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### Editorial

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**Mini Review Section** — With rapid advances being made in *Agrobacterium* and transgenic plant research, the possibilities for crop improvement are numerous. One recent advance epitomizes the current situation: coffee (*Coffea canephorea*) plants that have been genetically modified to contain less caffeine. The caffeine biosynthesis gene, coding for obromine synthase, was targeted for down-regulation using the concept of RNA interference.

**Current Trend Section** - Surface disinfection depends on several parameters and is widely used for cleaning, disinfection, and sterilization. Thanks to innovative technologies, protocols, and creativity, new methods have been developed with a continued evolution to improve performance. Despite the effective solutions given by traditional methods of surface disinfection, modern nanotechnology has proven to be an emergent innovation to protect against viruses.

In Profile Scientist - Ehrlich became interested in the selectivity of dyes for specific organs, tissues, and cells, and he continued his investigations at the Charité Hospital in Berlin. After he showed that dyes react specifically with various components of blood cells and the cells of other tissues, he began to test the dyes for therapeutic properties to determine whether they could kill off disease-causing microbes. He met with promising results using methylene blue to kill the malaria parasite.

**Bug of The Month** - *Chlamydophila psittaci* (formerly *Chlamydia psittaci*) is a small bacterium that undergoes several transformations during its lifecycle. It exists as an elementary body (EB) between hosts. The EB is not biologically active but is resistant to environmental stresses and can survive outside a host. The EB travels from an infected bird to the lungs of an uninfected bird or person in small droplets and is responsible for infection. Once in the lungs, the EB is taken up by cells in a pouch called an endosome by phagocytosis. However, the EB is not destroyed by fusion with lysosomes, as is typical for phagocytosed material. Instead, it transforms into a reticulate body and begins to replicate within the endosome.

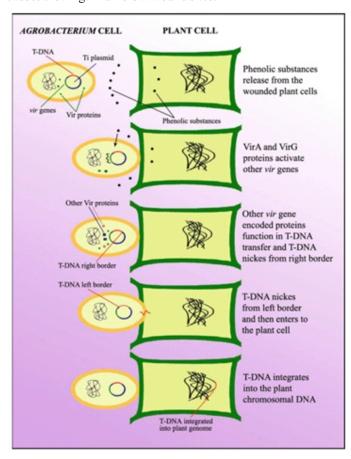
**Did You Know** – In the latest research published in Genes, it was shown how the population of bacteria on a person's skin leaves traces on the clothes they wear—and how these traces last for months and can be used to uniquely identify the wearer.

**Best Practices** - Microbes can spread and grow in the home, particularly in the kitchen, bathroom, and laundry areas. The highest counts of microbes in the kitchen and bathroom are found in wet areas around the sink, in sponges and cloths used for wiping and/or drying kitchen surfaces, and in the areas around the bathroom sink. Water temperature can influence the survival of microbes during dishwashing and laundry practices. For the laundry, drying is the most reliable method for destroying microbes.

# Importance of Agrobacterium tumefaciens in genetic manipulation II

### Molecular Mechanism of Gene Transfer

Agrobacterium, of the family Rhizobiaceae, is a genus of Gramnegative bacterium that genetically transforms host plants and causes crown gall tumors at wound sites.



Agrobacterium can transfer DNA to a broad group of organisms: plants, fungi such as yeasts, ascomycetes, and basidiomycetes, and protist such as algae.

Agrobacterium is usually classified by the disease symptomology (type of opine) and host range.

The process of gene transfer from A. tumefaciens into plant cells implies several essential steps: (1) bacterial colonization, (2) induction of bacterial virulence system, (3) generation of T-DNA transfer complex, (4) T-DNA transfer and (5) integration of T-DNA into plant genome.

Bacterial recognition of monosaccharide and phenolic compounds secreted by the plant wound site initiates the tumor induction. "Activated" Agrobacterium transfers a particular gene segment called transfer DNA (T-DNA) from the Ti plasmid. After T-DNA is stably integrated into the chromosomal DNA in the nucleus of the host plant, genes for opine synthesis and tumorinducing factors on the T-DNA are transcribed in the infected cells. This expression of the foreign gene in the host plant results

in neoplastic growth of the tumors, providing increased synthesis and secretion of opine for bacterial consumption. Opine is the condensation of an amino acid with a keto acid or sugar and is a major carbon and nitrogen source for Agrobacterium growth.

The region on the Ti plasmid is called the transfer DNA (T-DNA). The T-DNA region is a small segment that integrates into the plant genome by being transferred from bacterium into the plant cell. The second condition that plays an important role in gene transfer from Agrobacterium to the plant cell is the virulence (vir) region that is outside of the T-DNA and close to the left border with a nearly 25 kb length.

Vir region contains six main genes (virA, virB, virC, virD, virE, and virG). VirA codes for a receptor that detects and correlates with phenolic compounds leaking out of damaged plant cells, and consequently, virG is stimulated. Stimulated VirG takes charge of the transcription operator task for itself and the other vir genes. virC enables to separate from the borders, while virD gene provides the regeneration of the T-DNA strand; virB and virE genes facilitate the move of the T-DNA from the bacterium to the plant cell.

