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Novel Antimicrobial Agents and Processes for Textile Applications

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Abstract

The use of antimicrobial compounds in textiles has grown dramatically over the last decades. The potential application field is wide. It ranges from industrial textiles exposed to weather such as awnings, screens and tents; upholstery used in large public areas such as hospitals, hotels and stations; fabrics for transports; protective clothing and personal protective equipment; bed sheets and blankets; textiles left wet between processing steps; intimate apparel, underwear, socks and sportswear. Another large field of application is in filtration and disinfection of air and water for white rooms, hospitals and operating theatres, food and pharmaceutical industries, water depuration, drinkable water supplying and air-conditioning systems. The present chapter is a review of recent research works related to antimicrobial finishes for textile materials. Several examples of antimicrobial treatments (e.g. traditional pad-dry-cure technique, exhaustion bath, encapsulation, electrospinning, cross-linking, etc.) were reported. The antimicrobial agents were divided by their origin from synthesis or from natural sources. Quaternary ammonium compounds (QACs), Triclosan, metals (including metal oxides and salts), polyhexamethylene biguanide (PHMB), N-halamines and conjugated polymers (i.e. polypyrrole) were listed as synthetic biocides in textile applications. Extracts from plants (e.g. aromatic compounds, essential oils and dyes), antimicrobial peptides (AMPs) and chitosan were considered among natural-based biocides.

Keywords: textiles, cotton, chitosan, polypyrrole, antimicrobial, photo-grafting

1. Introduction

Fibres, both natural and man-made, have been widely used since the ancient past in the manufacture of other materials. World fibre consumption has strongly increased over the years, reaching a total demand of 94.9 million tons in 2015. In detail, 66.8 million tons were man-made fibres, in addition to natural fibres with a demand of 28.1 million tons [1].

Nowadays, besides the traditional clothing products, textiles find important applications also in home furnishing, food packaging, as fibre reinforcements for polymers, optical fibres, thermal and mechanical protection, sport equipment, fibrous materials for a large array of applications in medicine and hygiene such as medical devices, health care and hygienic coatings, air filters and water purification systems.

An important example of these functional fabrics, recently attracting the interest of the research, is antimicrobial fabrics. Due to the morphology of fibres, in particular those of natural origin, textiles are prone to microorganisms' growth on their surface, due to the large surface area and moisture affinity. Bacteria and fungi can be found everywhere, so the contact with textiles is extremely probable. Depending on moisture, nutrients, temperature and pH, their growth can be very fast: some bacteria can double every 20 min [2].

The undesirable effects caused by microorganisms' growth act both on the textile itself and on the user. Unpleasant odour, reduction of mechanical strength, stains and discolouration are all effects of the biodeterioration of textiles, affecting almost all the types of fibres. Natural fibres are generally more susceptible to biodeterioration than the man-made fibres, because their porous hydrophilic structure retains water, oxygen and nutrients, providing perfect environments for bacterial growth. Finishing agents can also promote microbial growth [3]. Even mild surface growth can make a fabric unattractive by the appearance of unwanted pigmentation; heavy infestation which results in rotting and breakdown of the fibres may cause the fabric deterioration, in fact microorganisms can accelerate the hydrolysis of cellulose. Man-made fibres derived from cellulose are susceptible to microbial deterioration. Viscose is readily attacked by mildews and bacteria while acetate and triacetate are more resistant, but discolouration can occur if the fabrics are incorrectly stored. Synthetic fibres show strong resistance to attack by microorganisms, due to the hydrophobic nature of the polymers, but the presence of contaminants can cause some bacterial attack [3].

Most of the microorganisms involved in textile contaminations can cause pathogenic effects. Many species, such as *Escherichia coli*, *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Acinetobacter baumannii*, can cause infections to human beings due to user contaminations. It is a great concern for textiles used mainly in hospitals, as medical devices or for health and hygienic care, and in crowded places, but also in sport and underwear clothing, water purification systems, animal feed and food industry. Therefore, the demand for antimicrobial textiles is gaining interest, showing a strong increase over the last few years: the global market for antimicrobial agents is expected to increase by about 12% each year between 2013 and 2018 [4].

Different methods were investigated to confer antimicrobial activity to textiles; they can be classified into the inclusion of antimicrobial leaching compounds in the polymeric fibres, the surface modification of the fibres by grafting reactions or by physical methods.

Besides the efficiency towards a broad spectrum of microorganisms, any antimicrobial treatment should consider other challenges. First, it has to be non-toxic to the end user of the textile, namely cytotoxicity, allergy, irritation or sensitization must be avoided. The treatment must have a good fastness to use, mainly to repeated laundering, dry cleaning and ironing, and should not compromise the quality, hand or appearance of the textile. The application method should be simple, easily implementable in the finishing process and environmentally friendly, avoiding side effects for the manufacturers. Finally, the antimicrobial agent should not kill the resident flora of non-pathogenic bacteria on the skin of the wearer. Thus, the study on new and efficient antimicrobial treatments for textiles, considering both the antimicrobial agent and the application method, is a relevant topic of the research.

