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# Nanoparticles for water disinfection by photocatalysis: A review

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**Keywords:** Nanoparticles, photocatalysis, water and wastewater disinfection

**Abstract:** Photocatalysis is an efficient and ecological method of water and wastewater disinfection. During the process, various microorganisms are deactivated, including Gram-positive and Gram-negative bacteria, for example *Escherichia coli*, *Staphylococcus aureus*, *Streptococcus pneumoniae*, and so on, fungi like *Aspergillus niger*, *Fusarium graminearum*, algae (*Tetraselmis suecica*, *Amphidinium carterae*, and so on) and viruses. Titanium dioxide (TiO<sub>2</sub>) is the most commonly used material due to its price and high oxidation efficiency; it is easy to modify using both physical and chemical methods, what allows for its wide use in industrial scale. Intensive research on novel photocatalysts (e.g. ZnO and carbon based photocatalysis like graphene, carbon nanotube, carbon nitride and others) has been carried out. The future development of nano-disinfection containing metal/metal oxides and carbon based nanoparticles should focus on:

- improving disinfection efficiency through different manufacturing strategies,
- proper clarification and understanding of the role and mechanism of interaction of the nano-material with the microorganisms,
- progress in scaling up the production of commercial nano-photocatalysts,
- determination of the extent of environmental release of nano-photocatalysts and their toxicity.

## Introduction

The access to fresh and sanitary safe water is a fundamental of sustainable development. However, it is estimated that more than 1/6 of human population suffers because of poor water quality, which leads to death to millions of people, especially children (Bodzek 2019). Due to these facts, novel methods of water treatment, targeted at the removal and decomposition of not only chemical pollutants, like organic compound and heavy metals, but also enabling elimination of microorganisms are sought for (Bodzek et al. 2019, Bodzek 2019, Feng and Astruc 2020, Reddy et al. 2016). Disinfection is found to be a principle technological action of every water treatment system. It is also said to be one of the most difficult and complicated operations, regardless of the scale of water treatment plant (Collivignarelli et al. 2018). In the case of treatment of water dedicated to potable purposes, disinfection should assure both, the production of microbiologically safe water and maintenance of its quality during transport, including prevention of secondary biological contamination of water in pipelines (Bodzek et al. 2019). In order to assure its biological safety, potable water is disinfected. The disinfection treatments are divided into conventional, advanced and natural processes (Bodzek et al. 2019, Collivignarelli et al. 2018). The conventional technologies include chlorine, chlorine dioxide,

ozone, peracetic acid and ultraviolet (UV) radiation. The advanced technologies include the combination of ozone and hydrogen peroxide, ozone and UV radiation, hydrogen peroxide and UV radiation, UV radiation with titanium dioxide, and membranes technologies (Bodzek et al. 2019, Collivignarelli et al. 2018). These techniques are found to be very efficient, but most of them require the use of either expensive chemicals or expensive devices for on-site disinfectants generation, e.g., in the case of chlorine dioxide or ozone. Moreover, many chemical disinfectants may lead to the formation of other harmful disinfection byproducts (DBPs), like bromates and brominated DBPs in the case of waters with elevated bromides content (Bodzek and Konieczny 2011).

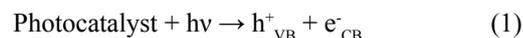
The use of photocatalysts as potential antimicrobial materials has been investigated since the 1990s. The research has covered conventional and novel materials, which have been evaluated in regard to their antibacterial efficiency (Feng and Astruc 2020, Molinari et al. 2017, Reddy et al. 2016). Photocatalysis has become widely used in order to overcome a range of environmental challenges faced by modern societies (Feng and Astruc 2020, Molinari et al. 2017, Reddy et al. 2016, Qu et al. 2013). The use of visible light for degradation or complete mineralization of contaminants is currently one of the most desired green features of the process (Feng and Astruc 2020). Despite a significant technological

progress observed within the last decade, there still exist a number of limitations, which require urgent actions. They cover modification of energy bandgap of photocatalysts, and development of novel photocatalyst characterized with narrow bandgap (Bodzek et al. 2021, Chong et al. 2010, Dalrymple et al. 2010, Bora and Dutta 2014). One option is to use nanosized particles (1–100 nm) as they reveal excellent photocatalytic potential due to their unique physico-chemical properties (Feng and Astruc 2020, Petronella et al. 2016, Bodzek and Rajca 2012, Chong et al. 2010, Dalrymple et al. 2010). Nanoparticles (NPs) are currently being viewed as a powerful nanotechnology to control hazardous microorganisms due to their intrinsic antimicrobial properties. More recently, several natural and engineered nanomaterials have also been shown to have strong antimicrobial properties. Unlike conventional chemical disinfectants, these antimicrobial nanomaterials are not strong oxidants and are relatively inert in water. Therefore, they are not expected to produce harmful DBPs. If properly incorporated into treatment processes, they have the potential to replace or enhance conventional disinfection methods. A large number of synthetic NPs have been explored for their antimicrobial properties. The most important nanomaterials that are commonly used for antibacterial activity in the water and food industry are oxides of zinc (Zn) and titanium (Ti) as well as carbon based nanoparticles (Amin et al. 2014). Titanium dioxide (TiO<sub>2</sub>) and zinc oxide (ZnO) nanoparticles are known to be one type of inorganic multifunctional substances that are able to inhibit the growth of microbes and they have been listed as Generally Recognized As Safe (GRAS) by the U.S. FDA (Zambrano-Zaragoza et al. 2018).

Photocatalytic inactivation of microorganisms is a complex process and its efficiency depends on a range of factors such as type of microorganisms, their concentration and physiological stage (Ganguly et al. 2018). Moreover, type, morphology, concentration and phase (suspended or immobilized) of photocatalyst are crucial for efficient process performance (Ganguly et al. 2018). Among different microorganisms, *Escherichia coli* (*E. coli*) is the most often used in testing and optimization of photocatalysis parameters as well as in designing of novel photoreactors (Byrne et al. 2018, Ganguly et al. 2018). However, other microorganisms like *Pseudomonas aeruginosa*, *Salmonella typhimurium* and *Enterobacter cloacae* have been used in research on their photocatalytic deactivation in order to prove the suitability of the process toward the removal of biologically active microbial species.

Photocatalysis process is defined as chemical reaction rate or the initiation of a chemical reaction under ultraviolet or visible light radiation at the presence of a photocatalyst (Molinari et al. 2017, Reddy et al. 2016, Bodzek and Rajca 2012). Homogeneous catalysis takes place when the catalyst and other reactants are all dissolved in the same solution. Heterogeneous catalysis typically involves the use of a catalyst that is insoluble, or perhaps only weakly soluble, in the solution in which the reaction takes place. Thus, in heterogeneous catalysis, the catalyst and solution may form a suspension, or the catalyst may simply be a solid that is placed in the solution. In the process, a light radiation is involved in initiation of photocatalytic reaction by bombarding of a photocatalyst surface with photons (Bodzek and Rajca 2012, Pelaez et al. 2012, Byrne et al. 2018). The radiation with a light of

a compatible wavelength (higher than the energy of band gap of a photocatalytic material) of a photocatalyst surface excites electrons (e<sup>-</sup>), which are present in its “conduction band” (Fig. 1) (Reddy et al. 2016, Qu et al. 2013, Bodzek and Rajca 2012, Bora and Dutta 2014). When e<sup>-</sup> leaves a valence band and is transferred to the conduction band (e<sup>-</sup><sub>CB</sub>), the positively charged hole (h<sup>+</sup><sub>VB</sub>) is formed (eq. 1 and Fig. 1) (Qu et al. 2013, Ganguly et al. 2018).