The third condition that is influential in gene transfer into the plant cells is the compounds coded by three loci (chvA, chvB and pscA) in the chromosomes of Agrobacterium, being of great importance for the bacterium to attach to the plant cell and to respond to the specific chemical (chemotaxis).



### Why is Agrobacterium used to make transgenic plants?

Agrobacterium is a useful tool for plant transformation because it can carry, transfer, and integrate a gene of interest into the plant genome.

In the development of transgenic plants, this system allows plants to stably harbor and pass a particular gene of interest to the next generations relatively quicker than by using the more traditional plant breeding method. This method is relatively inexpensive and



easy to perform. In addition, it provides convenient way to screen and select the transformed plant tissues.

There are at least three main components to prepare before performing Agrobacterium-mediated transformation:

- 1) T-binary system
- 2) Agrobacterium Competent Cells
- 3) Plants

### How to Choose Agrobacterium Competent Cells Agrobacterium-mediated transformation is a process of using Agrobacterium to transfer a gene of interest into the

plant cells, generating transgenic plants.

When choosing Agrobacterium competent cells, some factors to consider:

### 1. Transformation Efficiency

High transformation efficiency is an important feature for Agrobacterium cells because these cells enable to take up T-DNA efficiently.

### 2. Antibiotic Resistance

Some strains of Agrobacterium have a particular antibiotic resistance. Therefore, avoid using a vector with the same antibiotic marker to carry your gene of interest.

### 3. Compatibility with the Plants

Some factors can affect successful plant transformation, including the susceptibility of the plant to Agrobacterium infection, the efficiency of Agrobacterium-mediated gene delivery, and the ability of the plant to express the protein and regenerate whole plants from transformed cells.

### Adoption of plant molecular biology

A.tumefaciens has been used for plant genetic engineering extensively. Plants were genetically engineered for the purpose of developing resistance to herbicides, insect, or virus, tolerance to drought, salt, or cold, and increasing the yield. The Agrobacterium-mediated transformation method has not only been used for commercial purpose but also for basic biology

research to test study gene regulation or protein function in transgenic plants.

The Agrobacterium-mediated transformation method was improved by the strategy of developing modern binary Ti plasmid. Ti plasmids have been engineered to separate T-DNA and vir regions into two distinct plasmids, resulting in a binary vector and a vir helper plasmid, respectively.

With rapid advances being made in Agrobacterium and transgenic plant research, the possibilities for crop improvement are numerous. One recent advance epitomizes the current situation: coffee (Coffea canephorea) plants that have been genetically modified to contain less caffeine. The caffeine biosynthesis gene, coding for obromine synthase, was targeted for down-regulation using the concept of RNA interference. The transgenic coffee plantlets obtained after Agrobacteriummediated transformation had a 50% to 70% reduction in caffeine. These decaffeinated coffee plants will, in theory, bring an end to the expensive, industrial decaffeination that also results in a loss of taste. One problem remains. Even though the traditional breeding time of 25 years has been reduced to one year, the plants that were transformed were C. canephorea and not Coffea arabica.

The Agrobacterium-mediated transformation protocols differ from one plant species to other and, within species, from one cultivar to other. A. tumefaciens as a plant pathogen naturally infects the wound sites in dicotyledonous plants and induces disease known as crown gall, and this bacterium has been widely used for the introduction of foreign genes into plants and consequent regeneration of transgenic plants.

However, A. tumefaciens-mediated gene transfer is quite difficult in most of the plant species. The success of genetic transformation via A. tumefaciens is limited because plant's defence mechanism will be active when pathogen attacks. That is why manipulations of the plant and bacterium and physical conditions have been applied to increase the virulence of bacterium and to increase the transformation efficiency.

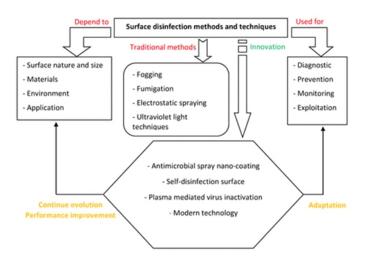
# Surface Disinfection to Protect against Microorganisms: Overview of Traditional Methods and Issues of Emergent Nanotechnologies

Sterilization methods for individuals and facilities are extremely important to enable human beings to continue the basic tasks of life and to enable safe and continuous interaction of citizens in

Sterilization methods, their availability in gatherings, and the efficiency of their work are among the important means to contain the spread of viruses and epidemics and enable societies to practice their activities almost naturally.

The objective was to understand how to control their microbial viability. One of the common ways of infection is contacting surfaces containing droplets from a person who is infected with the viruses by coughing, talking, or sneezing. However, it was revealed that it depends on the material from which the surface is made from hours to several days. The most important factor of surface decontamination is the surface type and properties and the substrate quality. Surface type is related to the goods material properties ((i) source (nature such as fruits and legume, hard or soft (such as a polymer)), (ii) permeability, (iii) stiffness, (iv) hydrophilicity, etc.). Besides, surface size related to the dimension of equipment and facilities is the second parameter which conditions the choice of the most suitable method.