The aim of this chapter is to provide an overview of recently developed antimicrobial treatments to produce antimicrobial textiles. Afterwards, the discussion will be focused and detailed on chitosan and polypyrrole (PPy), two promising antimicrobial agents deeply investigated by the authors for textile applications.

2. Antimicrobial treatments for textiles

Depending on the fibre type, that is morphology, composition and surface texture, and on the applied antimicrobial agent, different chemical or physical approaches are possible and under development to confer antimicrobial activity to textiles.

In the case of synthetic fibres, a specific antimicrobial agent can be directly incorporated into the polymeric matrix [5].

The application of the antimicrobial agent on the fibre's surface, during the finishing stage, is a viable method both for synthetic and natural fibres; it can be carried out by the traditional pad-dry-cure technique or exhaustion bath.

The recent growing interest on nanotechnology concerns also the textile field; in fact nanoscale particles can be prepared from natural or synthetic compounds with antibacterial activity and applied to textiles, for example, by foulard. The advantage is the lower add-on enough to confer the desired property due to the high surface area of the nanoparticles. Moreover, coupling the process with a final cross-linking, a good fastness of the finishing can be obtained [6]. Electrospinning to produce intrinsically antimicrobial nanofibres is another interesting application of nanotechnology; in this case, the nanofibres can be coupled with other natural or synthetic fibres to produce antimicrobial yarns [7].

Microencapsulation is a process by which droplets of liquid or particles of solid are covered with a continuous film of polymeric material [8]. This technology is one of the most promis-

ing techniques to confer functional properties to textiles: the capsules are applied to fibres as dispersion with a binder using padding, spraying, impregnation, exhaust or screen-printing techniques. It is more advantageous than the conventional processes in terms of economy, energy saving, eco-friendliness and controlled release of substances, but it can affect the handle of the textile [9].

An effective way to embed the antimicrobial agent in the fibre is cross-linking. Cross-linking happens when a cross-linker makes intermolecular covalent bridges between the polymer chains and the antibacterial molecule. Cross-linkers include glutaraldehyde, genipin, glyoxal, dextran sulphate, 1,1,3,3-tetramethoxypropane, oxidized cyclodextrins, ethylene glycol diglycidyl ether, ethylene glycol diglycidyl ether (EGDE) and diisocyanate [10, 11].

Cross-linking can occur by chemical [12], radiation [13] or physical method [14]. In radiation, cross-linking, heat or a catalyst are not needed, thus no additional toxic chemical is introduced into the system and the substrate is preserved by a thermal degradation. The physical method is based on ionic interactions between polymer chains, so it is not as durable as the chemical or radiation ones.

Finally, altering the surface properties of fibres is also an interesting way to ensure a strong adhesion of finishing agents to textiles. Surface modification methods, such as oxygen plasma treatment, ultrasound technology, UV radiation, surface bridging and enzyme treatment, have been recently investigated, with the aim to impart durable antimicrobial finishes to fabrics using mainly natural products [15].

Depending on the application method, the antimicrobial textile can act by contact or by diffusion. In the first case, the antimicrobial agent is placed on the surface of the substrate and no leaching occurs; it will act only in case of direct contact between the microorganism and the fibres. In the second case, the agent will migrate from the textile to the external environment, to attack the microorganisms. It means that the antimicrobial activity of the textile can decrease with time, and that the impact of the antimicrobial agent on the environment has to be considered.

3. Antimicrobial agents of synthetic origin

According to its action against the microorganisms' cell, an antimicrobial agent can be classified as biostatic or biocidal. The first ones can just inhibit the cell growth, whereas biocidal agents can kill the microorganisms. Most of the antimicrobial agents used in commercial textile finishing are biocides, acting by damage or inhibition of cell wall synthesis, inhibition of cell membrane function, of protein synthesis, of DNA and RNA synthesis or of other metabolic processes.

Quaternary ammonium compounds (QACs) are cationic agents carrying a positive charge at the N atom in solution ($R_4N^+X^-$); they are usually attached to an anionic fibre surface (polyester, cotton, nylon and wool) by ionic interaction. The molecule is a linear alkyl ammonium chain composed of a hydrophobic alkyl chain (C12–C18) and a hydrophilic counterpart.

The antimicrobial action, depending on alkyl chain length, presence of perfluorinated groups and cationic ammonium group, is due to the interaction of positive charges on the surface and cell membrane negative charges, with the consequent loss of membrane permeability and cell leakage. It causes the damage of cell membranes, the denaturation of proteins and the inhibition of DNA production [16].