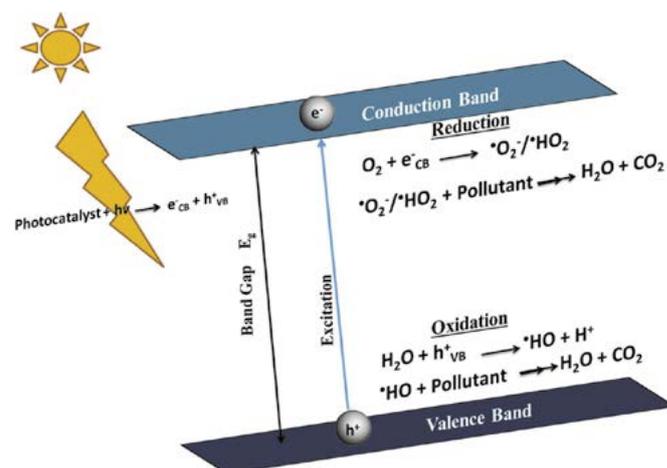


In the conduction band (e<sup>-</sup><sub>CB</sub>), e<sup>-</sup> reacts with oxygen (O<sub>2</sub>), what leads to a formation of superoxide anion (\*O<sub>2</sub><sup>-</sup>) or hydroperoxyl (\*OH<sub>2</sub>) radicals (Fig. 1 and eq. 2) (Bodzek and Rajca 2012, Chong et al. 2010, Dalrymple et al. 2010). Simultaneously, water is oxidized at positively charged hole (h<sup>+</sup><sub>VB</sub>) (Chong et al. 2010, Dalrymple et al. 2010). As a result of this reaction hydroxyl radicals (\*OH) and hydrogen ions (H<sup>+</sup>) are formed (eq. 3) (Chong et al. 2010, Dalrymple et al. 2010). Hence, pollutants present in a solution are degraded by formed radicals, called also reactive oxygen species (ROS), to water (H<sub>2</sub>O) and carbon dioxide (CO<sub>2</sub>) (Reddy et al. 2016, Chong et al. 2010, Dalrymple et al. 2010).



Despite the fact that the oxidation mechanisms can differ depending on type of photo-material and investigated pollutants, the degradation usually relies on redox reactions occurring between electrons and electron holes (Ganguly et al. 2018). Photocatalysis process starts on a surface of a semiconductor as only it absorbs photons (Bodzek and Rajca 2012), what consequently leads to the activation of valence band electrons and their transfer to the conduction band if the delivered energy is higher than the energy of band gap.

Initially ultraviolet light has been found to be most effective for photocatalysis process initiation, while recent research shows that photocatalysts may be activated either by visible or natural light (Pelaez et al. 2012). A number of research studies on the use of other materials than the most



**Fig. 1.** Mechanisms of photocatalysis (Bodzek and Rajca 2012, Chong et al. 2010, Dalrymple et al. 2010)

popular titanium dioxide ( $\text{TiO}_2$ ) are also carried out. Most of these initially involved research efforts have been focused on photo-catalytic water splitting, and then on a range of other applications such as a technique for water and wastewater treatment as well as air or exhaust gas cleaning (Byrne et al. 2018, Ganguly et al. 2018). Due to a large amount of research studies on photo-catalysts, they have been divided into two categories: oxide and non-oxide materials. Moreover, it has been observed that these groups differ also in the impact of pH change on conduction and valence bands (Byrne et al. 2018, Ganguly et al. 2016).

Modification of photocatalysts is an important aspect of practical use of photocatalysis in water and wastewater treatment, but also in other technical fields. The wavelength required to activation of most of photocatalysts usually covers ultraviolet (UV) range (300–400 nm) and it is delivered to the system by UV lamps or as a UV light present in sunlight. It results from high energy of bandgap of many photocatalyst, which for example in the case of  $\text{TiO}_2$  equals 3.2 eV. Hence, photocatalytic systems which are based of sunlight UV radiation require significant surface, what generates high operational costs (Dalrymple et al. 2010). In order to improve the absorption of light in visible range, what is especially important for photo-mineralization of pollutants present in water and wastewater, photocatalysts need to be properly modified (Anjum et al. 2019). The additional benefit resulting from the use of sunlight is the fact that it is commonly available and free. A number of successful modifications of semiconductive photocatalysts of a wide “transfer band” aimed at shift of absorption toward visible light range have been performed. However, such photocatalysts are usually characterized with lower photoactivity due to conditions favoring recombination process (Molinari et al. 2017, Chong et al. 2010). Among available modification techniques the following may be distinguished (Molinari et al. 2017, Bora and Dutta 2018):

- (a) use of different nano-techniques, such as application of nano-structural photocatalysts, modification of a surface of nano-structural catalyst by addition of dyes or organic polymers, dope of metals nanoparticles (NPs) on a catalyst surface,
- (b) addition of transient metals (manganese, copper, nickel, cobalt, etc.) and non-metals (nitrogen, sulfur, boron, halogens, etc.) to semiconductive catalyst,
- (c) combination of different semiconductive materials,
- (d) application of mesoporous supports,
- (e) induction of photocatalysis with visible light plasma.

The presented paper reviews the current state of the art on the use of nano-photocatalysis in water and wastewater disinfection. The fundamentals of process mechanism and discussion on microorganisms used in investigation of disinfection processes are also included. The results of the research studies on the use of different types of nano-photocatalysts and possible methods of their modification have been shown.

## Photocatalytic disinfection mechanism

The proper understanding of microorganisms inactivation by means of photocatalysis is crucial for definition of disinfection mechanisms. Matsunaga et al. (1985) have been the first to demonstrate an inactivation process induced with a light and

proposed that coenzyme A degradation by ROS generated during the reaction was the most probable mechanism of antibacterial action. The enzyme denaturation caused the stop of respiration activity, what led to final death of the cell (Matsunaga et al. 1985).

Further research, however, has shown that a cell’s death is caused by the damage of cellular wall or cellular membrane. The leakage of potassium ions followed by the flow of intercellular substances such as RNA and proteins leads to complete destruction of cells, what has been confirmed by transmission electron microscopy (TEM) images (Saito et al. 1992). Kikuchi et al. (1997) have observed that antibacterial activity decreases after the addition of ROS sweeper, but it is not completely stopped due to formation of highly reactive hydrogen peroxide. Hence, antimicrobial activity results from a synergic action of all types of ROS formed during photocatalytic reaction (Kikuchi et al. 1997). The progress in investigations of photocatalytic disinfection mechanism leads to understanding and description of the most probable mechanism of cellular wall damage. Jacoby et al. (1998) have finally proven the complete demineralization of damaged cells. Scanning electron microscopy (SEM) has served to observe disappearing cells colonies, while  $^{14}\text{C}$  radioisotope labelling has been used to analyze carbon content (Jacoby et al. 1998). The authors have found that peroxidation of phospholipid component of cellular wall due to the possible ROS attack results in its decay. Lipids peroxidation results in formation of malonic dialdehyde, while continuous radiation allows for aldehyde mineralization to carbon dioxide ( $\text{CO}_2$ ) and water ( $\text{H}_2\text{O}$ ) (Bagchi et al. 1993).

To sum up, ROS are crucial in the destruction of microorganisms using semiconductive materials and UV radiation, whereas the strongest effect is assigned to hydroxyl radicals ( $\cdot\text{OH}$ ) (Ganguly et al. 2018, Bogdan et al. 2014). The inactivating properties of hydroxyl radicals lie in high oxidation potential and non-specific reactivity (Reddy et al. 2007). Hydroxyl radicals formed on nano- $\text{TiO}_2$  surface, during their contact with water, are characterized with long lifespan and may penetrate cellular wall and damage DNA, similarly as  $\text{H}_2\text{O}_2$ , as well as cause peroxidation of lipids (Fig. 2) (Bodzek and Rajca 2012, Bogdan et al. 2014). Photocatalytic mechanism, which leads to antimicrobial effect, starts with the destruction of cellular wall membrane, what results in leakage in intracellular substances such as RNA and proteins, followed by complete cell destruction and death (Doong et al. 2017). The leaked material is oxidized at photocatalytic sites of a semiconductor. It has been found, however, that the presence of extracellular polymeric substance (EPS) may lead to the decrease of antibacterial activity due to its competition toward ROS. Hence, it is important to remove EPS from the reaction environment in order to obtain desired disinfection effect (Davididou et al. 2017). Moreover, generated ROS are able to create the so called oxidative stress in aqueous phase what additionally enhances the overall disinfection effect.

## Photocatalytic disinfection with titanium dioxide

$\text{TiO}_2$  photocatalysts show high efficiency to deactivation of microorganisms which appear in water and wastewater (Saito et al. 1992). There are numerous research studies discussing

the use of TiO<sub>2</sub> nanoparticles (NPs) (e.g. Degussa (Evonik) P25 nanopowder) activated with both UV- and sunlight to deactivate bacteria (*E. coli*, *Pseudomonas aeruginosa*), fungi (*Candida albicans*, *Fusarium solani*), protozoa (trophozoite stage of *Acanthamoeba polyphaga*), spores (*Bacillus subtilis*) and cysts (Kikichi et al. 1997). Investigations have shown that 120 min exposition of water contaminated with *Escherichia coli* and *Lactobacillus acidophilus* to radiation of UV range (315–400 nm) resulted in almost complete destruction of bacteria (Matsunaga et al. 1985).

