear behaviour of $I_v(\Omega)$. Then, the radiant flux Φ_{watt} has been derived as follows:

$$\Phi_{watt} = \frac{\Phi_v}{683\cdot V(\lambda)} \qquad (3)$$

where $V(\lambda)$ is the luminous efficiency function (photopic vision). For our verification purposes, we limited calculations to a very simple model, neglecting the LED spectral FWHM (Full Width at Half Maximum) and calculating $V(\lambda)$ in correspondence to the respective measured emission peak maximum (see Table I), as eq. 3 is strictly valid for a monochromatic source..

TABLE I
MEASUREMENTS OF THE LED TOTAL RADIANT FLUX

Measured peak wavelength (nm)	*Nominal peak wavelength (nm)*	*Measured radiant flux (mW)*	*Calculated radiant flux (mW)*
632	*625*	7.4±0.1	16
460	*460*	5.0±0.8	7.35
402	*405*	3.25±0.26	6.8*
379	*375*	6.86±1.6	9.9*

* information given directly in the LED datasheet as typical value

It is worth noting that there are discrepancies between theoretical and measured values. We have to consider that datasheet report typical emission values for I_v, whose variability may well be up to more than 50%. In addition, a more accurate calculation would need to account for the spectral width of the LED emission (typical FWHM is 15-25 nm) to obtain the radiant flux by modifying eq. 3, which is beyond the scope of this article. For all these reasons, we relied on our measured values rather than on datasheet ones.
By considering the LED datasheet, where I_v/I_{vmax} is plotted vs. α (data not shown, see Fig. 4 for α), we derived the behaviour of I_v/I_{vmax} vs. the solid angle for light emission Ω by considering its definition: $\Omega=2\pi(1-\cos\alpha)$, finding a very good agreement with a linear behaviour represented by the relationship:

$$I_v(\Omega) = I_{v\max}\left(1-\frac{\Omega}{2\pi}\right) \qquad (4)$$

with $0 \le \Omega \le 2\pi$, being $\Omega = 0$ the null solid angle corresponding to the forward LED axis direction ($\alpha=0$). Correspondingly, the luminous flux $I_v(\Omega)$ is derived by integrating over Ω, obtaining $I_v(\Omega) \propto \Omega^2$. For a monochromatic source and thanks to eq. 3, it follows that the radiant flux is:

$$\Phi_{watt}(\Omega) = k\cdot\Omega^2 \qquad (5)$$

where k is an unknown constant depending on the maximum value for $\Phi_v(\Omega)$. For our purposes, we can model the single LED irradiation by Fig. 4, where d is the distance between the LED and the illuminated surface, A is the irradiated circular area of radius r due to emission within the solid angle Ω and the LED emission is concentrated in its center point. We define $A_{90\%}$ as the "effective irradiated area" where 90% of the total radiant flux is delivered. This choice defines the value for $\Omega=\Omega_{90\%}$. Correspondingly we find $\alpha_{90\%}$ by applying the definition of Ω: $\alpha_{90\%} = \arccos(1/\sqrt{10})$. In turn, this defines $A_{90\%}$ as (see also Fig. 4):

$$A_{90\%} = \pi\cdot\left(d\cdot tg(\alpha_{90\%})\right)^2 = 9\pi\cdot d^2 \qquad (6)$$

It is worth noting that $\Omega_{90\%}$, and consequently $A_{90\%}$ do not depend on k (eq. 5), which means we do not need to know the maximum LED luminous intensity (often unknown) to define them. In this way we can associate an illuminated area to each LED. This determines an error of about 10%, which seems acceptable for our modeling purposes.

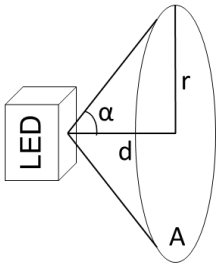

Figure 4 Irradiated area A at distance *d* for each LED.