QACs are effective against Gram-positive and Gram-negative bacteria, fungi and certain types of viruses; for this reason, these are widely used in industrial applications [17]. The disadvantage is the poor fastness of the treatment due to the fast leaching from the textile for the lack of chemical or physical bonding [18].

Commercial products based on QAC are BIOGUARD® (AEGIS Microbe Shield, New Zealand), Sanigard KC® (LN Chemical Industries, Switzerland) and Sanitized® (SANITIZED, Switzerland) [19].

Triclosan is a 2,4,4'-trichloro-2'-hydroxydiphenyl ether ($C_{12}H_7Cl_3O_2$), a synthetic chlorinated bisphenol not ionized in solutions, improving its durability to laundering. It can act against Gram-negative and Gram-positive bacteria and against some fungi and viruses [5, 20] by blocking lipid biosynthesis affecting the integrity of cell membranes [21].

Triclosan has become, in last decades, the most efficient and widely used bisphenol in many application fields. On textiles, it is mainly used in association with polyester, nylon, polypropylene, cellulose acetate and acrylic fibres [22].

This recent widespread use of Triclosan had the drawback to generate bacterial resistance. Moreover, the reported photochemical conversion of Triclosan to 2,8-dichlorodibenzo-p-dioxin in aqueous solutions is another great concern, due to its toxicity [23].

Commercial products based on Triclosan, either as an isolated agent for a finishing or incorporated in fibres, are Microban® (Cannock, United Kingdom), Irgaguard® 1000 (Ludwigshafen, Germany), BiofresH™ (Salem, MA, USA) and Silfresh® (Magenta, Italy).

Metals, oxide or salt compounds, based on silver, copper, zinc or cobalt, have a strong biocidal effect due to the metal reduction potential, metal donor atom selectivity and speciation. These compounds can bind to O, N or S donor ligands present in the microorganism cell, inducing an oxidative stress, damaging cellular proteins, lipids and DNA.

Among all, silver particles were widely exploited due to the broad spectrum of action against bacteria like *P. aeruginosa*, *S. aureus*, *S. epidermidis*, *E. coli* and *K. pneumoniae*. In textile fields, they are mainly applied in the form of salts (79%) rather than metallic (13%) or ionic (8%) form [24]. Recently, the application in the form of nanoparticles, obtained by sol-gel, is gaining interest for silver, CuO, ZnO and TiO_2 [25]. It is due to the higher surface area with respect to larger particles, higher solubility and faster release of the metal ions, turning in a stronger antimicrobial effect. On ZnO, it was found that the antibacterial activity is inversely proportional to the nanoparticle size [26].

The scale-up of the process to commercial scale, unfortunately, was contained due to cost, environmental and technical challenges. A plasma, UV or acidic pre-treatment is often required on fabrics to improve the treatment durability, otherwise not so good [27].

Available commercial products are mainly based on silver, in isolated form, for fibre finishing or incorporation, or already in fibre or fabric form. Some examples are Ultra-Fresh[®] and Silpure[®], SmartSilver[®], MicroFresh[®] and SoleFresh[®], Bioactive[®] and Silvadur[™].

Polyhexamethylene biguanide (PHMB) ($(C_8H_{17}N_5)_n$) is a polycationic amine in which the cationic biguanide groups are interdispersed between hydrophobic hexamethylene groups. Electrostatic and hydrophobic interactions occur with microbial cell membranes, resulting in cell membrane disruption and lethal leakage of cytoplasmic materials. Its antibacterial activity increases with the level of polymerization [28]. Some PHMB-based textile products, such as Biozac ZS and Reputex[®], have already appeared on the market as finishing products [29].

N-halamines are heterocyclic organic compounds, with one or two covalent bonds between nitrogen and a halogen, usually chlorine (N–Cl). N-halamines can be imide, amide or amine depending on the covalent bonds formed; the antimicrobial activity increases in the same order, while the stability decreases. N-halamines present a biocide action against a broad spectrum of bacteria, fungi and viruses, binding to the acceptor regions on microorganisms, precluding the cell enzymatic and metabolic processes and causing the consequent microorganism destruction [30]. Besides the low cost and wide range action, an advantage is the possibility to recharge their antimicrobial effect of the inactive substance by simply reacting them with Cl donor compounds [31]. As a disadvantage, textiles' treatment with N-halamine may result in a substantial amount of adsorbed Cl on the fibre surface. Those residues may produce an unpleasant odour or even discolour fabrics, which is a concerning disadvantage to the textile industry.