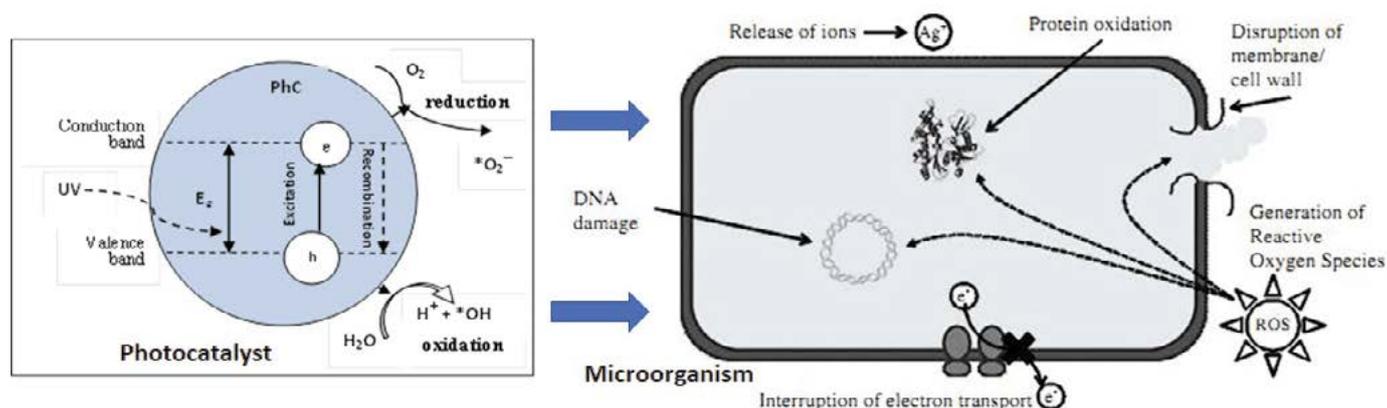
Antibacterial effect of TiO<sub>2</sub>/UV process has been confirmed by many laboratory research studies with use of a number of Gram-negative bacteria, i.e., *Escherichia coli*, *Klebsiella pneumoniae*, *Salmonella enteritidis*, *Salmonella typhimurium*, *Serratia marcescens*, *Shigella flexnerii*, *Legionella pneumophila*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Vibrio cholerae*, and Gram-positive bacteria, i.e., *Bacillus anthracis*, *Bacillus cereus*, *Bacillus pumilis*, *Bacillus subtilis*, *Listeria monocytogenes*, *Staphylococcus aureus*, *Clostridium perfringens*, *Lactobacillus acidophilus*, *Lactobacillus helveticus*, *Enterococcus faecalis*, and *Enterococcus faecium* (Bogdan et al. 2014). According to the authors (Bogdan et al. 2014, Blanco-Galvez et al. 2007), the sensitivity of bacteria to TiO<sub>2</sub>/UV process can be arranged in the following series: *Escherichia coli* > Gram-negative bacteria (other than *E. coli*) > Gram-positive bacteria (other than *Enterococcus sp.*) > *Enterococcus sp.* This sequence is confirmed also by Kuhn et al. (2003) who have investigated efficiency of inactivation of 4 bacteria species using UV-A 1 minute radiation and obtained the following sensitivity series: *Escherichia coli* > *Pseudomonas aeruginosa* > *Staphylococcus aureus* > *Enterococcus faecium*. The differences in affinity of particular microorganisms to photocatalytic inactivation are found to result from different structures of cellular walls and various susceptibility to oxidation.

Antibacterial effect of nanocatalysis depends on the activity and concentration of the photocatalyst, size of its particles, presence of other environmental contaminants, initial concentration of microorganisms, solution pH and temperature as well as light intensity (Friedmann et al. 2010). Maness et al. (1999) have shown that the highest photocatalytic activity, and hence the strongest antimicrobial effect, is revealed by two polymorphic nano-TiO<sub>2</sub> forms – anatase and rutile, which are

mixed at the ratio of 4:1 (trade name P25), and commonly used in research on disinfection properties of nano-TiO<sub>2</sub>. Regardless of initial *Escherichia coli* cells concentration (10<sup>2</sup>–10<sup>8</sup> CFU/mL) the highest inactivation can be obtained at 0.1 wt.% P25 (Maness et al. 1999). Etacheri et al. (2010, 2013) have found that hetero-bonds between anatase and brookite TiO<sub>2</sub> forms doped with carbon at the ratio of 80/20 assure sufficient inactivation of *S. aureus* under visible light radiation. The significant narrowing of bandgap is in this case caused by additional energy levels formed by carbonates, what results in excellent photocatalytic and antibacterial activity.

Rincon and Pulgarin (2003) have stated that nano-TiO<sub>2</sub> of diameter below 20 nm reveals better antibacterial properties than its less powdered form, because particles of diameter <20 nm are able to penetrate cellular wall and permeate to cytosol (Desai and Kowshlik 2009), whereas TiO<sub>2</sub> nanoparticles of size 20–80 nm are not able to pass through the wall (Wong et al. 2006). Studies have shown that the presence of organic contaminants may decrease the efficiency of TiO<sub>2</sub>/UV bacteria inactivation even up to 40% (Belapurkar et al. 2006). Other research studies indicate that photocatalytic effect of nano-TiO<sub>2</sub> is also related with high microorganisms concentration (above 10<sup>8</sup> CFU/mL) (Maness et al. 1999).

The use of TiO<sub>2</sub> in **powdered form** is, however, related to one significant disadvantage, which is the separation of nanoparticles from treated stream. In order to minimize this effect, the efficient antibacterial activity is obtained by immobilization of nanoparticles on relevant supports (Liu et al. 2008). Second solution is the application of the **TiO<sub>2</sub> nanotubes (NTs)**. NTs structure is one of the most promising morphologies for antimicrobial applications due to a range of desired features, such as high aspect ratio, enlarged specific surface area and improved light harvesting and absorption (Byrne et al. 2018, Podporska-Carroll et al. 2015, Garvey et al. 2016). Recently, the novel capacitive method of synthesis of very long nanotubes of high aspect ratio (of length 100 μm and diameter 20 nm) based on a simple electrochemical process involving chlorine (Richter et al. 2007) has been elaborated. The obtained, highly porous nanotubes can be directly involved in antimicrobial applications. Antibacterial properties investigations of TiO<sub>2</sub> nanotubes using *E. coli* and *S. Aureus* have been found highly efficient to inactivate both examined species, at removal rates obtained during



**Fig. 2.** The mechanism of destruction of microorganism cell by reactive oxygen species (ROS) (Bodzek and Rajca 2012, Bogdan et al. 2014)

24 hours UV radiation amounted to 97.53 and 99.94% for *E. coli* and *S. aureus*, respectively (Podporska-Carroll et al. 2015). Research on inactivation of *E. coli*, *P. aeruginosa* and *S. aureus* of TiO<sub>2</sub> nanotubes has also been performed (Garvey et al. 2016). Wang, et al. (2021) have developed a UV-assisted electrochemical oxidation (UV-EO) process that employs blue TiO<sub>2</sub> nanotube arrays (BNTAs) as photoanodes. Inactivation of tetracycline- and sulfamethoxazole-resistant *E. coli* along with degradation of the corresponding plasmid coded genes (tetA and sul1) is measured by plate counting on selective agar and qPCR, respectively. In comparison to UV irradiation alone, enhanced antibiotic resistant bacteria (ARB) and antibiotic resistance genes inactivation and ARG degradation is achieved by UV-EO. Chloride significantly promotes the inactivation efficiency due to the electrochemical production of free chlorine and the subsequent UV/chlorine photoreactions.

Studies have shown that the increase of calcination temperature and rutile content in TiO<sub>2</sub> NTs results in poorer disinfection effect. The photocatalyst, which has contained the highest percentage share of rutile and has been calcined at 800°C has been the least toxic to microorganisms. In regard to temperature, it has been also shown that NTs calcined at 600 and 800°C are not able to reach the inactivation coefficient higher than 1 log<sub>10</sub> (Garvey et al. 2016). On the other hand, non-calcined NTs or ones calcined at 200°C show high inactivation rate up to 2.7 log<sub>10</sub> (Garvey et al. 2016). NTs, which have been calcined at between 400 and 800°C inactivated *P. aeruginosa* in the same extent as P25, whereas the latter has been found more toxic in regard to *E. coli* than calcined NTs (Garvey et al. 2016).

