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Research Article

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Evaluation of Antibacterial Efficacy of Publically Available Hand Sanitizers for the General Public in Islamabad

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Abstract

Hand washing is recognized as a key element to prevent the spread of infectious diseases. Hand sanitation is the act of cleansing hands with sanitizers to ensure proper hand hygiene. Keeping in view, we conducted the study to evaluate the antimicrobial efficacy of sanitizers available for the public at twenty different locations in Islamabad. These locations included places like (ATMs, restaurants, labs, etc.). These 20 samples were applied on the hands of 40 different people with two different approaches and got disappointing results both ways. The Petri dishes showed almost the same amount of bacterial activity before sanitizing and after sanitizing. The research limelight the authenticity of the publically available hand sanitizers; whether they are efficient or not to the extent that they could be relied upon so effortlessly, whether or not the people governing the public places are being responsible for the check and balance of these sanitizers, all of these ingenious ways given to the public to keep their health and hygienic safety intact in these tough times of the pandemic just so that the social fear of going out can be adjusted to the new normal which is the excessive care of health and hygiene to lower the chances of the spread of Covid. The research study also removes the redundancies that cloud the judgments of a common man on the "do's and do not" on how to make the interactions and work in public places safe along with a concluded discussion on the results that were taken from the experimentations.

Keywords: *Sanitizer; COVID-19; Hygiene; Islamabad*

Introduction

Hand hygiene gains its utmost importance as it can be easily contaminated by direct contact with airborne microorganisms present in the atmosphere or the ones that may come in contact with our skin through coughing and sneezing. Particularly in situations when the global pandemic is at its full glory, it is essential to interrupt the chain of transmission of the virus through the practice of proper hand sanitation as hygiene can be the best barrier between the transmissions of all the viral and bacterial contaminants out in the environment that not only can be hazardous but also fatal if not taken care off. It can be achieved with contact isolation and a rigorous infection control tool such as maintaining good hand hygiene specifically in public. In these tough times, people tend to keep small pocket-sized hand sanitizers and wet wipes just so that they can be in the maximum assurance that they can be safe from the coronavirus as they believe to gain a barrier from it in the form of the sanitizers. Because of this kind of hygienic attitude amongst the people, the public places started to provide hand sanitizers in

their nameless containers which were then offered to the people who were leaving or entering the premises of that particular public place. This notion at the public places gave people ease of mind that even if they do not come out with their hand sanitizers, they'll still have the chance to sanitize their hands at whichever public place they turn out to be gaming up. One of the major reasons to conduct a study on hand sanitizers was due to the mere fact that despite its popularity, there is not much literature regarding their efficiency. To keep it basic, the main target of our research was to evaluate the efficiency of hand sanitizers against bacteria solely. So, other microorganisms were out of the equation to avoid complications. We did not have any objections to testing it against viruses or other microorganisms. At first, we thought that comparing different brands available in the market was a good idea but the enormous number of companies that recently started production would've made it impossible to buy all of them and compare which is best. So, to randomize it, we collected samples from public places. The samples of hand sanitizers were collected from (ATMs, Restaurants, Grocery stores, cafes, educational institutes, and labs, etc.). With the help of these samples, we could determine the efficiency of the publically available hand sanitizers which would then help us rely on the public places for our hygiene if the results of the experimentation prove to be satisfactory and reassuring.

COVID-19 and Hand Sanitizers

COVID-19 was officially recognized as a World pandemic disease by World Health Organization (WHO), March 11, 2020 (Saqlain et al 2020). During this pandemic situation, hand sanitizers became popular, and maintaining hand hygiene was on the top of the list of guidelines which were provided by WHO. To understand the correlation between Coronavirus outbreak and hand sanitizers. We need to analyze the mechanism of its spread. It was identified in Wuhan, China, in December 2019. COVID-19 is an infectious and extremely contagious disease that is caused by the SARS-COV-2 virus (Hu, et al 2021). COVID-19 is transmitted when people breathe air contaminated with droplets and small particles in the air. The risk of inhalation is greatest when people are nearby, but these small droplets can be inhaled over long distances, especially indoors. Hands touch too many surfaces and can quickly pick up viruses. Once infected, hands can transmit the virus to your face, from where the virus can enter your body. That is why hand hygiene is also very important to control the spread of COVID outbreaks. Washing hands with disinfectant detergents or hand washes is an effective measure in preventing infective disease transmission. In the context of coronavirus disease 2019 (COVID-19) prevention, the WHO and Centers for Disease Control and Prevention have recommended washing hands with soap and water after coughing and sneezing, visiting a public place, touching surfaces outside the home, and taking care of a sick person, as well as before and later eating.