For *in vitro* irradiation, a dedicated Delrin® support has been fabricated, as shown in Fig. 5. Afterwards, a single LED has been embedded in a 2 mm transparent resin and positioned directly onto the photodiode.

Fig. 5. Dedicated LED support for in vitro irradiation (left) of *H.pylori* cultures (right).

Measurement of irradiance uniformity at a 2 cm distance (data not shown) for a single LED in a fixed LED position has given a value of about 15%, defined as maximum deviation from mean value over the Petri dish surface. During *in vitro* irradiation, light diffusion due to the Delrin® support occurred, so that almost all the radiant flux was impinging on the Petri dish surface (data not shown).

B. In vitro experiments

H.pylori laboratory-adapted strain (ATCC 49503) was routinely grown in liquid medium consisting of Brucella broth at 37°C in a microaerophilic atmosphere. After 96 hours it was streaked on Blood Agar plates until colonies were easily countable.

Experiments have been performed to assess the killing efficacy of the chosen LEDs at the different wavelengths. After growing on agar plates, an aliquot corresponding to a bacterial concentration of 800000 Colony Forming Units/ml (CFU/ml) was used and *in vitro* light bacterial irradiation was performed on phosphate-buffered saline (PBS) in Petri dishes. During light delivery, dishes with *H.pylori* in liquid medium were maintained in plastic bags at 37°C in microaerophilic atmosphere and sterile conditions. For this reason, the experimental setup of Fig. 5 (LED in a fixed position) has been equipped with a magnetic reed switch allowing to turn the LED modules on and off in an easy way without opening the envelop. The final configuration of the experiment is shown in Fig. 6. Irradiation time was 30 minutes for all samples, separately with red (625 nm) and blue (405 nm) light. These LED modules correspond to those integrated in the capsule, as described in Section IV. During irradiation, envelops with cultured bacteria were kept at 37°C inside a cell incubator in microaerophilic atmosphere and sterile conditions. A control sample (no irradiation) was subject to the same procedure.

Fig. 6. Transparent envelop with inside the dedicated irradiation LED support and the *H. pylori* cultures (idle).

Then, survival fraction determination was performed. Following light irradiation, a suspension aliquot was removed from each Petri dish, subjected to serial tenfold dilutions and plated at 37°C in microaerophilic conditions on Blood Agar for 4 days. Surviving CFU were counted and recorded for analysis and survival fractions were determined relative to unilluminated bacterial suspensions. Irradiation results are shown in Table II.

TABLE II
IRRADIATION OF *H. PYLORI* CULTURES

Nominal wavelength (nm)	*H. pylori concentration after irradiation (CFU/ml)*	*CFU reduction after irradiation (%)*
625	40000	95
460	30000	96.25
405	30000	96.25
375	50000	84

IV. SYSTEM INTEGRATION

After proving the *in vitro* efficacy of the LEDs, the ingestible capsule device has been designed in order to integrate all the needed components and considering dimensions that allow an easy swallowability (i.e. the target dimensions are the ones of commercial capsules: diameter 11-14 mm; length 26-30 mm). In order to prevent capsule retention, the clinical protocol foresees since now the use of a dissolvable (dummy) capsule to verify the functional patency of the small bowel and consequently identify those patients who can safely undergo a capsule therapy [20]. Two capsule types have been designed and built: a research capsule, a prototype including the LEDs and a more performant battery that is still not acceptable for clinical use, and a pre-industrial

prototype, including components that are certified or certifiable for clinical use. Both types have been equipped with 8 LEDs positioned on an electronic board including a magnetic switch (Memscap, USA) and a battery. The magnetic switch is normally in a closed reed state and is used to switch on the capsule power by removing the device from a magnetized external case when in use.