**TiO<sub>2</sub> composites** have a higher inactivation efficiency against microorganisms in water and wastewater disinfection than pure titanium dioxide.

Recent studies have also shown that **Ag nanoparticles** doped TiO<sub>2</sub> reveals significantly improved inactivation of bacteria (Page et al. 2007). For example, the presence of 1wt.% of Ag in TiO<sub>2</sub> shortens UV-A radiation period required for complete removal of 10<sup>7</sup> CFU/mL of *E. coli* from 65 to 16 minutes (Reddy et al. 2007). It is assumed that Ag nanoparticles improve TiO<sub>2</sub> photoactivity by slowing the charge carriers recombination and/or by the increase of absorption surface (Sung-Suh et al. 2004). It is also supposed that Ag nanoparticles induce transfer of electrons to TiO<sub>2</sub>, what leads to charge separation and thus allows for activation of the material under visible light range (Seery et al. 2007). Hence, Ag-TiO<sub>2</sub> nanocomposites are photocatalytic nano-materials suitable to be used in processes at visible light radiation. It has been also shown that the introduction of Ag nanoparticles to **TiO<sub>2</sub> foil** (Ag/TiO<sub>2</sub>) as well as the construction of mesoporous composite foil Ag-TiO<sub>2</sub>/Ag/anatase-TiO<sub>2</sub> have antibacterial effect toward *E. coli* during photocatalysis under visible and sunlight radiation (Akhavan and Ghaderi 2009). The formed composite possess larger surface and higher number of active sites available for microorganisms degradation than TiO<sub>2</sub> without modification. Similar nanocomposite material can be obtained by means of sol-gel method by deposition of 30 nm Ag-TiO<sub>2</sub> coat on 200 nm anatase (a-TiO<sub>2</sub>) layer preliminary doped with silver nanoparticles. The antibacterial activity investigated toward *E. coli* in the dark has been 5.1 times better than observed for pure a-TiO<sub>2</sub>. Moreover, photo-antibacterial

activity of nanocomposite layer radiated with sunlight has been 1.35 and 6.90 higher than the activity of Ag/a-TiO<sub>2</sub> and a-TiO<sub>2</sub>, respectively. Both OH<sup>-</sup> bonds as well as H<sub>2</sub>O content have been concentrated on the developed foil surface and in boundary phase, what influences the release of nano-silver ions through pores of TiO<sub>2</sub>. The durability of nanocomposite foil has been at least 11 times greater than the durability of Ag/a-TiO<sub>2</sub> foil. Hence, Ag-TiO<sub>2</sub>/Ag/a-TiO<sub>2</sub> photocatalyst may be regarded as an effective and durable antibacterial, nanocomposite material.

Interesting option of nanocatalyst is the nanocomposite comprised of Ag-TiO<sub>2</sub> and hydroxyapatite (HAP) (Reddy et al. 2007). Photocatalytic properties of HAP, antibacterial Ag nature and improved disinfection effect of Ag-TiO<sub>2</sub> have all together enabled the complete inactivation of *E. coli*. Such catalysts can be obtained by co-precipitation method, while Ag-TiO<sub>2</sub> (AT1) of 1wt.% Ag content and Ag-HAP of 5wt.% Ag content using wet impregnation method. The analysis has indicated that in all prepared photocatalysts titanium appears in the form of Ti<sup>4+</sup>, while silver as Ag<sup>0</sup>, and the change of photocatalytic activity results from surface changes of PO<sub>4</sub><sup>3-</sup>. Another composite of heterostructure comprised of TiO<sub>2</sub>/Ag<sub>3</sub>PO<sub>4</sub> has also been investigated toward inactivation of *E. coli* bacteria and *F. graminearum* fungi (Liu et al. 2017). After 100 min VIS radiation, TiO<sub>2</sub>/Ag<sub>3</sub>PO<sub>4</sub> composite reduced the survivability of *F. graminearum* to ca. 2%. The mechanism of photocatalytic disinfection has been assigned to the damage of cellular cell related with the attack of reactive oxygen species, mainly hydroxyl radicals (Liu et al. 2017). The obtained material has been characterized with effective charge separation and improved visible light absorption.

Similarly, composites comprised of **AgI/TiO<sub>2</sub>** and **AgBr/TiO<sub>2</sub>** have assessed antimicrobial effect toward *E. coli* and *S. aureus* under visible light radiation ( $\lambda > 420$  nm) (Hu et al. 2007, Lan et al. 2007). The results have indicated that photocatalytic complete degradation of cellular wall is the main reason of microorganisms death, what has been shown by TEM images research, whereas degradation products such as aldehydes, ketones and carboxylic acids have been identified using FTIR spectroscopy. Moreover, electrostatic interaction between bacteria and catalyst significantly influences the disinfection efficiency basing on *E. coli* inactivation results obtained at different conditions.

In addition to silver, copper **Cu** sputtered onto **TiO<sub>2</sub> foil** also shows inactivation of bacteria (Rtimi et al. 2013). Cu and TiO<sub>2</sub>/Cu sputtered films have allowed for complete inactivation of *E. coli* in dark, while such an effect has not been observed in the case of TiO<sub>2</sub>. The presence of Cu has enhanced inactivation of bacteria under simulated sunlight radiation of low intensity, what influences the practical use of such materials. Composite layers formed during plasma sputtering allow for significant metal savings in regard to layers formed using conventional sputtering methods.

The similar, strong antibacterial effect has also been revealed by **TiO<sub>2</sub>-ZrO<sub>2</sub>** composite on polyester (PES) sputtered with **Cu**. Three times better inactivation has been obtained for Cu containing composite in comparison to material deprived of the metal, even at a very low copper content (0.01–0.02 wt.%) (Rtimi et al. 2015). Antibacterial properties are obtained due to strong interactions between phosphate and thiol group of negatively charged cellular wall and copper (a highly active

electron donor). Also, **TiO<sub>2</sub>(Eu)/CuO** nanocomposite has been formed and investigated toward its antibacterial effect against *Enterococcus* bacteria (Michalski et al. 2016). The photoactivity of the material is connected by selective CuO binding with microorganism cell.

Fisher et al. (2014) have described **molybdenum** doped titanium dioxide structures, which can be used as both antibacterial and photocatalytic agent for efficient inactivation of brewery microorganisms. Such material has revealed an inactivation rate of 5-log and 1-log toward bacteria and fungi, respectively. TiO<sub>2</sub>-Mo surface has also been active in the dark and the indicated inactivation rates have been reached within 4–24 hours in the case of bacteria and up to 72 hours in the case of fungi. The research also suggests that TiO<sub>2</sub>-Mo layer may act as a secondary barrier, which prevents accumulation of microorganisms on the surface brewery tanks and other elements of installation during long production periods.

**Boron** and/or **cerium** doped titanium dioxide has been investigated as an antibacterial agent using *Staphylococcus aureus* species (Wang et al. 2016a). The main crystalline phase of TiO<sub>2</sub> is anatase phase, while its doping with B/Ce results in small sizes of crystals, which possess large specific surface area. It has been found that B/Ce-TiO<sub>2</sub> nanomaterial is characterized with satisfactory antibacterial properties, mainly due to narrowing of bandgap of titanium dioxide, enlargement of specific surface area, presence of B partially substituting O and forming B-O-B bonds in borate ions and co-appearance of Ce<sup>3+</sup> and Ce<sup>4+</sup> ions. The results indicate that B and Ce doped TiO<sub>2</sub> deactivated microorganisms better than in the case of other materials, while the presence of both additives reveals synergic antimicrobial effect.

Stan et al. (2016) have discussed photocatalytic and antimicrobial properties of cotton woven cloths coated with dispersed TiO<sub>2</sub>-1wt.% containing **Fe-N** nanoparticles formed using hydrothermal method and secondary calcination at 400°C. The obtained composite has mainly comprised of anatase phase and smaller amount of brookite (~15%–20 wt.%) containing Fe<sup>3+</sup> ions and nitrogen. The cloths have revealed specific antibacterial properties, which depended on their composition, but also bacteria species and incubation time. The assessment of in vitro biocompatibility has confirmed lack of cytotoxicity after a short-term exposition.