Conjugated polymers, such as **polypyrrole**, are generally applied in textile field as electrically conductive coating in order to produce electrically conductive textiles [32, 33]. PPy can be easily produced by chemical oxidative polymerization from water solutions of the monomer. Textile materials (e.g. fibres, yarns and fabrics) soaked in the polymerization bath are coated with an even and uniform layer of PPy by in situ chemical oxidative polymerization. During the oxidative polymerization, positive charges are introduced along the backbone chain of PPy. The charges are counter-balanced by counter-ions (also called dopants or doping agents), namely anions present in the polymerization bath. The anions in the polymerization bath are embedded in the polymer matrix improving the formation and stability of positive charges along the backbone chain of PPy, delocalized over several monomer units. PPy has been a subject of several works that evaluate its properties as biocidal agent. Excellent antimicrobial properties have been shown against both Gram-negative and Gram-positive bacteria. Such a bioactivity of PPy is likely due to the presence of positive charges, even if no leaching of biocidal substances has been proven on PPy-coated fabrics. The 'non-leaching' approach would avoid or limit the release of toxic biocide agents to the environment or to the skin of the wearers, in the case of garments.

PPy nanoparticles were synthesized by chemical polymerization using ammonium persulphate as oxidant following different methods in order to evaluate the influence on the morphology of resulting nanoparticles and bactericidal activity [34]. Five systems were synthesized: conventional PPy (without surfactants), highly soluble PPy (in SDS solution), PPy/Ag colloid (in PVA solution), branched PPy and branched PPy/Ag nanocomposite (in CTAB/

SDBA solution). Resulting polymer particles were investigated as active bactericidal materials against *E. coli*, *S. aureus* and *K. pneumoniae*. Inhibition zones, minimum inhibitory concentration (MIC) and time-kill assays were evaluated. The results indicate that the incorporation of silver nanoparticles improves the biocidal action of PPy and confirm that the size of PPy nanoparticles represents a relevant parameter for the bactericidal activity. In particular, it is possible to list the biocidal activity as follows: highly soluble PPy > branched PPy with silver > branched PPy > colloidal PPy > conventional PPy.

Antimicrobial activity of PPy on fabrics was first reported by Seshadri and Bhat [35] in 2005. In particular, they deposited PPy on cotton fabrics by in situ chemical oxidative polymerization at cold temperature (0–5°C). The fabrics were impregnated with monomer solutions and then the oxidant solution (ferric chloride) was added producing PPy-coated fabrics. CuCl_2 was used to treat samples of PPy-coated fabrics as an additional antimicrobial agent. The biocidal properties were tested by AATCC Test Method 147-1993 and ASTM E 2149-01 using *S. aureus*, *E. coli* and *Candida albicans*. The microbial reductions were 65, 59 and 73%, respectively. The addition of CuCl_2 to PPy increased the biocidal efficiency to 93, 98 and 100%, respectively.

In another paper [36], cotton fabrics were coated with PPy at room temperature using different oxidizing agents in order to assess their antimicrobial efficacy. The fabrics were soaked in a water solution of the oxidant. The monomer was added drop-wise to the stirred bath, and the reaction lasted for 4 h producing an even black layer of PPy on the fibres of the fabrics (Figure 1).

To obtain information about the influence of the oxidation agents on the biocidal activity, the synthesis of PPy was carried out using three different oxidants: ferric chloride, ferric sulphate and ammonium persulphate.

With ferric chloride and ferric sulphate, PPy is produced by the redox reaction between the monomer and ferric ions, reduced to ferrous ions. Using ammonium persulphate, the oxidative component is persulphate ion reduced to $\text{SO}_4^{=}$. Using ferric sulphate and ammonium persulphate, PPy embeds $\text{SO}_4^{=}$ ions as counter-ions, whereas the PPy produced with ferric

Figure 1. (a) Picture of the PPy-coated cotton fabric and (b) SEM image of cotton fibres coated by PPy.

chloride embeds Cl^- . Both ferric chloride and ferric sulphate give a high acidic pH to the polymerization bath due to the production of ferric complexes with OH^- . On the contrary, solutions of ammonium persulphate have a relatively low pH, due to the hydrolysis equilibrium of ammonium ions and water.

Antibacterial activity of PPy-coated fabrics was evaluated following the ISO 20645:2004 procedure using *E. coli* by placing the fabrics in contact with bacteria. No inhibition zone was observed after 24 h and the colonies grew around the fabric. The absence of colonies was observed under the fabrics in the contact zone. Therefore, there is an antibacterial activity on the fabric surface just by contact because PPy cannot diffuse being linked to the fabric. The absence of bacterial growth, even without inhibition zone, may be considered as a good antibacterial compound. Finally, the results pointed out that the antibacterial property is independent on either the oxidant used in the synthesis of PPy or the dopant embedded in the polymer matrix.

Cotton fabrics have also been coated with PPy using dicyclohexyl sulposuccinate (DSS) [37]. DSS has two functions: (a) it is embedded into PPy as counter-ion, similarly to several other dopants with an $-\text{SO}_3^-$ group, (b) it greatly enhances the deposition process of PPy on the fabric by lowering the surface tension as a surfactant, and in turn it increases the evenness and weight uptake of PPy. In fact, the weight uptake of PPy has been 12% without DSS and 18% with DSS. The difference has been attributed primarily to the more efficient deposition due to the increased wetting of fibre surface caused by the surfactant action of DSS.