The research prototype has been built in two different versions, one emitting red light only (625nm) and the other emitting blue light only (405nm). The light sources correspond to the above-described LED modules (405 and 625nm). Each prototype has a polymeric shell 27 mm in length and 14 mm in diameter. Two symmetric transparent windows are integrated in the lateral area of the capsule for enabling illumination from the internal light sources. The LEDs are positioned on a PCB having dimensions of 13 mm x 16 mm x 2 mm, designed and dimensioned for embedding the LEDs and the electronic components. The 3D sketches of both research and pre-industrial prototypes with main components are shown in Figs. 7 and 8. A LiPo (Lithium-Polymer) battery (3.7 V, 30 mAh, Plantraco, Canada) has been used for the research prototype. Although this battery has high energy density (200 Wh/kg) and a convenient form factor (13 mm x 17 mm x 5.7 mm), still it is non-certified for clinical use. However, it is commonly used for capsule research devices and miniaturized robots as main power source [14][15][17].

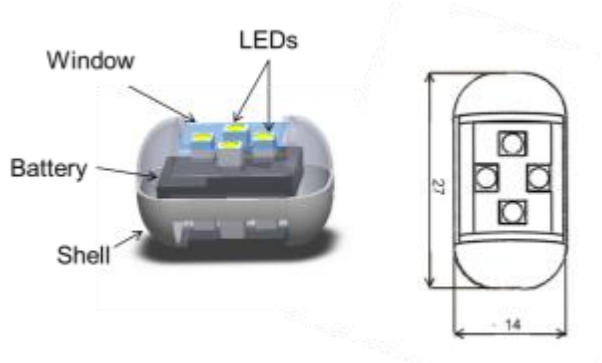

Fig. 7. Research prototype (14 mm diameter, 27 mm length); 3D sketch (Left) and top view (Right).

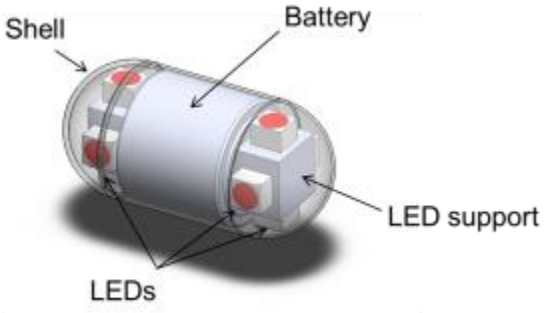

Fig. 8. Pre-industrial prototype 3D sketch (12 mm diameter, 27 mm length).

The pre-industrial prototype has been built in one version only emitting red light at 625nm; it has a transparent polycarbonate shell having dimensions of 27 mm length and 12 mm diameter. The capsule has been equipped with 8 LEDs positioned with radial symmetry, in order to maximise the total irradiated area and for adapting the internal geometry to the battery shape factor. The LEDs are positioned on a LED support having dimensions of 6 mm x 6 mm x 4.5 mm embedding both LEDs and electronic components. The power source consists in a 2L76 3-Volt Lithium cylindrical battery (160 mAh, 11.6 mm diameter, 10.6 mm height). A reed switch has been included as well in analogy with the research prototype. The used battery is certifiable for clinical use, being a traditional Lithium-Ion battery commonly used in implanted medical devices (i.e. pacemakers).

A comprehensive model of the light power per unit surface impinging on the various regions of the stomach (and stomach antrum in particular) due to capsule irradiation is beyond the scope of this article. Nevertheless, for both prototypes we present two exemplary cases, corresponding to a "short" (2mm) and "long" (15mm) mean distance (d, see eq. 6) between the capsule and the stomach wall in the collapsed and extended stomach conditions respectively. This very simple model considers the capsule ideally positioned along the symmetry axis of the antrum region, whose dimensions in the distended state are compatible with a total surface area of the order of 50 cm^2 and lateral dimensions up to more than 4cm. The mean time for therapy was estimated to be about 30 minutes, corresponding to the mean gastric emptying time.

