Plasma-based Cleansing Techniques for Biopharmaceutical Research: An Introduction to and Explanation of the Technology

Introduction

Recently, IonField Systems™ introduced the TipCharger™, a device that generates a self-contained, low-temperature, atmospheric plasma for cleaning pipette tips used in automated life science laboratories. The patented technology uses a dielectric barrier discharge (or gap plasma) that is generated in small, well-like cavities and conforms to a standard SBS footprint. Currently, the device is passive in that plasma generation is triggered via the insertion of pipette tips, making it easy to install and integrate with most automated liquid handlers. The result is the ionization and molecular breakdown of biological and organic contaminants on both the exterior and interior surfaces of the tips. Unlike traditional solvent-based pipette tip wash methods, plasma as a state of matter (neither a gas nor a liquid) generates no solid or liquid waste and only small amounts of gaseous by-products. Unlike traditional solvent-based tip washing methods, plasma cleaning does not simply dilute contaminants, but removes them at a molecular level, allowing laboratories to reuse polypropylene tips and reduce the number of tip boxes needed. This paper discusses the mechanism and application behind this revolutionary cleaning method.

Background and early studies

The term plasma was first adopted by Nobel Prize winning chemist Irving Langmuir in 1927 to describe the properties of gases ionized by high voltage to form free electrons and positive ions in a neutral background gas. These ionized gases conduct electricity and are strongly affected by magnetic fields. Although common in nature, it is estimated that over 99% of all matter in the known universe is in the form of plasma,lightning and flames are the only two naturally occurring forms on Earth. Through advancements in physics and chemistry as well as the harnessing of alternating currents, the breadth of man-made plasmas continues to widen.

Most of the early studies on man-made plasmas were performed using vessels either under low-pressure or vacuum -filled with inert gases providing a ready source of electrons and positive ions. Through the application of a high voltage to metal electrodes within these vessels, electrons are accelerated towards the anode (positive electrode) while positive ions move toward the cathode (negative electrode). Further, by constantly and simultaneously switching the polarity of each electrode, electrons and charged ions continuously move between poles forming what is known as a dielectric barrier discharge.

In the presence of a gas, free electrons and activated ions collide with other atoms and molecules present in the gas field resulting in a sustained and steady-state plasma. These experiments ultimately led to the development of neon signs, streetlights and fluorescent light bulbs (Figure 1).

Figure 1. *Man-Made Plasma Lights Up the Las Vegas Sky*

Atmospheric-Pressure 'Cold' Plasma

Although originally reported by Werner von Siemens in 1857 as a method for generating ozone, plasma technology remained underdeveloped until modern materials allowed for the production of more effective and efficient dielectric materials. Over the past thirty years advancements in dielectric materials, i.e the development of ceramic coated electrodes, enabled the use of insulating layers that allow for stable discharges to occur. The result is the generation of atmospheric pressure plasmas that form in the absence of inert gases or a vacuum.

The induction of atmospheric plasma begins with the capturing of free electrons to charged dielectric plates. Through changes in polarity, these electrons are forced to mobilize between a set of plates resulting in an electron avalanche (Figure 2).

By drawing air in-between these plates and through the electron field, the components of air are bombarded with electrons, causing the fracturing and/or activation of CO_2, H_2O, O_2, and N_2. The result is a 'cold' or 'non-thermal' plasma field where most of the input energy is channeled to the electron component of the plasma while the resulting ions and neutral components remain at or near room temperature.

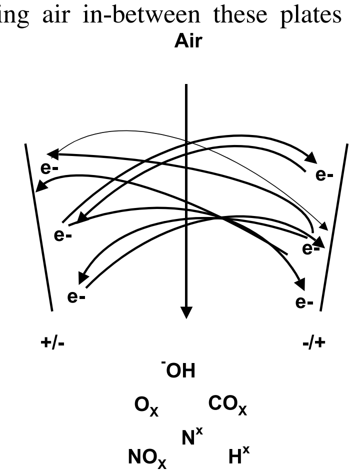


Figure 2: *Generation of atmospheric plasma and the subsequent ionization/activation of room air.*

In addition, highly energetic metastable atomic species are generated.during the process, These species have the appropriate number of electrons, but are displaced to higher shells and exhibit high reactivity and some selectivity in their reaction capabilities.