Antibacterial activity of the PPy-coated fabrics has been evaluated following the ASTM E 2149-01 procedure. Both the fabrics coated with PPy without DSS and PPy with DSS show 100% bacterial reduction, while untreated cotton fabric had practically no antibacterial activity. The stability of the biocidal action has been evaluated after different kinds of laundering. In particular, after dry-cleaning, fabrics coated with PPy without DSS and PPy with DSS show high bacterial reductions, 99 and 98%, respectively, whilst antibacterial efficiency decreases after launderings with non-ionic and anionic surfactants. In particular, the antibacterial activity of fabrics coated with PPy without DSS has been severely degraded by anionic laundering.

Moreover, the paper evaluated the biocidal mechanism of PPy by carrying out scanning electron microscopy (SEM) analysis of *E. coli* bacteria on cotton fibres and PPy-treated fibres. Bacteria on untreated cotton fibres had typical and regular bacterial shapes indicating that cells survived on the fibre surface. On the contrary, *E. coli* cells on PPy-coated fibres showed altered shapes probably due to the opening of their membrane and leakage of intracellular components from bacterial cells.

Recently, PPy has also been used for antimicrobial applications in combination with silver [38, 39]. In particular, Omastová et al. [40] prepared polypyrrole/silver composites by a single-step chemical oxidative polymerization using silver nitrate as an oxidant in water at room temperature. The reaction needed several days in order to yield more than 70%. The silver content in the PPy was estimated in the range of 70–80 wt%.

PPy/silver composites are composed of globules of about 1- μm diameter. This globular morphology is typical of PPy prepared with classical oxidants, such as iron(III) salts. The molecular

structure of PPy produced was characterized by Fourier transform infra-red spectroscopy (FTIR) and Raman spectroscopy showing the same features as in PPy prepared with other oxidants. The morphology of silver nanoparticles was evaluated by transmission electron microscopy (TEM). Silver is present in particles of 50–100 nm size and occasionally larger polygonal crystals.

PPy/silver nanocomposites were used to coat cotton fabrics by in situ chemical oxidative polymerization using silver nitrate [39]. In a redox reaction, silver ions oxidize the pyrrole monomer and reduce to Ag^0 . The reduced silver was deposited on/into the polypyrrole/cotton matrix layer as nanoparticles. In the beginning, the cotton fabric was impregnated in a solution containing pyrrole. Silver nitrate was added into this solution and stirred. After completion of reaction, the cotton fabric was coated with a PPy/silver nanocomposites layer.

The antimicrobial activity of PPy/silver-coated fabrics against *E. coli* and *S. aureus* bacteria was evaluated by the assessment test and agar diffusion test. The antimicrobial property of PPy/silver composites was measured by the clear zone of inhibition around the fabrics after incubation in agar plate method. Untreated cotton shows no antimicrobial activity against both bacteria. Moreover, the bacteria were grown over the surface. The PPy-coated cotton shows a small inhibition zone, whereas in PPy/silver composite-coated fabrics the inhibition zone was found to increase with increasing concentration of silver in the composites.

The antibacterial activity of the PPy/silver composite-coated cotton fabrics was also quantified according to the AATCC 100-1999 procedure. The PPy/silver composite-coated cotton fabrics show a gradually increased bacterial reduction percentage over the contact time. The paper reported that the bacterial reduction reaches likely 100% within 6 h against *E. coli* and 12 h against *S. aureus*.

Commercially available antimicrobial fabrics already include fabrics composed of silver-coated fibres. Therefore, another possible approach could be to treat this kind of fabrics with PPy instead to synthesize silver nanoparticles during PPy deposition as previously reported.

In a work [41], PPy deposition was carried out on cotton fabrics containing 10% of silver-coated fibres. PPy was synthesized at room temperature using ferric sulphate as oxidant. Antibacterial activity was evaluated following the AATCC Test Method 100–2012 against Gram-positive bacteria on textiles with different amount of PPy on fabrics with silver-coated fibres and pure cotton fabrics (without silver-coated fibres). A synergic biocidal effect between silver ions and PPy was observed. In fact, silver-containing fabrics used in this work alone does not guarantee a complete biocidal effect, but the addition of just 2 wt% of polypyrrole showed a bacterial reduction of 99%. On the other hand, excellent bacterial reduction (>99%) was found on pure cotton fabrics containing more than ~9 wt% of PPy, but the amount of PPy can be reduced to 5% in the presence of silver to reach the same level of efficiency.

Few papers reported the applications of PPy to man-made fibres for antimicrobial purposes. In particular, a study [42] was investigated where a polyethylene terephthalate (PET) fabric was coated with reduced graphene oxide (RGO) sheets, and then a PPy layer was deposited by in situ polymerization in order to cover RGO.