According to the World Health Organization, cleaning and disinfecting are two effective ways to stop this virus and eliminate it from surfaces. Disinfection can be carried out either with traditional methods like liquid solutions such as sodium hypochlorite (bleach/chlorine) and alcohol at 70-90%, or ultraviolet (UV) radiation which has the capacity to destroy the DNA of the virus.



Surface disinfection depends on several parameters and is widely used for cleaning, disinfection, and sterilization. Thanks to innovative technologies, protocols, and creativity, new methods have been developed with a continued evolution to improve performance. Despite the effective solutions given by traditional methods of surface disinfection, modern nanotechnology has proven to be an emergent innovation to protect against viruses.

### Traditional Methods: -

### Fogging Method

There are different fogging disinfection systems. The difference exists in the used chemical product: Sterilox hypochlorous acid, tartaric acid solution, liquid peracetic acid, alkyl amine/peracetic acid, the 7.5% of the hydrogen peroxide, and 0.2% of the chlorine dioxide solutions. Surfaces are huger and more extended such as hospital-acquired infections. These locations are fertile zones for virus development. Consequently, increased transmission of pathogens is noticed, from patient to patient or patient to hospital staff. The use of hydrogen peroxide vapor in clinical surfaces provided better results compared to the steam vapor system. The use of airborne hydrogen peroxide gives excellent results in surgical wards, single isolation rooms, and bathrooms. However, this technique is obliged to the removal of patients and has a high acquisition cost and increased room turnover time.

### **Fumigation**

Fumigation is the operation of introducing a gas or a substance giving rise to a gas in the atmosphere of a partially or completely closed enclosure with a view to destroying so-called "harmful" living organisms. It was formerly the combustion of plants producing vapors charged with the active ingredients of the plant.

### Wide-Area or Electrostatic Spraying Techniques

Most surface areas are uncharged or negative. The surface disinfection using electrostatic application consists of using a disinfectant registered with the Environmental Protection Agency on the surfaces with electrostatic spraying according to Coulomb's law. Electrostatic spraying can minimize preventable infections in large surfaces in hospital environments, improving patient experience and increasing hospital revenues. There are many elements that can affect its efficiency: the charge/mass ratio, spraying distance, and liquid deposition efficiency target.

### Ultraviolet Light

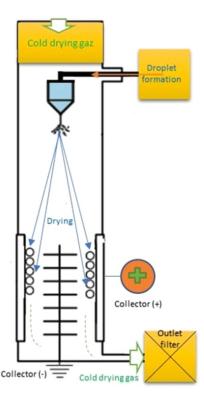
The World Health Organization approved that ultraviolet (UV) is effective as a no-touch technology in the case of healthcare settings. Ultraviolet light technology in the field of disinfection can reduce contamination compared with manual techniques, especially in the medical field and hospital environment. However, this method is effective from the point of view of sterilization results and limited to closed places without the presence of people because, as known, UV has a negative effect on the human skin and eyes.

### **Innovation in Surface Disinfection Method Antimicrobial Spray Nanocoating**

Dry nanospray is one of the most innovative technologies and has many advantages in terms of its ability to produce nanoparticles with much smaller droplets and a narrow area distribution. Spray drying can produce different forms of particle shapes. Nanospray drying allows volume reduction, defined particle size, changing chemical, and physical properties with chemical and biological stability and high specific surface. Additionally, nanospray drying offers easy dosage administering and handling. It was initially utilized in the pharmaceutical field.

The nanospray drying process consists of six fundamental steps.

The **first stage** is heating the inlet air to the desired temperature not to exceed 220 °C. The **second stage** is droplet formation using a two-fluid nozzle or ultrasonic spray head. Before the collection of the particle using cyclone technology or electrostatic particle collector, the drying step between drying gas and



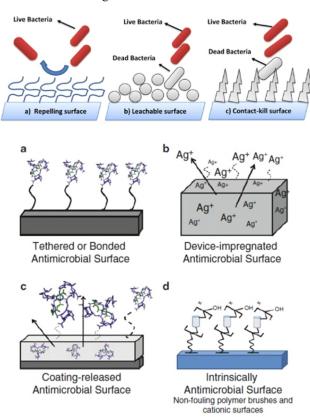
sample droplets is important. Then, the finest particles were collected to protect the user and environment using an outlet filter. Finally, drying gas delivered by aspirator or with compressed air. A piezoelectric actuator vibrates a small replaceable spray cap, containing nanometric holes, to generate droplets. The latter leads to rapid vertical movement of the spray mesh, ejecting many droplets in size accurately through the holes in the drying chamber. The size of the droplets depends on the size of the holes and the physicochemical characteristic of the disinfectant.