Composites comprised of **nickel ferrite** and TiO<sub>2</sub> reveal magnetic and photocatalytic properties (Rana et al. 2005, Rawat et al. 2007). The magnetic core simplifies photocatalyst recovery, whereas TiO<sub>2</sub> gives photocatalytic features. The obtained composite has shown antibacterial properties against *E. Coli*. Additionally, inactivation of bacteria under UV radiation in the presence of TiO<sub>2</sub>-NiFe<sub>2</sub>O<sub>4</sub> is faster than the one observed for TiO<sub>2</sub> NPs. Chen et al. (2017) have developed three-component composite comprised of TiO<sub>2</sub>/NiFe<sub>2</sub>O<sub>4</sub>/diatomite doped with Ce/N. The hybrid catalyst has shown multifunctional properties. The presence of diatomite has improved the adsorption, Ce/N doping has positively influenced visible light absorption, while NiFe<sub>2</sub>O<sub>4</sub> has enhanced magnetic recycle of the photocatalyst. Basing on the obtained results it has been found that after 90 min of *E. coli* degradation in the dark only 24.2% can be deactivated, while photodegradation process enables up to 83.6% deactivation effect.

Also, magnetic Fe<sub>2</sub>O<sub>3</sub>-AgBr composite inactivates both Gram-negative (*E. coli*) and Gram-positive (*Staphylococcus aureus*) bacteria (Ng et al. 2016). The antibacterial effect has been favored by high temperature and alkaline pH. The examination of photocatalytic bacteria inactivation has shown that Fe<sub>2</sub>O<sub>3</sub>-AgBr material causes oxidation by H<sub>2</sub>O<sub>2</sub> generated using photogenerated electron as well as by direct contact with photogenerated electron gap. The antibacterial properties have been stable within 5 proceeding recycles, hence the material possesses high potential in regard to its use in water disinfection.

Rengifo-Herrera et al. (2009, 2010) have performed wide research on doping **nitrogen** and **sulphur** compounds N-S to TiO<sub>2</sub> and the impact of semiconductor modification on *E. coli* photocatalytic inactivation. Thiourea was used as the source of N-S dopes to TiO<sub>2</sub> (Rengifo-Herrera et al. 2009). After calcination at the temperature of 400 and 500°C different types of catalysts can be obtained (Fagan et al. 2016). The authors suggest that hydroxyl radicals probably do not directly participate in oxidation processes, which occur during N, S doped Degussa (Evonik) P25 radiation with visible light. Reactive oxygen radicals (ROS) have been found to depend on dope properties, particles size and surface hydroxylation (Fagan et al. 2016, Rengifo-Herrera et al. 2009, 2010).

Helali et al. (2014) studied photocatalytic and photolytic disinfection of *Escherichia coli* in water under natural sunlight using different types of photocatalyst (TiO<sub>2</sub> P-25, PC500, Ruana and Bi<sub>2</sub>WO<sub>6</sub>) at different concentrations. The solar photo-inactivation yielded complete inactivation results, which varied with the solar light intensity. Meanwhile, dark control samples in the lab (temperature constant at 25°C) remained at constant concentration and dark samples outside laboratory showed a decrease due to the mild solar heating occurring during the experiments.

## Photocatalytic disinfection with zinc oxide

Zinc oxide is a nanomaterial, which in recent years has been widely investigated as a potential antibacterial agent, mainly because of its high activity at relatively low concentrations (Dimapilis et al. 2018). It keeps its stability in severe environmental conditions, while it is harmless to animals and humans (Dimapilis et al. 2018). ZnO has been investigated in regard to deactivation of variety of microorganisms in order to sufficiently establish its antibacterial potential.

Investigations on antibacterial effect of ZnO, using different bacteria species including both Gram-positive (e.g. *Sarcinic pseudomycosis*) and Gram-negative (e.g. *E. coli*) microorganisms, show that efficiency is better for Gram-positive than for Gram-negative bacteria (Dimapilis et al. 2018, Tayel et al. 2011, Zhang et al. 2011, Jones et al. 2008). It has been found that it is due to the simpler structure of cellular wall. It has also been stated that thick cellular wall of Gram-negative bacteria may prevent permeation of ZnO to the interior cell and thus disable its further interaction with intracellular substances. Among the investigated bacteria species, *Bacillus cereus* has been found to be the most sensitive to ZnO, while *Pseudomonas spp.* is recognized as the most resistant. On the other hand, 2 hour exposition of *S. aureus* to ZnO nanoparticles results in cells destruction or lysis, while the complete disappearance is obtained after 4 hours.

Jones et al. (2008) have suggested that despite bacteria type, nanoparticles size may be more important considering inactivation mechanism due to possible accumulation of NPs inside cellular membrane. Yamamoto (2001) has shown that the impact of ZnO nanoparticles size was poorer in regard to *S. aureus* than in the case of *E. coli*. It has been probably caused by the difference in structure and chemical composition of cellular components. In *E. coli* wall, peptidoglycan, lipid and lipopolysaccharide layers can be found on cell surface, whereas in *S. aureus* only peptidoglycan layer can be detected.

He et al. (2011) have studied the effect of ZnO concentrations on the deactivation of two pathogenic fungi, i.e., *Botrytis cinerea* and *Penicillium expansum*. The obtained results have shown that concentration higher than 3 mM significantly inhibits the growth of fungi and the effect improves with fungi concentration increase. *P. expansum* is found to be more sensitive than *B. cinerea* to ZnO action, what results from the differences in growth morphology. Moreover, the results suggest that the inhibitive mechanism of ZnO is different against fungi than the one against bacteria.

Photo-catalytic deactivation of Gram-negative *E. coli* and Gram-positive *Bacillus subtilis* bacteria present in water using **ZnO nanorods** under visible light radiation has been twice higher than their inactivation observed in the dark conditions (Bhadra et al. 2011, Eskandari et al. 2011). The degradation of both cellular wall and DNA has been obtained. The high efficiency against pathogenic microorganisms revealed by ZnO nanorods at the presence of visible light allows for their potential ex situ use in water decontamination carried out at ambient conditions. Moreover, ZnO nanorods immobilized on cellulose paper also reveal antibacterial properties under visible light radiation (Eskandari et al. 2011).

He et al. (2013) have elaborated immobilization of Au nanoparticles on ZnO using photoreduction method. The relatively small share of Au significantly improves antibacterial features of the material. The increase in the amount of ROS, which are generated during the process, enhances the overall efficiency of the disinfection. Other research studies have been focused on the inactivation of Gram-negative *Vibrio cholera* 569B bacteria present in water using sunlight activated photocatalysis on nanocomposite particles of core-coat structure of Ag@ZnO type (Das et al. 2015). Silver nanoparticles have been obtained via the reduction of silver perchlorate followed by precipitation on ZnO coat. The final material has been applied to photocatalytic deactivation of the model pathogen and the impact of photocatalyst dose and reaction temperature on process kinetics have been checked. The efficiency of the process has been established for deionized and actual water matrices and compared with pure ZnO and commercial TiO<sub>2</sub> (Degussa (Evonik) P25). The nanocomposite system has shown the optimal disinfection (ca. 98%) during 40–80 minute exposure to sunlight and at the catalyst load 0.5 mg/L. Hence, the reduction of aquatic bacteria by photocatalytically active Ag@ZnO nanocomposite under sunlight radiation may be potentially applied to water decontamination at ambient conditions.

Elmi et al. (2014) have investigated antibacterial activity of ZnO nanoparticles (NPs) **using municipal wastewater** containing *K. pneumoniae*, *E. coli*, *Proteus* and *Staphylococcus epidermidis*. The antibacterial effect of ZnO

has been compared with conventional disinfection methods using chlorine and UV radiation. The research has shown that the antimicrobial activity of nanoparticles depends on the type of bacteria and nanoparticles concentration. The disinfection effect can be observed in regard to *E. coli* and *S. epidermidis*, whereas there is no inhibition observed to *K. pneumoniae* and *Proteus*. The antibacterial efficiency increases with ZnO nanoparticles concentration increase. In general, the experiments indicate that ZnO can be a potentially useful agent in wastewater disinfection, especially as a method supporting UV disinfection.

Nanostructured ZnO photocatalysts immobilized on cellulose and polyester supports have also been developed in order to improve **water disinfection** carried out with the use of sunlight (solar disinfection – SODIS) (Danwittayakul et al. 2020). Research on disinfection using UVA has been performed using 200 mL transparent polyethylene bags acting as SODIS reactors, which contained ZnO catalyst and water contaminated with 106 CFU *Escherichia coli*. The photocatalyst placed on polyester support has revealed 15% higher efficiency than the one immobilized on cellulose support (Danwittayakul et al. 2020). The on-site SODIS test operated with the use of natural sunlight has also been performed and involved the use of dedicated SODIS reactors equipped with ZnO immobilized in polyester support. Almost complete disinfection effect (97–98% efficiency) have been reached within 15 minutes of the test run (Danwittayakul et al. 2020). The treated water has contained below 2 mg/L of zinc released from the photocatalytic material.