Antibacterial activity was assessed qualitatively against *S. aureus* (ATCC 25923) and *E. coli* (ATCC 25922) with AATCC 100-2003 standard method. PPy-coated sample showed antibacterial activity against both types of bacteria. The sample treated with RGO/PPy composite layer also showed excellent antibacterial activity against both bacteria that can be attributed to the existence of PPy with its antibacterial activity. No antibacterial activity was found on RGO-coated fabrics.

4. Antimicrobial agents of natural origin

Bacterial resistance to biocides, their inadequate activity, toxic effects on households and the environment and poor durability on textiles have become important issues of concern.

Some antimicrobial agents are commercially marketed as 'eco-friendly', such as Ultrafresh by Thomson Research Associates, Tinosan AM 110 (2,4,4'-Trichloro-2'-hydroxyl-diphenyl ether) by Ciba Specialty Chemicals, Sanitized AG by Clariant, Ecosy by Unitika, Utex by Nantech Textile Company Limited and Vantocil IB by Zeneca. However, investigating the chemistry behind these purportedly natural biocides, it is clear that they are not entirely natural.

As a consequence, certain synthetic antimicrobial agents, such as Triclosan, have been banned by a number of leading retailers and governments in Europe, for their potential to cause skin irritation, non-biodegradable and bioaccumulation effects [43].

Due to these concerns, coupled with the high level of consumer awareness about clothing safety, many kinds of eco-friendly antimicrobial agents such as peroxy acids, chitosan and its derivatives or specific dyes have been developed for textiles.

In the last years, significant progresses in the discovery of new compounds with antimicrobial activity, from natural products, were made. These substances may present an efficient antimicrobial effect, with safety, easy availability, non-toxicity to skin and environmental friendliness. Moreover, no resistance of pathogenic bacteria was reported towards these natural chemicals.

Plants have received interest as a major source of natural antimicrobials in nature [44, 45]. Materials extracted from different parts of plants such as bark, leaves, roots and flowers containing tannin, flavonoids and quinonoids but also alkaloids, saponins, terpenoids and phenolic compounds, with strong antimicrobial properties, have been investigated [46, 47].

Even essential oils, a mixture of a variety of aromatic compounds which can give cologne, can provide protection from a broad spectrum of microbes. The application of essential oils for antimicrobial effect on textiles has increased in recent times, due to their high efficiency, even if the real action against microbes is not clear.

A synergistic effect was noted, for example, for carvacrol and some hydrocarbon monoterpenes showing good antimicrobial properties: probably the hydrocarbons interacted with the cell membrane of the microbes and facilitated quick penetration of carvacrol into their cells. Similar effects were reported for eugenol/carvacrol and eugenol/thymol towards *E. coli*,

suggesting that carvacrol and thymol disintegrated the outer membrane of *E. coli*, making it easier for eugenol to enter the cytoplasm. The advantages of synergy are the reduction of the concentration required to yield the same antimicrobial effect when compared with the sum of the purified components [48, 49].

Natural dyes, extracted from bark, leaves, roots, fruits, seeds and flowers, or from micro-organisms such as fungi, algae and bacteria, have an inherent antimicrobial property due to the presence of different colouring materials such as tannin, flavonoids and quinonoids. Moreover, they offer a wide range of colours, are environmentally friendly and can be used in low-cost treatments with the additional benefit of colouring and confer antimicrobial activity in a single step [50].

Natural antimicrobial peptides, present in every living organism, are also promising natural candidates for antimicrobial textile applications. They are characterized by their small size (12–50 amino acids), the arginine and lysine residues responsible for their positive charge, and an amphipathic structure that interacts with microbial membranes. Some examples are daptomycin (Cubicin®, Cubist Pharmaceuticals), pexiganan, psoriazyna and plectasin NZ2114. Another efficient AMP is L-cysteine, successfully used to promote the biofunctionalization of wool and polyamide, conferring a durable antimicrobial finishing [51] to those fibres.

5. Chitosan

Among the antimicrobial agents of natural origin, chitosan is gaining great interest in the last decades; in fact, chitosan and its derivatives appear to be the most effective natural antimicrobial agent on the market.

5.1. Structure and properties

Chitosan (2-amino-2-deoxy-(1→4)- β -D-glucopyranan) is a natural biopolymer, resulting from the deacetylation of chitin, constituting 20–30% of the exoskeleton of crustaceans.

It is the second most abundant biopolymer in the world, following cellulose, meaning an easy availability at low cost [52]. Its natural origin makes it biodegradable, biocompatible, non-toxic and non-carcinogenic, that is, an eco-friendly product avoiding any environmental or hygienic issues.

This biopolymer shows excellent film- and coating-forming properties when cast from organic acidic water solutions and, last but not least, it shows a strong antimicrobial activity against a wide spectrum of microorganisms, including fungi, algae and some bacteria.