The plasma ions and metastable atomic species react rapidly with biologicals and organic solvents to facilitate their breakdown and eventual removal.

In general, there are four prototypical reaction mechanisms that play a role in the removal of material from a contaminated surface by plasma cleaning. Oxidation (the addition of oxygen to a compound to form an oxide) of the contaminant is the most common mechanism. Reduction of the contaminant (the removal of oxygen) is another possible route. Electron- and ion- induced decomposition processes are two other reaction routes.

Common to all processes is the fact that electron-driven dissociation and ionization of the plasma constituents are critical in the formation of reactive radicals and ions. In laymen's terms, the various species and mechanisms of action provide for both electrophilic and nucleophilic attack of organics at the molecular level (Figure 3).

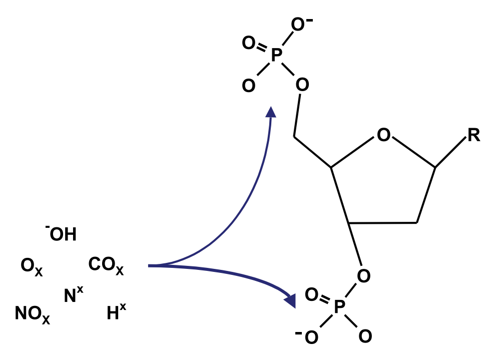


Figure 3: *Electron- and ion-driven constituents provide multiple opportunities for oxidation and reduction reactions via electrophilic and nucleophilic attack against Deoxyribonucleic Acid.*

Cold Plasma in the Automated Laboratory

At present, most analytical methods used in the biopharmaceutical laboratory require at least one liquid transfer operation, and nearly 100% of those using viable cells or intact organisms call for the use of sterile, disposable pipette tips in order to minimize effects of cross-contamination.

Exhaustive solvent-based washing procedures using organic or caustic materials have been the only alternative to the use of disposable pipette tips until IonField Systems introduced cold plasma technology to the lab as a safe, reliable and practical cleaning method using the TipCharger system.

Summary

Safe

Since the arc-free dielectric barrier discharge plasma is produced by the TipCharger at near room temperature, it draws very little power. The induced plasma field is fully enclosed within a self-cleaning, inert chamber. All by-products of the cleaning process are effectively trapped by the TipCharger's internal filtration system, and vented to an external exhaust system.

Reliable

With essentially no moving or serviceable parts, the TipCharger cleaning station provides a robust solution for cleaning plastic disposable, and metal or ceramic tips. The unique combination of oxidizing free radicals and metastable atomic species formed within the cleaning station's cold-plasma field promotes the nearly instantaneous conversion, volatilization and ultimate removal of potential chemical or biological contaminants from pipetting surfaces.

The metastable species are readily pipetted in and out of a pipette when the opening of the pipette tip is within the plasma field (Figures 4 and 5).

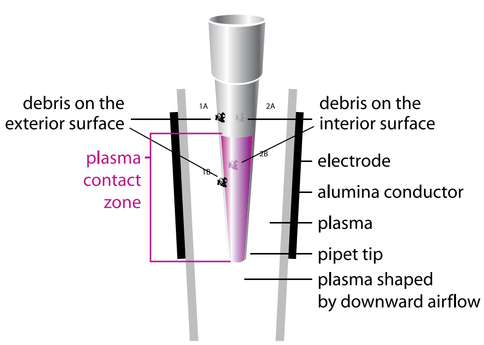


Figure 4 *Plasma Cleaning Action In and Around A Pipet Tip*

This technique will actively cleanse the inner, wetted surfaces of liquid transfer devices such as cannulae, pin tools and disposable pipette tips. Plasma will flow easily into all microscopic surface imperfections and cavities, reducing tip-to-tip variations due to random surface properties.