Other tests have involved ZnO immobilized on Si plates and such material has shown excellent antibacterial effect against *E. coli* above 3 log<sub>10</sub> CFU/mL under double UV radiation (i.e. UV-A and UV-C) (Jin et al. 2019). Moreover, immobilized ZnO nanoparticles have been much more effective than the suspended form of the photocatalyst. Among main deactivation mechanisms one can distinguish membrane sorption and ROS generation without Zn ions release.

In Tab. 1 the summary of research on antibacterial activity of ZnO revealed against different bacteria species is presented.

## Carbon based photocatalysts in water and wastewater disinfection

Among important photocatalysts used in water disinfection are those that are based on coal. There can be distinguished here composites based on TiO<sub>2</sub> and other materials by their combination with activated carbon, graphene, carbon nanotubes, fullerenes and graphitic carbon nitride g-C<sub>3</sub>N<sub>4</sub>. This group of photocatalysts has also gained attention regarding their use in photocatalytic disinfection processes. Recently, there have been reports on the use of photocatalysis, specifically carbon nanomaterials, to inactivate viruses, including the SARS-CoV-2 virus (Nasir et al. 2021, Sengupta and Hussain 2021). SARS-CoV-2 belongs to the RNA virus family and its spread has created a medical emergency worldwide because of its deadly nature and rapid transmission rate. To combat the present situation various carbon nanomaterials, such as fullerene, CNT, graphene and GO can be employed as antiviral substances because of their capability of inhibiting RNA type virus (Sengupta and Hussain 2021), biocompatibility and low

toxicity (Sengupta and Hussain 2021). Thus, it can be believed that these unique carbon nanomaterials will pave the way to combat the fatal SARS-CoV-2 in near future.

Composites comprised of titanium dioxide and **graphene oxide** (TiO<sub>2</sub>-GO) exposed to visible light radiation have been most frequently investigated in their possible use against *E. coli* and *F. solani*, and the results compared with Degussa (Evonik) P25 (standard nanoparticles of TiO<sub>2</sub> with mixed anatase and rutile phases) (Cao et al. 2013, Fernández-Ibáñez et al. 2015). The composite has shown faster disinfection effect than one obtained for commercial photocatalyst and the reason of this fact has been related to the formation of singlet oxygen in reaction mixture. Liu et al. (2011) have also prepared graphene oxide-TiO<sub>2</sub> composite, which has shown better inactivation of *E. coli* in comparison to titanium dioxide nanowires. This phenomenon has been related to the cooperation of 2 dimensional graphene sheets deposited on large surface of TiO<sub>2</sub> nanowires. Also nanocomposites comprised of TiO<sub>2</sub> and reduced graphene oxide show improved antimicrobial properties under sunlight radiation (Akhavan and Ghaderi 2009). This activity has been assigned to better light absorption or acceptance of electrons revealed by reduced graphene oxide, which sufficiently participates in charge separation and delays recombination process. Three component composites of nano-TiO<sub>2</sub>, carbon dots (C-dots) and reduced graphene oxide (rGO) have been investigated for disinfection properties in dark as well as exposed to simulated and natural sunlight (Zeng et al. 2017). The research has shown

that in the dark and under simulated sunlight *E. coli* has not been inactivated in sufficient way, whereas its exposition to natural light has resulted in 1.03 log and 0.58 log inactivation for TiO<sub>2</sub>/rGO (TR) and C-dots/TiO<sub>2</sub>/rGO (CTR) respectively, after 60 minutes of radiation. The mechanism of the process is based on transfer of electrons from TiO<sub>2</sub> to carbon dots using rGO sheets. Electrons captured by C-dots are next transferred to O<sub>2</sub> molecules and superoxyl radicals \*O<sub>2</sub><sup>-</sup> are formed and they are responsible for destruction of cellular wall. The addition of C-dots improved charge separation and slowed the recombination down, what in consequence improved overall antibacterial effect.

ZnO/graphene oxide nanoparticles have excellent antibacterial activity against *E. coli*, *Salmonella typhimurium*, *Bacillus subtilis* and *Enterococcus faecalis* (Raizada et al. 2019). The mechanism of antibacterial action has been based on the large amount of reactive oxygen species generated on the surface of the developed nanocomposites. Wu et al. (2015) have also used nanocomposite photocatalyst comprised of graphene oxide/zinc oxide (GO-ZnO) toward inactivation of *Escherichia coli* K-12. The obtained results have indicated that GO-ZnO nanocomposite has possessed much improved antibacterial features in comparison to separately used ZnO and GO and, moreover, it has revealed strong interactions between ZnO and GO in the composite structure.

In Tab. 2 the selected results on bacteria inactivation using bi- and three component photocatalysts containing graphene oxide in their structure are presented.

**Table 1.** ZnO antibacterial activity against different bacteria species and at different ZnO concentrations.

Bacteria species	ZnO concentration	Antibacterial test results	Reference
<i>Staphylococcus aureus</i>	0.1–0.3 µL	ZOI, mm: 1.1 µL-10 mm, 1.2 µL-13 mm, 1.3 µL-11 mm	Menaka et al. 2016
<i>Escherichia coli</i>	5–100 mM	Efficient inhibition at investigated concentration range. No inhibitive effect observed for concentration below 1 mM.	Padmavathy and Vijayaraghavan 2008
<i>Escherichia coli</i> <i>Bacillus subtilis</i>	0.1–2.0 wt.%	At 2 wt.% dose complete inhibition of both bacteria species is achieved. Below 2 wt.% <i>B. subtilis</i> is more sensitive than <i>E. coli</i> .	El Saeed et al. 2015
<i>Staphylococcus aureus</i> <i>Salmonella typhimurium</i>	20–100 µg/mL	MIC for both bacteria ca. 40 µg/mL. Inhibitive effect increase with concentration increase.	Navale et al. 2015
<i>Escherichia coli</i> <i>Bacillus cereus</i> <i>Pseudomonas aeruginosa</i>	1 M ZnO	ZOI, mm (6 mm disc diameter): <i>E. coli</i> ca. 21, <i>B. cereus</i> ca. 34, <i>P. aeruginosa</i> ca. 17	Tayel et al. 2011
<i>Escherichia coli</i> <i>Klebsiella pneumoniae</i> <i>Staphylococcus epidermidis</i>	0.01–50 mM	ZOI, mm at 25 mM: (5 mm diameter disc): <i>E. coli</i> ok. 12, <i>S. epidermidis</i> ok. 18, <i>K. pneumoniae</i> ok. 0	Elmi et al. 2014
<i>Staphylococcus aureus</i> <i>Escherichia coli</i> <i>Klebsiella pneumoniae</i> <i>Enterococcus faecalis</i> <i>Pseudomonas aeruginosa</i>	(20–100) µg/mL	ZOI, mm at 20 µg/mL (6 mm diameter well): <i>S. aureus</i> ca. 18, <i>E. coli</i> ca. 13, <i>K. pneumoniae</i> ca. 10 <i>E. faecalis</i> ca. 10, <i>P. aeruginosa</i> ca. 25	Narayanan et al. 2012
<i>Escherichia coli</i> <i>Pseudomonas aeruginosa</i> <i>Staphylococcus aureus</i> <i>Bacillus subtilis</i>	0.02925–30 mg/mL	ZOI, mm at 0.938 mg/mL: <i>E. coli</i> ca. 22, <i>P. aeruginosa</i> ca. 24, <i>S. aureus</i> ca. 18, <i>B. subtilis</i> ca. 15	Elkady et al. 2015

ZOI – zone of inhibition; MIC – minimum inhibitor concentration

**Table 2.** Graphene oxide based photocatalysts in water disinfection

Photocatalytic material	Light source	Characteristics	Reference
rGO@ film TiO <sub>2</sub>	Sunlight	100% <i>E. coli</i> inactivation.	Akhavan and Ghaderi 2009
GO-TiO <sub>2</sub> nanowires	Sunlight	100% <i>E. coli</i> inactivation.	Liu et al. 2011
GO-ZnO-Ag	Visible	99.99% <i>E. coli</i> inactivation.	Gao et al. 2013a
GO-CdS	Visible	100% <i>E. coli</i> and <i>B. subtilis</i> inactivation.	Gao et al. 2013a
rGO-C <sub>3</sub> N <sub>2</sub> -S	Visible	100 % <i>E. coli</i> inactivation.	Wang et al. 2013
TiO <sub>2</sub> - GO	UV and sunlight	Enhanced and faster photodegradation under both UV and sunlight radiation.	Gao et al. 2014
TiO <sub>2</sub> -C dots/rGO	Sunlight	1.03 log inactivation of <i>E. coli</i> after 60 min.	Zeng et al. 2017

**Carbon nanotubes** (CNTs) are promising antibacterial agents, which can be used against *Escherichia coli*, *Micrococcus lysodeikticus*, *Streptococcus mutans*, *Salmonella spp.* and other microorganisms appearing in water (Kang et al. 2009, Akasaka and Watari 2009, Liu et al. 2009). CNTs cytotoxicity toward bacteria present in water is a complex function of solution chemistry, transport behaviors and physico-chemical properties of nanomaterials (Kang et al. 2009). Additionally, functionalization of CNTs leads to the improvement of antibacterial features by enhancement of their dispersion ability and stability (Pasquini et al. 2012). Another potential application of CNTs in water disinfection is their use in synthesis or in situ formation of membranes used for the elimination of bacteria and viruses from treated water (Brady-Estévez et al. 2010). Several research studies have shown that CNTs can be combined with polymers and nanomaterials for preparation of nanocomposite membranes further involved in efficient water and wastewater disinfection.