The antimicrobial action of chitosan is influenced by intrinsic factors and environmental conditions, such as the chitosan molecular weight and polymerization degree, its deacetylation degree, the pH of the medium and the microorganism type.

Chitosan is considered to be both bacteriocidal and bacteriostatic although the exact action mechanism is not fully understood. The most acceptable models describe the interaction

between positively charged chitosan groups and negatively charged microbial cell membranes due to electrostatic interactions. It promotes changes in the properties of membrane wall permeability causing internal osmotic imbalances and consequently inhibiting the growth of microorganisms. Even the hydrolysis of the peptidoglycans in the microorganism wall occurs, leading to the leakage of intracellular electrolytes as proteins, nucleic acids and glucose. Another proposed mechanism is the binding of chitosan with microbial DNA, which leads to the inhibition of the mRNA and protein synthesis via the penetration of chitosan into the nuclei of the microorganism, reaching the plasma membrane. A third mechanism is based on the excellent metal-binding capacity of chitosan due to the amine groups which are responsible for the uptake of metal cations by chelation, suppression of spore elements and binding to essential nutrients to microbial growth [53, 54].

Due to its diversified application fields, chitosan is a biomolecule with great potential. The antimicrobial activity was undoubtedly the most interesting application in recent years, leading to a wide application of chitosan, mainly in the field of food packaging and edible films, for biomedical and pharmaceutical purposes (drug delivery or tissue engineering), cosmetics and dermatological, agriculture, paper, enzyme immobilization and, of course, in textile field.

5.2. Use as antimicrobial agent for textiles

The use of chitosan and its derivatives on fibres seems to be the more realistic prospect since this product does not provoke any immunological response.

Besides the biocidal properties of chitosan on textiles, it also has several other advantages considering the further colouration, because the amine group present readily reacts with dyes for successful dyeing/printing [55].

Chitosan is mostly applied by the traditional pad-dry-cure process using chitosan/citric acid mixture mainly on cotton fabrics, even though other techniques have been used to impart antimicrobial property to fabrics. The use of binders with chitosan has also been reported [56] with the advantage that it can be applied to all manner of fabrics due to the presence of the binder.

Complexes based on chitosan and other biocidal agents have been studied to increase treatment efficiency and durability [57, 58]. Promising results were found with bivalent metal ions, such as Cu(II), Zn(II) and Fe(II), showing an antimicrobial effect much higher than the single components, due to the stronger positive charge after complexation [59], and with nanocapsules based on antibacterial polypeptide-grafted chitosan [60].

Despite some disadvantages in the use of chitosan in textile field, namely some temperature and pH activity dependence and poor handling, Eosy[®], a commercial finishing product based on this biopolymer, and a composite fibre of chitosan and viscose, named Crabyon[®], presenting a durable antimicrobial efficacy, are already available [61].

In textile field, the most common way to apply chitosan to fabrics is by wet thermal curing, involving relatively high temperature with energy consumption, costs and possible fabric degradation; moreover, the addition of toxic reagents, such as glutaraldehyde, is requested as cross-linking agent.

In recent research works, ultraviolet radiation in the presence of a suitable photoinitiator was proposed as an alternative process to graft chitosan molecules to textile fibres by radical process [62, 63]. In detail, in case of a cellulosic substrate such as cotton, the same cellulose molecule can be involved in the reaction by the formation of radicals, which can react with those formed on the chitosan molecule, conferring a strong fastness to the finishing. Moreover, UV grafting is a fast and eco-friendly process, carried out at room temperature, with lower cost than the traditional thermal process.

Cotton, silk and synthetic fabrics were considered as substrates. Obtained results showed that chitosan UV curing yielded strong antimicrobial properties, reaching 100% m.o. reduction on all considered fabrics, as confirmed by antimicrobial tests carried out also on chitosan film. Moreover, low add-ons, 1–3% o.w.f., are enough to confer the desired property to the fabrics, so the hand properties of cotton or silk and the filtration capacity of synthetic fabrics are not compromised. In order to have a good treatment fastness, chitosan has to be diluted with acetic acid solution before spreading on fabrics and an impregnation time of 12 h at an ambient temperature or 1 h at 50°C is necessary before the curing to ensure a good penetration inside the fibres.

The homogeneous distribution of chitosan on fabrics was confirmed by dyeing tests with an acid dye and by SEM analysis (**Figure 2**), which showed the optimal distribution of the finish on single fibre surface, while the presence of amino groups before and after the washing test, responsible for the antimicrobial activity, was revealed by ninhydrin assay and FTIR-attenuated total reflectance (ATR) spectra.