The flow of concurrent drying gas directs the particles to the electrostatic particle collector. The electrostatic particle collector can capture nanometric particles (<1  $\mu m$ ) with separation efficiency. The particles are gently removed from the inner surface of the collecting electrode cylinder using the particle scraper. The sizes of the particles formed through encapsulation are nano (<1  $\mu m$ ) and known as nanocapsules. The smaller the particles, the better the solubility of the encapsulated liquids. The droplets obtained at the nanoscale lead to an increase in the volume distribution in the surface, which generally occurs by reducing the size of the drops, thereby increasing the particles' efficiency. The cost reduces by using nanodrop sterilization instead of other sprayers.

### **Self-Disinfection Surface**

The most emergent is the self-defensive antimicrobial known as the self-disinfection surface. It consists of the creation of a bacteria cell wall to resist the adhesion of bacteria or to kill bacteria by chemical or physical changes. This technique is classified in three categories: surface resisting the bacteria attachment using the photoactivation process for example, surface leaching antibacterial agents called intrinsically antimicrobial ability, and surface killing or delivery bacteria by contact using antimicrobial loading, based on the coating technology or incorporation process. The most important

functional materials are photoactive building monocrystals (CuInZn4S6), both TiO<sub>2</sub> and SiO<sub>2</sub> nanocoatings, antimicrobial peptide substance, and membrane-active polycations. Research have successfully clarified the long-term efficacy, durability, and mechanical stability of the antiadhesive action in relation to the environmental conditions, the scalability of production, and the real cost of these settings.



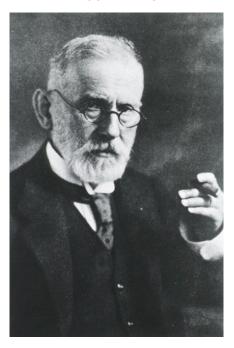
### Plasma-Mediated Virus Inactivation

Plasma generation is attributed to the creation of anodized gas which contains reactive chemical elements such as electrons, ions, and charged species. Nitrogen-gas plasma and UV radiation have the most effective impact on viruses and about a six-log reduction/inactivation performance for bacteria. Other techniques used innovative methods by radiofrequency plasma treatment using an  $Ar/O_2$  gas mixture. They concluded that plasma can not only disinfect surfaces but also degrade toxins. The mechanisms of degradation are directly related to the used gases as well as the method used to generate plasma and the target microorganism.

## **Modern Technology for Healthcare Environment and Surface**

Considered as the highest exposed environment to microorganisms, healthcare workers and relative equipment and devices are suspected to be what is called high-level disinfection. The objective was to protect the staff and to prevent the transmission of infectious agents. Their use was associated with an improvement in the cleaning and disinfection of high touch surfaces with a specific protocol and guidelines considering (i) the potential of the environmental surfaces to transmit infection, (ii) mode and duration of contact, and (iii) toxicologic risk assessment. The use of ultraviolet germicidal irradiation as a disinfection method, kills airborne viruses using shortwavelength ultraviolet.

### **Paul Ehrlich**



German biochemist Paul Ehrlich (1854-1915) developed a chemical theory to explain the body's immune response and did important work in chemotherapy, coining the term magic bullet. Ehrlich received the Nobel Prize in Physiology or Medicine in 1908.

In 1906 Ehrlich prophesied the role of modern-day pharmaceutical research, predicting that chemists in their laboratories would soon be able to produce substances that would seek out specific disease-causing agents. He called these substances "magic bullets." Ehrlich himself met with signal successes in the emerging fields of serum antitoxins and chemotherapy.

### Early Work with Dyes

Ehrlich was born near Breslau-then in Germany, but now known as Wrocław, Poland. He studied to become a medical doctor at the university there and in Strasbourg, Freiburg im Breisgau, and Leipzig. In Breslau he worked in the laboratory of his cousin Carl Weigert, a pathologist who pioneered the use of aniline dyes as biological stains.

Ehrlich became interested in the selectivity of dyes for specific organs, tissues, and cells, and he continued his investigations at the Charité Hospital in Berlin. After he showed that dyes react specifically with various components of blood cells and the cells of other tissues, he began to test the dyes for therapeutic properties to determine whether they could kill off diseasecausing microbes. He met with promising results using methylene blue to kill the malaria parasite.

### Antitoxins from Blood Sera

After a bout with tuberculosis and his subsequent cure with tuberculin therapy, developed by fellow German Robert Koch, Ehrlich focused his attention on bacterial toxins and antitoxins. At first he worked in a small private laboratory, but then he was invited to work at Koch's Institute for Infectious Diseases in Berlin. The post-Pasteur era was an exciting time to be looking for cures and preventives, and Koch's Institute was one of the best places to be.

Among Ehrlich's new colleagues were Emil von Behring and Shibasaburo Kitasato, who had recently developed "serum therapies" for diphtheria and tetanus. Whereas Louis Pasteur's vaccines and Koch's tuberculin were made from weakened bacteria, these new serum therapies used blood serum, or cellfree blood liquid, extracted from the blood of naturally or artificially immunized animals to induce immunity. Von Behring and Kitasato evolved the concept of "antitoxin" to explain the immunizing properties of sera.

One of Ehrlich's jobs at the institute was to make von Behring's diphtheria antitoxin in quantity and later to review the quality of the product produced by the chemical-pharmaceutical company Hoechst. In carrying out this work, he determined how to boost immunity systematically and how to produce high-grade sera.