Microbial Diversity on Human Skin

(Kong & Segre, 2012) reported that approximately 1 billion bacteria inhabit a typical cm2 of human skin, covering the surface and extending down into the appendages and glands. These microbes play their role in human health and diseases (Kong & Segre, 2012). Microorganisms in two forms on the skin are resident microbes and transient microbes. Resident flora normally resides under the superficial cells of the stratum corneum and can be found on the surface of the skin from birth to end of the life (Zubair et al., 2014). They are often considered to be commensal which means that the microbes are not harmful and may provide benefit to the host. Whereas the transient flora comes from the environment and does not remain on the surface permanently (Kong & Segre, 2012). These microbes have a very low growth rate on the skin and are responsible for the transmission of infections (Zubair et al., 2014).

An interesting feature of the microbiota inhabiting the dry sites of the skin, as captured by molecular analysis, is the plenty of Gram-negative organisms as contaminants from the gastrointestinal tract, which were formerly thought to colonize the skin only infrequently (Kong & Segre, 2012). Environmental factors, intrinsic factors such as age, genetic makeup, and immune response, and hygiene conditions influence the composition of skin microbial communities (Kong & Segre, 2012). Make-ups, soaps, hygienic goods, and conditioners are also contributing to the distinction of skin microbiota by changing the skin barrier. Generally transient and resident flora differs significantly among individuals, it is often comparatively constant for any given individual.

Hand Hygiene

Today, hygiene is connected with disease prevention and health promotion. The hygiene is globally documented and evidence-based. Many of the research studies have described an association between improvements in hand hygiene and decreases in rates of infectious diseases. The behavior, customs, and concerns of the people largely govern their hygienic conditions Ojima et al., (2002). Palm counts the most dynamic skin microbial habitats given the nearly constant and varied exposure to environmental surfaces A-Wahab et al., (2016). Transient pathogenic bacteria that are possibly present on the hand include Salmonella 8 typhi, Escherichia

coli, Pseudomonas aeruginosa, Proteus mirabilis, Klebsiella species, Enterobacter species, Streptococcus species, and Staphylococcus aureus Biswas et al., (2018). Several hygienic measures can be taken to prevent cross-contamination from one surface to another Al-Harbi et al., (2018). Hand washing, also known as hand hygiene, is the act of cleaning hands to remove soil, dirt, and microorganisms. It is estimated that simple yet effective hand washing could save one million lives annually and many public health campaigns worldwide have addressed "hand hygiene" with varying success. As part of a major global effort to improve hand hygiene in health care, the WHO in 2009 launched a global campaign named "SAVE LIVES: Clean Your Hands". A study in the north of England found that only 43% of mothers washed hands after changing a dirty nappy and studies have found low rates of handwashing in public washrooms (Judah et al., 2010). Previous studies of the prevalence of fecal and total coliforms found 20% of public surfaces were positive for coliform bacteria and 7% were positive for the presence of fecal coliforms (Mc Michael, 2016).

The influence of hand hygiene depends not only on the consistency and carefulness of the measures used but also on the type of handwashing agent carefully chosen (Bhatia & Dehankar, 2017). The efficacy of different handwashes in removing the bacterial flora from hands indicates the probability of bacterial transmission (Ataee et al., 2017; Fuls et al., 2008). Although specialists approve that washing hands with soap and water is efficacious at reducing the spread of pathogenic bacteria, there remains uncertainty on the benefit of antimicrobial hand washes over non-antimicrobial soap and water. The variations in the results 10 may be attributed to confounding factors, such as soap volume, wash time, type of antimicrobial product, and lack of uniformity among these factors in the published studies. Another study demonstrated that the soap volume and wash time can influence the number of resident bacteria persisting on the hands after multiple hand washes but not after a single hand washes. The efficacy of non-antimicrobial handwash and antimicrobial handwash is measured based on bacterial reduction alone and not the subsequent transfer of bacteria after the use Fuls et al., (2008).