Chitosan film was characterized by differential scanning calorimetry (DSC) and FTIR analysis. Data found are perfectly in agreement with literature data related to thermally cured

Figure 2. Chitosan UV grafted to fibres: cotton (a), PET (b), PA (c) and silk (d).

chitosan, meaning that UV curing leads at the same polymer structure. Finally, on the FTIR-ATR spectra of treated cotton or synthetic fabrics, in comparison with untreated samples, the presence of the typical bands of chitosan is evident, showing its presence again.

Chitosan was applied by UV grafting also to wool fibres, to confer a multifunctional finishing to the fabric, improving its value and application fields [64].

The antimicrobial activity, reaching 67% *E. coli* reduction, was obtained by a surface modification of wool fibres with 2% grafted chitosan, preceded by an oxidative wool pre-treatment and 1-h impregnation at 50°C to enhance chitosan penetration in wool fibres. It was coupled to an improvement of wool dyeability towards acid dyes and some anti-felting properties with respect to the untreated substrate, while the treatment fastness to laundering depended on the used surfactant and no anti-pilling properties were conferred.

A semi-industrial scale-up of the process was carried out on cotton fabrics, following an encouraging preliminary laboratory research, on samples of reduced dimensions, aimed to optimize the main process parameters. To test the feasibility of the proposed treatment at larger scale, large white or dyed fabrics were impregnated by foulard with a commercial chitosan solution, significantly reducing the add-on to restrain the costs. Then, they were irradiated, both dried and wet, with a high-power UV lamp, in air.

Obtained results confirmed the previous, laboratory scale, ones: a strong antibacterial activity with good washing fastness (99.9% microorganisms reduction after 30 washing cycles) was achieved by irradiation of the samples even wet and in air. It was obtained with a chitosan add-on percentage lowered till 0.3 wt% with a negligible affection of colour or hand properties of the fabric [65].

5.3. Wastewater purification

Wastewater treatment is one of the major current applications of chitin/chitosan-based products due to their coagulating, flocculating and chelating properties.

Ecological and health problems associated with heavy metals and pesticides accumulation in water and, as a consequence, through the food chain prompted the need for purification of industrial water in an efficient way.

The ability of the free amino groups of chitosan to form coordinate/covalent bonds with metal ions is of great interest: chitosan in the form of a film or a powder or suitably grafted to an inert substrate can be used in metal ion complexing, in particular above its pK_a value (about 6.5).

Chitosan, carboxymethyl chitosan and cross-linked chitosan showed a strong efficiency in removing Cd²⁺, Cu²⁺, Hg²⁺, Ni²⁺ and Zn²⁺ ions from wastewater and industrial effluents [66–69].

Chitosan was tested also as a sequestering agent for dye molecules, mainly present in wastewater from dyeing plants, showing a high efficiency towards different dye classes: acid, reactive, anionic and direct dyes [70–72].

Cotton gauzes coated with chitosan using a UV-curing process were also tested, in static and dynamic conditions, as water filter for biological disinfection against both Gram-negative and Gram-positive bacteria. The material showed good antibacterial activity against *E. coli*, *S.*

aureus and *K. pneumoniae*, in both static assessment and dynamic conditions: chitosan-treated gauze showed a high antimicrobial efficiency in few seconds of contact time. Results are of interest even if compared with those related to the same cotton gauze cationized with a quaternary ammonium salt. A certain pH sensitivity was found, but in all cases microorganism reduction never fell under 80% [73].

It makes this composite a good candidate for its real use as biological filter.

6. Conclusions and future perspectives

The presence of Gram-positive and Gram-negative bacteria and fungi is common on textiles, involving both synthetic and natural fibres. Due to their structure and chemical composition, textile products are prone to host microorganisms whose proliferation is responsible for diseases, unpleasant odours, colour degradation and deterioration of fabrics. Many of these microorganisms are pathogens, quite often related to nosocomial infections; therefore, the development of non-toxic processes for the preparation of antimicrobial textiles is gaining interest from both the academic researches and industry.

Emerging applications for biocidal finishes in textiles are required in different fields: sportswear, underwear, textile-based medical devices, home furnishing, filtration and depuration of water or air in crowded public areas. The required effect, depending on the application, can vary from the simple odour control to disease and infection control.

Among the novel, natural and eco-friendly antibacterial finishing of textiles, a strong antibacterial finishing of textile substrates, with good fastness and stability, was obtained with both photo-grafted chitosan and polypyrrole coating on textiles.

Moreover, the morphology of PPy particles seems to effect antimicrobial performances, but the works, at the moment, were focused on particles not linked to a substrate. Further studies to improve PPy properties will appoint to produce PPy coating on the fabrics with particles having a designed shape.

In another study not dealing with textile applications, polypyrrole-graft-chitosan (PPy-g-CS) copolymer was chemically synthesized and characterized [74]. PPy-g-CS showed an antibacterial activity stronger than chitosan and PPy alone, comparable with the antibiotics considered as reference. It suggests a synergic effect of polypyrrole-chitosan coating, exploitable in textiles.

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