In recognition of Ehrlich's accomplishments and of his promise as a researcher, in 1896 the Institute for Serum Research and Serum Testing was established for him in a Berlin suburb. In 1899 the institute moved to Frankfurt to more suitable quarters and was renamed the Royal Prussian Institute for Experimental Therapy.

### A Nobel Prize and Magic Bullets

In 1908 Ehrlich shared the Nobel Prize in Physiology or Medicine with Élie Metchnikoff for their separate paths to an understanding of the immune response: Ehrlich presented a chemical theory to explain the formation of antitoxins, or antibodies, to fight the toxins released by the bacteria, while Metchnikoff studied the role of white blood corpuscles (phagocytes) in destroying bacteria themselves. By that time most scientists agreed that both explanations of the immune system were necessary.

Early in his career Ehrlich began to develop a chemical structure theory to explain the immune response. He saw toxins and antitoxins as chemical substances at a time when little was known about their exact nature. Up to that time, those scientists who were synthesizing therapeutic agents came at their tasks with few hypotheses about where and how these agents interacted with living systems.

Ehrlich supposed that living cells have side chains—a shorter chain or group of atoms attached to a principal chain in a molecule—much in the way that dye molecules were known to have side chains that were related to their coloring properties.

These side chains can link with particular toxins. According to Ehrlich, a cell under threat from foreign bodies grows more side chains, more than are necessary to lock in foreign bodies in its immediate vicinity. These "extra" side chains break off to become antibodies and circulate throughout the body. It was these antibodies, in search of toxins, that Ehrlich first described as magic bullets.

### Chemotherapy

Serum therapy was for Ehrlich the ideal method of contending with infectious diseases. In those cases, however, in which effective sera could not be discovered, Ehrlich would turn to

### In Profile



synthesizing new chemicals, informed by his theory that the effectiveness of a therapeutic agent depended on its side chains. These "chemotherapies" were to be the new magic bullets.

In Frankfurt, Ehrlich turned from his work on serum therapy to chemotherapies and dyes. First targeting the protozoa that were known to be responsible for certain diseases, such as sleeping sickness, he and the Japanese bacteriologist Kiyoshi Shiga synthesized trypan red as a highly effective cure for that disease.

In 1906 Georg-Speyer-Haus, a research institute for chemotherapy, was established with its own staff under Ehrlich's direction. Soon this institute and the Hoechst and Cassella chemical companies reached an agreement that gave the companies the right to patent, manufacture, and market preparations discovered by Ehrlich and his colleagues.

The companies further agreed to supply chemical intermediates for the syntheses that the staff of the institute would undertake.

Salvarsan

The researchers, now including an organic chemist, Alfred Bertheim, and a bacteriologist, Sahashiro Hata, broadened the targeted microorganisms to include spirochetes, which had recently been identified as the cause of syphilis.

Beginning with an arsenic compound, atoxyl, in three years' time and three hundred syntheses later—for that day an amazingly large number—they discovered Salvarsan (1909).

Salvarsan was first tried on rabbits that had been infected with syphilis and then on patients with the dementia associated with the final stages of the disease. Astonishingly, several of these "terminal" patients recovered after treatment. More testing revealed that Salvarsan was actually more successful if administered during the early stages of the disease. Salvarsan and Neosalvarsan (1912) retained their role as the most effective drugs for treating syphilis until the advent of antibiotics in the 1940s.



# Jokes



A man receives a phone call from his doctor.

The doctor says, "I have some good news and some bad news."

The man says, "Ok, give me the good news first."

The doctor says, "The good news is, you have 24 hours to live."

The man replies, "Oh no! If that's the good news, then what's the bad news?"

The doctor says, "The bad news is, I forgot to call you yesterday."

A: I have the perfect son.

B: Does he smoke?

A: No. he doesn't.

B: Does he drink whiskey?

A: No, he doesn't.

B: I guess you really do have the perfect son. How old is he?

A: He will be six months old next Wednesday.

My friend said he knew a man with a wooden leg named Smith. So, I asked him, "What was the name of his other leg?"

Teacher: "Nick, what is the past participle of the verb to ring?"

Nick: "What do you think it is, Sir?"

Teacher: "I don't think, I Know!"

Nick: "I don't think I know either, Sir!"

Two boys were arguing when the teacher entered the room.

The teacher says, "why are you arguing?"

The other boy answers, "we found a ten-rupee bill and decided to give it to whoever tells the biggest lie."

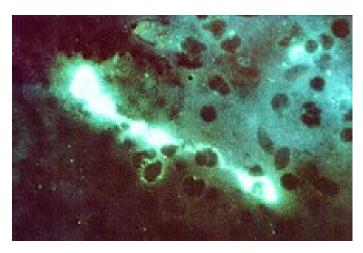
"You should be ashamed of yourselves," said the teacher,

"When I was your age, I didn't even know what a lie was."

The boys gave the ten rupees to the teacher.