Risk of Irritant Contact Dermatitis Due to Excessive Washing of Hands

Excessive washing of hands is also dangerous as it can damage the skin barrier. Washing your hands with soap and water helps wash away dirt and germs, but it also removes the beneficial oils your skin needs to stay healthy and the beneficial bacteria that protect against disease. Unfortunately, every product you practice to preserve your hands clean, and even the water itself, is stripping your hand of a much-needed barrier to keep them safe from damage. It has been found that frequent hand washing over a long period can cause long-term skin changes, leading to skin conditions such as chronic skin damage, irritant contact dermatitis, and eczema. Damaged skin can upsurge the risk of infection and transmission of infectious microbes. This is where the use of sanitizers has an advantage over soap and other water-based hand washing techniques. Alcohol-based sanitizers typically are less likely to cause ICD than handwashing with detergent-based or antimicrobial soaps. Antimicrobial ingredients in soaps such as chlorhexidine, chloroxylenol, and triclosan are frequent culprits. Detergents in soap cause more skin irritation and trans epidermal water loss than alcohol. In the context of this, this study focuses on the below objectives. To test the efficiency of the hand sanitizers that are available for the general public at the open public places (e.g., restaurants, ATM's or any other institution that may accommodate a large number of people at the same time). To check the hygienic factor of using the publically available hand sanitizers in the actual sanitization of the hands.

Methodology

Study Area

The research study includes the different areas of the capital of Pakistan, Islamabad. The nature of this study revolves around in-vitro type examination where the growth rate of bacteria and its behavior is observed in a controlled environment which is given inside Petri-dishes for each collected sample. Despite the popularity of sanitizers, there is not enough literature to prove the efficacy of the sanitizers that are used in public areas, against bacteria. We collected 20 samples of sanitizers in sterilized bottles from different locations in Islamabad to initiate our experimentations.

Sample No.	Name	Location	Latitude	Longitude
ATMs				
1	Muslim Commercial Bank	E11 Markaz	33.70362	72.97886
2	Silk Bank	F10 Markaz	33.69812	73.01135
3	Habib Bank Limited	F11 markaz	33.68486	72.98612
4	Alfalah Bank	E11 markaz	33.70395	72.97864
5	United Bank Limited	E11 Markaz	33.70349	72.97889

Cafes				
6	Mocca Café	F-6 Markaz	33.72065	73.07387
7	Burning Brownie	F-6 Markaz	33.72071	73.07395
8	Chikachino	Bahria Enclave	33.68625	73.21063
9	Juice Time	F11 Markaz	33.6848	72.98807
10	Gloria Jeans	Bahria Enclave	33.68629	73.21069
11	Tayto	I-8	33.66841	73.07527
12	What A Paratha	F-7 Markaz	33.72136	73.05914
13	Downtown	F-7 Markaz	33.71977	73.05302
14	Monalo Gelato	Centauras mall	33.70792	73.04984
Other Public Places				
15	Najeeb Pharmacy	E11/3	33.70021	72.97292
16	Shaheen Chemist	G-11 Markaz	33.66954	73.00022
17	D.Watson, G-11	G-11 Markaz	33.6701	72.9994
18	Chugtai Lab	Blue Area	33.70946	73.05723
19	Bahria University Quaid-e-Azam Block	E-8, Shangrilla road	33.71818	73.03436
20	Bahria University Iqbal Block	E-8, Shangrilla road	33.71818	73.03436