Nanocrystalline TiO<sub>2</sub> and CNTs are important functional materials which in recent years have been used in water disinfection showing *E. coli* inactivation under visible light radiation (Akhavan et al. 2009). Frequently, MWCNTs have been coated with TiO<sub>2</sub> using immersion sol-gel method followed by calcination at 400°C, which led to crystallization of TiO<sub>2</sub> coat and formation of Ti-C and Ti-O-C bonds within the obtained composite. Photoinactivation of bacteria induced with visible light has increased in the following series: MWCNTs < TiO<sub>2</sub> < TiO<sub>2</sub>/MWCNTs. Koli et al. (2016a) have also investigated the efficiency of disinfection of TiO<sub>2</sub>/MWCNTs toward *E. coli* and *S. aureus* under visible light radiation. The composite has revealed faster disinfection and better cell viability in comparison to pure TiO<sub>2</sub> due to more efficient charge separation enabled by MWCNTs.

Nanocomposites comprised of Fe doped TiO<sub>2</sub> and MWCNTs (0.1–0.5 wt.%) prepared using sol-gel method reveal photoactivity against gram-positive *Bacillus subtilis* and Gram-negative *Pseudomonas aeruginosa* bacteria (Koli et al. 2016b). The best antimicrobial effect is in the case of the nanocomposite containing 0.5 wt.% of MWCNTs in comparison to other materials. Photoluminescence studies have shown that Fe-TiO<sub>2</sub>-MWCNTs nanocomposites are able to generate higher amounts of reactive oxygen species than other nanocomposites.

Nanocomposites comprised of **fullerene** and titanium dioxide (C<sub>70</sub>-TiO<sub>2</sub>) show also antibacterial features under

exposure to visible light (Oujang et al. 2016). Disinfection experiments have shown that 73% of *E. coli* cells have died within 2 hours, which is three times higher than one revealed by bare TiO<sub>2</sub>. The obtained results have indicated that the mechanism of cells inactivation is related to hydroxyl radicals, which play an important role in photocatalytic initiation of cell's death. Bai et al. (2012) have described the preparation of transparent nano-TiO<sub>2</sub> coats containing polyhydroxy-fullerene (PHF) of improved antimicrobial potential. The material has been applied to photocatalytic inactivation of spores of *Aspergillus niger* (*A. niger*), a fungus which is recognized to cause asthma. The developed nanocomposite has been able to eliminate spores three times faster than the coat comprised only of TiO<sub>2</sub>. The changes in morphology of *A. niger* spores caused by photocatalysis have been examined using SEM.

The novel photocatalyst is based on graphitic **carbon nitride** g-C<sub>3</sub>N<sub>4</sub>, which is characterized with efficient disinfection activity due to a large adsorption surface, suitable bandgap energy and high chemical and thermal stability and 2D structure. It is an important photocatalytic material widely applied in environmental protection to deactivate many toxic contaminants, including microorganisms and harmful pathogens (Hao et al. 2016, Belver et al. 2019). The mechanism of disinfection is mainly based on the generation of reactive oxygen species (Murugesan et al. 2019). Different strategies dedicated to improvement of disinfection features of g-C<sub>3</sub>N<sub>4</sub> include: (1) modification of g-C<sub>3</sub>N<sub>4</sub> texture and morphology; (2) doping g-C<sub>3</sub>N<sub>4</sub> with cations (including noble metals: gold (Au), platinum (Pt) and silver (Ag) and transient metals: copper (Cu), nickel (Ni), iron (Fe) and tin (Sn)) and non-metals (sulphur (S), carbon (C), fluorine (F), boron (B) and phosphorus (P)); and (3) combination of g-C<sub>3</sub>N<sub>4</sub> with suitable semiconductive materials (e.g. TiO<sub>2</sub>, ZnO, CuO, Cu<sub>2</sub>O, BiOCl, BiVO<sub>4</sub>, Bi<sub>2</sub>MoO<sub>6</sub>, Bi<sub>2</sub>WO<sub>6</sub>, In<sub>2</sub>S<sub>3</sub>, SrTiO<sub>3</sub>, WO<sub>3</sub>, AgX (X = Br, Cl and I) and GO) (Hao et al. 2016, Murugesan et al. 2019). These methods are focused mainly on the narrowing of energy gap and widening of visible light absorption both promoting photocatalytic disinfection. Nitrogen rich polymeric g-C<sub>3</sub>N<sub>4</sub> may be regarded as alternative to photocatalysts based on metals oxide (TiO<sub>2</sub> and ZnO) applied in photocatalytic disinfection.

Huang et al. (2014) have shown that *E. coli* K12 bacteria can be efficiently deactivated due to the use of g-C<sub>3</sub>N<sub>4</sub> of mesoporous structure. The surface of g-C<sub>3</sub>N<sub>4</sub> of such structure is 20 times larger (190 m<sup>2</sup>/g) than the one of g-C<sub>3</sub>N<sub>4</sub> of regular structure, while electron holes photogenerated on the surface

enhance bacteria inactivation (Kang et al. 2018). The use of mesoporous photocatalyst enables 100% inactivation of bacteria after 4 hour radiation with visible light, whereas g-C<sub>3</sub>N<sub>4</sub> of different structure reveals only 77.1% deactivation efficiency. Porous g-C<sub>3</sub>N<sub>4</sub> nanosheets possess better water disinfection efficiency in regard to *E. coli* due to large surface, low band gap and better electrons transport (Kang et al. 2018). Thurston et al. (2017) have evaluated biocide activity of two different g-C<sub>3</sub>N<sub>4</sub> foils against gram negative (*E. coli*) and gram positive (*S. aureus*) bacteria (Thurston et al. 2016). The improved activity of g-C<sub>3</sub>N<sub>4</sub> toward pathogenic microorganisms has been related to a large surface (72.2 m<sup>2</sup>/g), decreased energy of bandgap (2.86 eV) and efficient separation of photogenerated electron-hole pairs.

**Functionalization with carboxylic (-COOH) and carbonyl (-C=O) groups** introduced on g-C<sub>3</sub>N<sub>4</sub> edges may significantly promote inactivation of *E. coli* up to 6 log during 30 min exposure to visible light. Functionalization of nano-networks of g-C<sub>3</sub>N<sub>4</sub> efficiently favors charge separation and changes the location of surface bands upwards, what influences H<sub>2</sub>O<sub>2</sub> formation and improves overall disinfection effect (Teng et al. 2018). g-C<sub>3</sub>N<sub>4</sub> produced using urea reveals sporecide activity toward *Bacillus anthracis* (*B. anthracis*) endospores during exposure to visible light. In other research, almost 2×10<sup>7</sup> CFU/mL have been completely inactivated using single layer of g-C<sub>3</sub>N<sub>4</sub> within 4 hours and the reduction of 5–3 log *E. coli* by g-C<sub>3</sub>N<sub>4</sub> nanosheets has been obtained (Zhao et al. 2014). Inactivation of MS2 phages using g-C<sub>3</sub>N<sub>4</sub> has been investigated for the first time by Li et al. (2016). Almost all virus species (1×10<sup>8</sup> PFU/mL) have been completely deactivated within ca. 6 hours.