# Chlamydia psittaci



Chlamydiae are ubiquitous, obligate intracellular bacteria that grow in eukaryotic cells and are responsible for a wide spectrum of human diseases. *Chlamydia Psittaci* (Cp) is the etiologic agent of psittacosis, which represents an infection caused by exposure to infected animals.

Chlamydophila psittaci (formerly Chlamydia psittaci) is a small bacterium that undergoes several transformations during its lifecycle. It exists as an elementary body (EB) between hosts. The EB is not biologically active but is resistant to environmental stresses and can survive outside a host. The EB travels from an infected bird to the lungs of an uninfected bird or person in small droplets and is responsible for infection. Once in the lungs, the EB is taken up by cells in a pouch called an endosome by phagocytosis. However, the EB is not destroyed by fusion with lysosomes, as is typical for phagocytosed material. Instead, it transforms into a reticulate body and begins to replicate within the endosome. The reticulate bodies must use some of the host's cellular machinery to complete their replication. The reticulate bodies then convert back to elementary bodies, and are released back into the lung, often after causing the death of the host cell. The EBs are thereafter able to infect new cells, either in the same organism or in a new host. Thus, the lifecycle of C. psittaci is divided between the elementary body which is able to infect new hosts, but cannot replicate, and the reticulate body, which replicates, but is not able to cause new infection.

Psittacosis represents a zoonotic bacterial infectious disease caused by the obligate intracellular organism, Chlamydia psittaci. Psittacosis, also called parrot fever and ornithosis, is transmitted from contact with infected birds and causes a wideranging spectrum of disease and severity. Birds serve as the major epidemiological reservoir.

Psittacosis can affect any age group and gender, but incidence tends to peak in middle age, with an age range of 35 to 55. Psittacosis is regarded as a rare zoonotic infection. Thus, there is a decreased awareness of this disease entity among the public and health care professionals. When coupled with the need for specialized testing, underdiagnosis of psittacosis is likely when examining reports of prevalence and incidence. Individuals with exposure to pet shops, veterinary hospitals, bird exhibitions, and occupational exposure in the poultry industry are considered at

the highest risk of contracting the disease.

Psittacosis primarily manifests as respiratory symptoms, though they can vary greatly. The infection can spread hematogenously to impact several organ systems following replication in the respiratory system. Initially, it's frequently described as an influenza-like illness with fever, chills, headaches, and coughing. Case studies have shown that the infection can range from an asymptomatic state to a fulminant invasive disease with an average incubation period of 5 to 14 days.

Symptom onset is typically abrupt, with a headache cited as the most prominent complaint, in addition to fever, myalgias, nausea, vomiting, diarrhea, and cough. Studies have cited the presence of a severe headache as being a characteristic feature. Other signs of psittacosis that have been documented include altered mental status, mild neck stiffness, photophobia, hepatosplenomegaly, and pharyngitis. Psittacosis can affect multiple organ systems, and a multitude of manifestations have been reported in case reports of psittacosis.

Psittacosis is noted for a normal white cell count with toxic granulation or a left shift on laboratory testing. markers of acute inflammation, such as the erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP), are often elevated. Creatine level is usually elevated, and hyponatremia is quite common. Liver function testing, specifically aspartate and alanine aminotransferase and gamma-glutamyl transpeptidase (AST, ALT, GGT), are also variably elevated. Chest imaging is abnormal in the majority of cases, most often revealing lobar infiltrates.

Traditionally, serologic testing has been used to confirm suspected cases of psittacosis. Available serologic tests for psittacosis include complement fixation (CF) testing and microimmunofluorescent (MIF) antibody testing with paired sera. MIF testing is more sensitive and specific for C. psittaci when compared to CF. psittaci when compared to CF. A MIF test with an immunoglobulin (Ig)M antibody titer greater than or equal to 16 or a four-fold increase in antibody titer two weeks apart is considered diagnostic for psittacosis. CF testing does not differentiate between chlamydial species and is much less specific for psittacosis. The diagnostic threshold for CF is a fourfold increase in antibody titer two weeks apart. Monoclonal antibody techniques are being developed for diagnosing psittacosis, but studies demonstrating the sensitivity and specificity of these techniques are still lacking. DNA-based polymerase chain reaction (PCR) techniques have also been developed. They show promise as rapid diagnostic tools for diagnosing psittacosis but are not widely available. An emerging diagnostic tool is metagenomic next-generation sequencing (mNGS), which was shown to increase the rate of pathogen identification in severe community-acquired pneumonia cases from 40.8% (when using PCR) to 74.2% (when using mNGS). A 2022 study reported higher sensitivity of mNGS than PCR in blood for diagnosing psittacosis because mNGS can detect pathogens with an extremely low DNA load in the sample. Culture is the most specific and accurate method to diagnose psittacosis; however, it should be noted that isolation of C. psittaci requires a biosafety level three facility due to the risk of transmission to laboratory personnel.