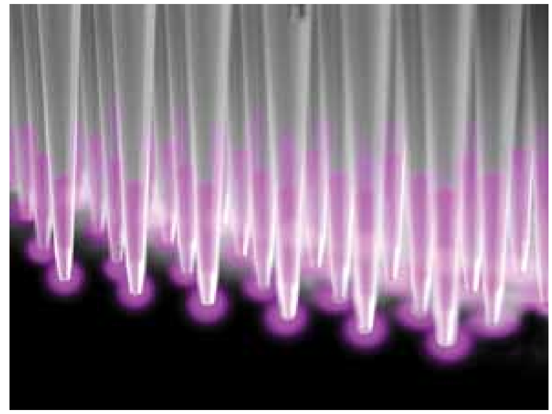


Figure 5. *Artist's drawing of plasma cleaning action while tips are inserted into the TipCharger.*

Practical

By cleaning pipette tips on-line and in-process, integration of the TipCharger offers multiple benefits to the automation process. Throughput is significantly increased, which is an important consideration for labs operating at high volumes and for business models based on per-test pricing. Direct and indirect costs for tip and solvent purchase, storage, use and disposal are dramatically decreased when using the TipCharger. Plasma cleaning does not simply dilute contaminants, but removes them at a molecular level, with the important benefit of eliminating contamination and carryover and improving overall process results.

In summary, integrating cold plasma cleaning technology within automated laboratories offers an opportunity to deliver better science as well as increase the efficiency and productivity of liquid handling processes.

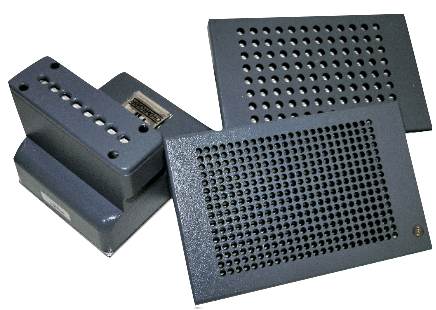
Integrating the TipCharger into Automated Assays Provides the Following Benefits:

Confidence TipCharger cleans better than any other washing technology - in most applications the TipCharger will clean to background, so there is no difference between plasma cleaning and a new tip.

Cost Benefit TipCharger can save up to 98% on the cost of the disposable tips and extends the life of fixed tips.

Speed Incorporating the TipCharger System can result in a time savings of 10-30 seconds for every microplate processed or rack of tips cleaned.

Convenience Clear away the clutter and save time: Integrating the TipCharger System eliminates the need to store cases of new pipette tips and dispose of racks of hazardous used tips.



TipCharger Plasma Cleaning Stations
Available in 8, 96 and 384 channel versions

USA and ROW
IonField Systems
3411 Silverside Road
Hanby Building
Wilmington, DE 19810 USA

Call: +1.302.442.6901
Fax: +1.302.792.7407
Email: info@ionfieldsystems.com
www.ionfieldsystems.com

Europe and Middle East
IonField Systems Limited
7 Paynes Park, Hitchin
Hertfordshire, SG5 1EH,
UK

Call: +44 (0) 1462 429 727
Fax: +44 (0) 1462 429 798
Email: info@ionfieldsystems.com
www.ionfieldsystems.com

About the TipCharger™

The TipCharger Cleaning System replaces existing wash stations and easily integrates with most existing and new automation platforms. The system utilizes a low temperature, atmospheric pressure plasma process that cleans metal and plastic pipette tips and pin tools. Treated surfaces are clean, dry and have uniform surface properties.

The TipCharger cleaning process reduces the incidence of micro-bubble formation and other random surface effects that degrade liquid handling precision and accuracy, even with new disposable tips.

IonField Systems' TipCharger improves the reproducibility of process results, shortens automation cycle times, reduces the number of lost runs, and eliminates environmental waste and liquid handling disposables. The overall result is increased confidence in results and a more effective and productive laboratory operation.

About IonField Systems™

IonField Systems is an advanced technology company focused on the development of state-of-the-art pipette tip cleaning for life science laboratory research applications. IonField Systems provides on-site technical support services to assist laboratories in rapidly integrating the system into day-to-day operations.