**Noble metals** introduced to g-C<sub>3</sub>N<sub>4</sub> photocatalysts may increase their photocatalytic activity by formation of charge carriers and widening of spectral absorption to visible light range. Additionally, they act as free electrons absorber enhancing the separation of photogenerated charge carriers and increasing photocatalytic efficiency of g-C<sub>3</sub>N<sub>4</sub> (Xue et al. 2015, Qin et al. 2015). It means that electron is transferred from g-C<sub>3</sub>N<sub>4</sub> CB to nanoparticles of a metal deposited on g-C<sub>3</sub>N<sub>4</sub> surface, whereas electron hole formed on g-C<sub>3</sub>N<sub>4</sub> surface remains available. The effective separation of charge carriers enhances photocatalytic disinfection by formation of ROS. The latter has been observed for Ag/g-C<sub>3</sub>N<sub>4</sub> photocatalyst, which has revealed better efficiency of *E. coli* inactivation and improved capability for destruction of biofilms containing peptides, nucleic acids and polysaccharides in reference to bare g-C<sub>3</sub>N<sub>4</sub> under visible light radiation (Bing et al. 2015, Ma et al. 2016). Xu et al. (2019) have also shown satisfactory antibacterial effect toward *S. aureus* using Ag doped g-C<sub>3</sub>N<sub>4</sub>. During 3 hour exposition, almost 29.6% and 99.4% of bacteria cells have been inactivated using g-C<sub>3</sub>N<sub>4</sub> and Ag/g-C<sub>3</sub>N<sub>4</sub>, respectively (Xu et al. 2019). The antimicrobial action of Ag/g-C<sub>3</sub>N<sub>4</sub> is obtained due to the presence of h<sup>+</sup> and \*O<sub>2</sub><sup>-</sup> formed during photocatalysis. Except for Ag, also the integration of Au with g-C<sub>3</sub>N<sub>4</sub> (CNA) may assure exceptional peroxidase activity toward decomposition of H<sub>2</sub>O to x\*OH radicals, and thus enable efficient deactivation of gram-positive and Gram-negative bacteria. Additionally, CNA reveals high toxicity toward cancer cells (Wang et al. 2016b).

There are also studies on photocatalytic disinfection activity of **non-metal** (N, S, C, P, etc.) doped g-C<sub>3</sub>N<sub>4</sub> available.

In one of them it has been shown that red phosphorus (r-P) nanoparticles doped on g-C<sub>3</sub>N<sub>4</sub> surface widen visible light absorption length to 700 nm and the material reveals better photodisinfection activity than bare g-C<sub>3</sub>N<sub>4</sub> and r-P (Wang et al. 2018).

Hybridization of g-C<sub>3</sub>N<sub>4</sub> with **other semiconductive** nanomaterials is regarded as attractive approach to improve its photocatalytic activity. Crucial features cover the improvement of visible light absorption and effective separation of charge carriers through transfer of electrons from higher CB to lower CB and transfer of electron holes from higher VB to lower VB. The research studies cover hybridization of TiO<sub>2</sub> with g-C<sub>3</sub>N<sub>4</sub> (g-C<sub>3</sub>N<sub>4</sub>/TiO<sub>2</sub>), and the obtained composites enable complete *E. coli* inactivation within 180 min under visible light radiation (Li et al. 2015). Xu et al. (2016) have described the visible light induced activity of g-C<sub>3</sub>N<sub>4</sub> layer deposited on TiO<sub>2</sub> nanotubes (TiNTs). The survival ratios of bacteria (the ratio of the number of bacteria colonies present on the test surface and the number of colonies on a reference glass substrate with none antibacterial activity) obtained for g-C<sub>3</sub>N<sub>4</sub>/TiNT and g-C<sub>3</sub>N<sub>4</sub> composite have reached ~16% and ~86%, respectively. Hybridization of semiconductive materials based on bismuth has also been checked in regard to their use in photocatalytic disinfection. For example, the efficiency of Bi<sub>2</sub>MoO<sub>6</sub>/g-C<sub>3</sub>N<sub>4</sub> (BM/CNNs) composite is higher than in the case of bare g-C<sub>3</sub>N<sub>4</sub> during exposure to visible light and the optimum BM share is 20 wt.% (Li et al. 2017). Another bismuth based composite comprised of dibismuth tetroxide and g-C<sub>3</sub>N<sub>4</sub> (g-C<sub>3</sub>N<sub>4</sub>/Bi<sub>2</sub>O<sub>4</sub>) inactivates *E. coli* K-12 with the efficiency reaching up to 6 log<sub>10</sub> CFU/mL during 1.5 hour exposure to visible light and is far more effective than g-C<sub>3</sub>N<sub>4</sub> (1.5 log<sub>10</sub>) and m-Bi<sub>2</sub>O<sub>4</sub> (4 log<sub>10</sub>) (Xia et al. 2017).

Carbon composites may also be combined with g-C<sub>3</sub>N<sub>4</sub> enhancing electron transfer to the latter material. For example, conjugation of g-C<sub>3</sub>N<sub>4</sub> of with graphene oxide (GO) of 2D structure is proved to improve the separation of photogenerated charges. Almost 97.9% inactivation of *E. coli* bacteria has been obtained using GO/g-C<sub>3</sub>N<sub>4</sub> composite at the concentration of 120 µg/mL and visible radiation time 120 min (Sun et al. 2017). A new class of metal-free heterojunction photocatalysts was prepared by wrapping reduced graphene oxide (RGO) and g-C<sub>3</sub>N<sub>4</sub> (CN) sheets on cyclooctasulfur crystals (α-S8) (Wang et al, 2013). Such structures exhibited antibacterial activity under visible-light irradiation. Also fullerenes (C<sub>60</sub> and C<sub>70</sub>) in combination with g-C<sub>3</sub>N<sub>4</sub> nanocomposites have been examined in regard to their antimicrobial activity using *E. coli* O157:H7 under visible light radiation. C<sub>70</sub>/g-C<sub>3</sub>N<sub>4</sub> composite has shown higher photoinactivation activity than C<sub>60</sub>/g-C<sub>3</sub>N<sub>4</sub> (ca.86%), whereas bare g-C<sub>3</sub>N<sub>4</sub> has deactivated only 68% of bacteria cells during 4 hour exposure to visible light (Ouyang et al. 2016, 2017).

## Conclusions and future perspectives

Advances in water purification using nanomaterials are mainly driven by their high efficiency in biological, physical, and chemical contaminants' removal at the nano-scale. It is to be noted that extensive research investments have recently been spent and earmarked for the future in this area due to the tremendous impact and potential that nanotechnologies have shown in various applications and particularly in water

and wastewater sanitation. Photocatalysis has appeared as an efficient and ecological method of water and wastewater disinfection. It has been found that the process allows for deactivation of various types of microorganisms. Despite many research studies available in this area, the search for simple, cheap and suitably active photocatalysts is still carried out. The dominant photocatalysts are characterized with specific limitation, whereas their industrially unattractive costs accompanied by troublesome synthesis protocols is the main challenge for the technology scale up. Up to this date, wide and independent research studies on disinfection and inactivation of variety of microorganisms have been performed and supplemented with individually designed research on such processes modelling. It has been clearly stated that operational parameter and factors influencing the occurring processes are very complex and thus, they require holistic approach. The research on photocatalytic disinfection combining both simulated data and results of processes performed using real water and wastewater streams would be very useful in complete understanding of the process pathways and mechanisms. However, regardless of these concerns, photocatalytic disinfection carried out with the use of sunlight appears an effective process suitable for commercial applications.

The rate of photocatalytic degradation strongly depends on adsorption of a given contaminant on a photocatalyst surface (Ganguly et al. 2018). It is also valid for photocatalytic inactivation of microorganisms. In order to assure efficient photoinactivation, microbes need to interact with a photocatalytic material surface (Ganguly et al. 2018). Reactive oxygen species (ROS) generated during the process firstly damage lipopolysaccharide layer of the cellular wall, next they react with peptidoglycan layer, and finally oxidize lipid layer and peptides (Ganguly et al. 2018). The destruction of this layer results in the leakage of potassium ions from cellular interior, what affects cells viability. This leakage leads to the loss of crucial cell functions and finally causes death of a cell (Ganguly et al. 2018).

The future development of nano-disinfection containing metal/metal oxides and carbon based nanoparticles should focus on:

- improvement of disinfection efficiency through different manufacturing strategies,
- proper clarification and understanding of the role and mechanism of interaction of the nano-material with the microorganisms,
- progress in scaling up the production of commercial nano-photocatalysts,
- determination of the extent of environmental release of nano-photocatalysts and their toxicity.

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