Treatment for this bacterial infection is based on intracellular activity, pharmacokinetics, and evidence from clinical trials that recommend tetracycline antibiotics, particularly doxycycline, as the preferred treatment. Case studies have shown that with treatment, most infected individuals will have an improvement in fever and clinical symptoms within 48 hours. In pregnancy and in

patients where doxycycline is contraindicated, the infection is best treated with macrolide antibiotics. Macrolides remain the agents of choice in children with mild to moderate infection. However, as has been proposed for the treatment of Rocky Mountain spotted fever, in cases where the benefit of treatment with doxycycline outweighs the potential risk, especially if the alternative therapy is found to be ineffective against psittacosis, a tetracycline, such asdoxycycline can be considered in pediatric patients.

# You leave a 'microbe fingerprint' on every piece of clothing you wear—and it could help forensic scientists solve crimes



When you think of a criminal investigation, you might picture detectives meticulously collecting and analyzing evidence found at the scene: weapons, biological fluids, footprints and fingerprints. However, this is just the beginning of an attempt to reconstruct the events and individuals involved in the crime.

At the heart of the process lies the "principle of exchange" formulated by the French criminologist Edmond Locard in the early 1900s, which states that "every contact leaves a trace." The transfer of materials between the parties involved in a crime (the victim, the perpetrator, objects, the environment) forms the basis for reconstructing the events.

In Locard's time, these traces were typically things you could see with a magnifying glass or microscope, such as pollen, sand and fibers. However, such evidence is limited because much of it is not directly associated with a specific individual.

In the latest research published in Genes, it was shown how the population of bacteria on a person's skin leaves traces on the clothes they wear—and how these traces last for months and can be used to uniquely identify the wearer.

### Microbial traces

Imagine a crime scene where an investigator finds a victim and a piece of clothing that doesn't belong to them. Pollen or grains of sand might help the investigator find out where it came from, but what about identifying the owner of the clothing?

Skin cells, hairs and biological fluids are good contenders.

However, another thing very specific to an individual is the unique community of microorganisms on and within their body. These microbes are specific to different parts of the body, can persist over long periods of time and can be transferred to other people and to the environment. This makes them useful to address a variety of questions in forensics.

"Forensic microbiology" got its start in the early 2000s, as scientists set out to find ways to defend against bioterrorism. Today forensic microbiology is used to identify individuals after death, understand what their health was like before they died, determine how and why people have died, how long it has been since they died, and where they came from. In a nutshell, today's update on Locard's principle is that "every contact leaves a microbiological trace."

### The 'touch microbiome'

While this principle has been established, we still want to know more about how much of an individual's microbiome is transferred to their surroundings. We also need to know how long it persists, and whether certain microbes may be more useful than others for identification. We also want to understand how microbial traces may be contaminated by other items or the environment, and how different receiving surfaces affect microbial populations.

In 2021, two of the authors (Procopio and Gino) and colleagues at the University of Central Lancashire in the UK and the University of Eastern Piedmont in Italy first described the "touch microbiome"—the unique bacterial populations on individuals'

skin. This work also studied how these bacteria could be transferred and persist for up to a month on non-porous surfaces, such as a glass slide, in uncontrolled indoor surroundings.

This team also analyzed DNA from samples belonging to dead bodies from old cases, which had been frozen for up to 16 years. They were able to identify specific populations of microbes linked to the manner of death and the decomposition stage of the bodies. This showed the microbial signature can be used to improve our understanding of cold cases when DNA extracts are still available.

### **Tracing T-shirts**

In the most recent work, the third author (Magni) joined the collaboration to improve the potential of individual identification from clothes, items often collected as evidence at the crime scene. In this study, cotton T-shirts were worn by two individuals for 24 hours in Australia. The T-shirts were then placed in a controlled environment for up to six months, alongside unworn items used as controls. Samples from both worn and unworn T-shirts were taken at various points in time and frozen. The samples were then shipped (still frozen) to Italy for microbial DNA extraction. Next, sequencing was conducted in the UK, with the goal of identifying the microbial species present in the samples.

Results showed the two volunteers transferred distinct and recognizable microbes onto the clothing, each unique to the respective individual. Additionally, they could distinguish

between worn and unworn items even after an extended period of time. The microbiome remained stable on the worn garments for up to 180 days.

It was also observed the transfer of specific bacteria from the worn items to the unworn ones stored closest to them, showing the possibility of microbe transfer between items.

### Learning more from clothes

Clothes at any crime scene can provide key evidence for the investigation process.

They can aid in profiling individuals by revealing indicators of gender, occupation, income, social status, political, religious or cultural affiliations, and even marital status.

Additionally, they can provide clues regarding the manner of death, the location of the crime, and in certain cases, even support the estimation of the time since death.

Clothes play a crucial role in reconstructing events associated with the crime and establishing the identity of individuals involved.

This research shows clothing can provide even more evidence. The discovery of unique microbiomes capable of identifying individuals from clothing marks a significant stride forward.

## Personal Health Bringing Good Hygiene Home

"Hygiene" refers to conditions or practices by which people maintain or promote good health by keeping themselves and their surroundings clean.

### HYGIENE NEEDS IN THE HOME

In the home, the first line of defence against infectious disease is **cleaning** and **disinfecting**.