The energy dose released from each capsule prototype has been calculated by using the measured radiant flux and capsule geometry, considering the presence and positioning of 8 LEDs for each prototype represented in Fig. 7 and 8. In the pre-industrial prototype, the irradiated area (eq. 6) is estimated to be ~9 cm^2, corresponding to eight times the spot produced by the single LED for the collapsed case (no significant lobe superposition) and >50 cm^2 for the distended case. In the research prototype, LED lobe superposition (Fig. 9) defines a smaller irradiation area of ~2·3.5 = 7 cm^2 in the collapsed case. In the distended antrum case, eq. 6 gives a value >50 cm^2, like with the pre-industrial prototype, meaning that the whole antrum surface is illuminated.

TABLE III
CAPSULE PROTOTYPE IRRADIATION PARAMETERS

PROTOTYPE	*STOMACH CONDITIONS*	*IRRADIATED AREA* ($A_{90\%}$, cm^2)	*MEAN ENERGY FLUENCE* (J/cm^2)	*MINIMUM N. OF CAPSULES TO REACH THE TARGET DOSE*
Pre-industrial	collapsed	9	11.8	2
	distended	50	2.1	11
Research	collapsed	7	6.7	3 (2)
	distended	50	0.94	17 (10)

Table III – Irradiation parameters for the capsule prototypes in the collapsed and distended stomach models (red light emission). Numbers in parenthesis refer to blue light emission.

In the cases where $A_{90\%}$>50cm^2, we assume that the mean power per unit surface illuminating the gastric wall is equal to the sum of all the LED emitted power divided by 50cm^2 (see also Table III). It is clear that in the collapsed stomach case, the target dose is reached only in the fraction of surface reached by the illumination. The pre-industrial red-light emitting prototype has a mean fluence of ~12 J/cm^2 calculated as 7.4 mW ·8·1800s over 9 cm^2. In order to reach the target energy dose of 16 J/cm^2 for red light, at least 2 or 11 capsules will be necessary in the collapsed and distended stomach case respectively. To test the performance of a different geometry for the capsule LED sources, we have also analyzed the case of an hypothetic *in vivo* application of the research prototype. This emits a mean fluence of about 6.7 or 0.94 J/cm^2, equal to 3.25mW·8·1800s over 7 or 50 cm^2 (collapsed and distended case respectively). Considering as before the respective cases of collapsed and distended stomach antrum wall, this means that at least 3 or 17 capsules would be necessary to treat the gastric target area for red light (~16 J/cm^2) and that 2 or 10 capsules would be necessary for blue light (~9 J/cm^2).

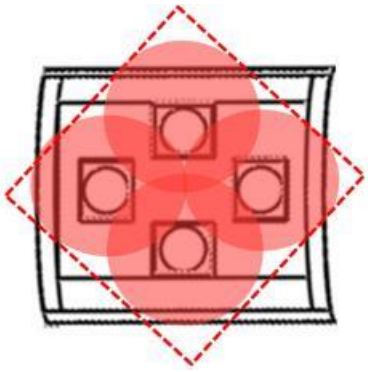

Figure 9: LED lobe superposition in the research prototype. Dotted red line represents the total irradiated area.

Fig. 10 and Fig. 11 show the red and blue light research and pre-industrial prototypes.

Fig. 10. Red (Left) and blue (Right) research prototypes.

Fig. 11. Pre-industrial red prototype: idle (Left) and alight (Right).

The pre-industrial prototype has been qualitatively tested on a distended stomach 3D model made in silicone (Fig. 12) in order to assess the overall light diffusion with respect to the capsule position.