**Cleaning** is the mechanical removal of dirt and soil from an object or area. Detergents and water are the preferred products for cleaning.

**Disinfection** is the chemical inactivation or killing of microbes. Products containing substances such as alcohol, sodium hypochlorite bleach, quaternary ammonium compounds, and phenolics, can be disinfectants, depending on the formulation and use of the product.

### Microbes in the Home: Where they are found

Microbes can thrive wherever there is an ample source of nutrients and water. Studies have shown that areas in the kitchen, bathroom, and laundry can serve as reservoirs for the growth of microbes. Bacteria, like *Pseudomonads* and *E. coli*, as well as molds, prefer areas with high humidity, such as drains, sinks, shower stalls, toilets, and basements. Other bacteria such as *Staphylococci* and *Bacilli* prefer drier surfaces like counter tops or skin.



### Disinfectant Cleaner for floor and hard surfaces

### Transfer of Microbes Elsewhere Around the Home

Other surfaces around the home can be sites of bacterial and viral transfer.

Infection from the transfer of bacteria and viruses from common household articles to the hands are possible from daily contact with these objects. Transmission from door handles, telephone receivers, faucet handles, and sponges has been shown to occur, with transfer to hands from hard, nonporous surfaces being highest.

### Controlling infectious microbes at home:

The Kitchen So much activity and food preparation take place in the kitchen that it is a virtual hot spot for bacterial growth and spread. When counter tops, cutting boards, and other kitchen surfaces are not properly cleaned and/or disinfected, microbes survive and proliferate. Kitchen studies frequently follow Gram negative bacteria like *Enterobacteria*, *Campylobacter*, and *Salmonella*, as these bacteria are sometimes found as natural contaminants on foods. If they are not eliminated during cooking, they can cause severe food poisoning.



Disinfectant glass and surface cleaner

### **Sponges and Dishcloths**

Sponges and dishcloths used with hypochlorite disinfection products have significantly lower bacterial contamination.

### In the Bathroom

In the bathroom, splashing and aerosol droplets are responsible for transferring contamination from toilets and sinks to surrounding areas in the bathroom. Using a chlorine bleachbased, in-toilet block effectively reduces the level of contamination in the toilet.



Toilet bowl cleaner

### In the Laundry

Reductions in infection risk have been associated with the use of hot water and bleach during laundering. Warmer washing temperatures, such as 1318° F (558 °C), are effective in reducing bacterial levels. Colder washing temperatures may increase the cross-contamination rate of articles that are washed together.

Sodium hypochlorite bleach is effective in reducing bacterial counts when either hot or cold water is used.



Aldehyde free disinfectant cleaner for linen

### Hand hygiene: A timeless defence against infection

Cleaning hands is very important in preventing infection. Good hand hygiene practices lead to reduced risk of infection. The major benefits of hand hygiene for the public are the removal of infectious agents found on hands and spread by the faecal-oral route, from the respiratory tract, and from contaminated food. Handwashing is necessary before and/or after behaviours that are associated with microbial contamination, especially using the toilet, diapering, and preparing or eating food.

For cleaning hands, there are generally three types of products available:

### 1. Plain Soaps

Generally, plain soaps do not kill microorganisms, but rather wash them off with the soap, with the help of friction and rubbing. As a result, most microorganisms picked up in daily life are removed. Handwashing with plain soap and water for 15 seconds reduces skin bacterial counts by 50 to 90% and washing for 30 seconds reduces counts by 90 to 99%.

### 2. Antibacterial Soaps/Handwash

In addition to washing off microorganisms, antibacterial soaps contain ingredients that inhibit the growth of and/or kill germs on the hands. Antibacterial soaps can also reduce bacteria on the skin and the rates of superficial skin related infections. Triclocarbon and triclosan are common antimicrobial active ingredients used in soap.

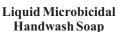
### 3. Hand Sanitizers (nonsoap products)

Hand sanitizers are non-detergent-based, antibacterial products in the form of hand rinses, gels, or wipes, which usually contain alcohol as the antibacterial ingredient. They rapidly kill a broad spectrum of microbes, including bacteria, viruses, and fungi.

Good hand hygiene, surface cleaning and disinfection, and laundering practices can lessen the chances of spreading these diseases. Microbes can spread and grow in the home, particularly

in the kitchen, bathroom, and laundry areas. The highest counts of microbes in the kitchen and bathroom are found in wet areas around the sink, in sponges and cloths used for wiping and/or drying kitchen surfaces, and in the areas around the bathroom sink. Water temperature can influence the survival of microbes during dishwashing and laundry practices. For the laundry, drying is the most reliable method for destroying microbes. Attaining maximal reduction in bacteria in both the machine and fabrics depends on the use of bleach or disinfecting detergents, as well as the water temperature.







Alcoholic Handrub Gel with Moisturizer

To maximize the removal of microbes, care should be taken to use disinfecting products according to their instructions. It is increasingly important that proper home hygiene and cleaning practices are followed to reduce the risk of spreading disease



**Personal Care Antiseptic Solution** 





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