The proposed capsule has no special parts for distending the stomach naturally collapsed in the fasting state. The distended condition can be obtained e.g. by making the patient drinking plain or sparkling water, that provide negligible light absorption; stomach distension reduces blind areas as well. Although stomach distension can help to expose the gastric internal surface to light, at the same time it increases the distance between the capsule and the irradiated tissue which is illuminated by a lower fluence (Tab. III), thus decreasing the single capsule efficiency. The need for stomach distension during the capsule treatment and the way it affects the overall efficacy will be evaluated in the future, as soon as a pilot clinical test in humans will be performed. In addition, bacteria located both on the surface and deeper in the tissue plicae can be targeted thanks to the red and blue light emission respectively.

Additional methods for enlarging the irradiated area will be evaluated. Among them, the use of a light diffusing material for the capsule surface can be an option, even if the use of LEDs with a wider emission angle seems to be a more promising solution, together with the use of additional LED sources.

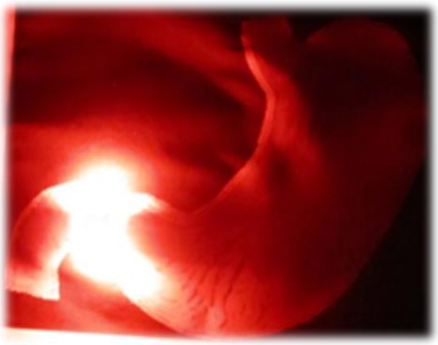

Fig. 12. Qualitative red light diffusion in a distended stomach model.

V. Conclusion

In this paper, an ingestible capsule for the photodynamic treatment of *H. pylori* has been proposed, built and characterized for the first time. As a first step, we confirmed the feasibility of *in vitro* photodynamic therapy of *H. pylori*, by means of tiny LEDs powered by a battery, in a completely wireless approach. LEDs at 375, 405, 460, 625 nm wavelengths have been used and their radiant fluxes measured, whose values fit the requirements for *H. pylori* killing, taken from scientific literature (threshold irradiation dose: ~16 J/cm^2 for 625 nm light and ~9 J/cm^2 for 405 nm, 30 minute therapy, 2 and 15 mm working distance). Both red and blue lights (625 nm and 405 nm respectively) were associated with a killing efficiency of about 96%. On the basis of these results, the LEDs, a battery and dedicated electronic boards have been embedded into both a research and a pre-industrial capsule

prototype. These have been fabricated in swallowable dimensions (27 mm length- 14 mm diameter and 27 mm length - 12 mm diameter respectively). The capsule can be considered as the first photodynamic swallowable device ever proposed. We believe that our device might pave the way to a promising and innovative approach towards the eradication of *H. pylori*, alternative to pharmacologic therapy. Considering previous work that found a bacterial killing between 77% and 99.9% [7][7][8] we are confident that our device will be capable to provide an effective treatment of *H. pylori* infections. It is worth noting that, as for example stated in [7], the presence of a single treatment session with non-wireless stomach illumination methods was one of the main causes of eradication failure, due to *H. pylori* re-growth in the subsequent days. This substantial problem is overcome by the easy and frequent repeatability of our wireless capsule approach, mainly due to its minimal invasivity.

Further steps of experimentation will consist in animal tests (mini-pigs) to assess the capsule mechanical resistance, the overall safety within the whole gastrointestinal tract and the capsule permanence time in the stomach antrum in a preclinical model (i.e. with the presence of peristalsis), as indications of the overall efficacy of the capsule. The assessment of the capsule treatment efficacy in animals is not scheduled, due to substantial differences between *H. pylori* strains in animals and those in humans. For this reason, experimental tests on humans are being planned to assess the efficiency of the capsule and the minimum light dose required (i.e. the number of capsules). Clinical tests will also help understanding whether the area irradiated by the capsule is sufficient for eradication. In this perspective, it is encouraging the fact that the capsule naturally locates in the stomach antrum, where the highest bacterium concentration is found.

From an economic viewpoint, we expect that the cost for eradication with our device is possibly comparable with the one of antibiotic therapy (about 50-60 €). In a first phase our targets will be antibiotic resistant patients, where the search for innovative and effective therapeutic solutions is paramount.

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