Table 1: *Illustrate sampling locations in Islamabad.*

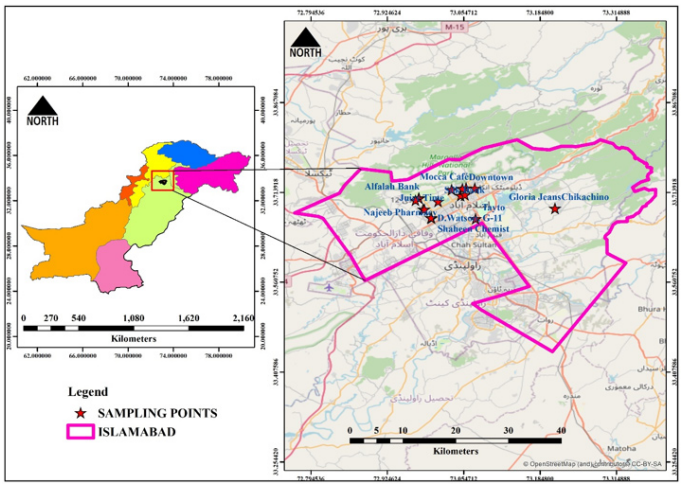

Figure 1: *Represents Sampling location in Islamabad, Pakistan.*

Procedure

To appraise the efficiency of sanitizers against bacteria, we prepared two nutrient agar plates for each sample. We chose nutrient agar as our culture media because it supports the growth of almost all types of bacteria. After sterilizing the culture media in an autoclave for 15 minutes at 121°C, it was poured into Petri dishes and left for some time so it can solidify. After solidification, one plate was inoculated before sanitizing as we needed to compare how many bacteria were present on the hands of the test subject and the other was inoculated after sanitizing to see how many bacteria were successfully killed by the sanitizer. The process of inoculation was done directly by gently touching the agar plate inside a vertical laminar flow cabinet. It was necessary to operate all the inoculation procedures inside the vertical laminar flow cabinet to avoid foreign contamination as a vertical laminar flow cabinet consists of a fan that is positioned on the cabinet ceiling and the contaminated air is sucked through this fan and conveyed from the top of the counter downwards in a vertical direction with a positive pressure while because of the HEPA filter placed under this fan, no contaminant can enter the cabinet from the top either. When both of the Petri dishes were done with inoculation, they were sealed and left inside an incubator at 37°C for 24-48 hours.

Culture Media Preparation

The culture media used for micro-organisms and pathogens Nutrient agar (a general-purpose medium supporting growth of a wide range of non-fastidious organisms generally including a variety of bacteria and fungi). Nutrient agar was made by suspending 23 g of nutrient agar powder in 1 liter of distilled water and mixing both these ingredients until the suspension became uniform. For sterilization, both these media were autoclaved at 121 degrees Celsius for about 20 minutes. Then this sterilized culture media was poured into Petri dishes inside a vertical laminar flow cabinet and it was left for 5-10 minutes to solidify and form a jelly-like structure which would be then ready for introducing micro-organisms.

First Approach

It was decided that the research will dedicate two Petri dishes for each sample; one for unsensitized hands and the other was for after sanitizing. Twenty random students from our university were asked to participate so that we can allocate one sample to each student. These students were briefed about the study design beforehand to avoid complications. Each test subject was supposed to fill the first agar plate with his/her dirty fingerprints to inoculate microbes and the second plate or petri dish was touched after the test subject had sanitized his/her hands with the allocated sample of sanitizer.

Incubation

The inoculated plates were then incubated in a bacterial incubator at 37°C for 24-48 hours. The inoculated Petri dishes must be incubated upside down to reduce the risk of contamination from airborne particles that settle on them and to prevent the accumulation of water condensation that could disturb or compromise a culture. A researcher would incubate a particular strain of bacteria at its optimum temperature so that they can study it when the growth rate is healthy. These bacterial organisms grow best at the temperature of the human body, which is around 37 degrees Celsius (98.6 degrees Fahrenheit). Therefore, the Petri dishes were incubated upside down to prevent the condensation formed on the lid from dripping thus drowning the colonies.

Second Approach

Culture media was arranged in the same way, by suspending 23g of Nutrient Agar in 1 liter of distilled water. Then, the solution was sterilized by placing it in an autoclave machine at 121°C for 15 minutes. Then it was poured into Petri dishes and left to solidify for 10 minutes. Now the transition from the first method was that after solidification, the agar plate was divided into four equal parts with the help of a red-hot inoculation loop which was heated with the help of a spirit lamp. The first portion was left untouched/controlled so that there's a section in the petri dish that would be kept in controlled conditions. When it comes to the second section, the test subjects disinfected their hands with methylated spirit as it was the only proven bacteria killer we had. The bottom left section was inoculated before sanitizing and the bottom right was dedicated to each sample of sanitizer.

Results

This study was conducted with two methods so that the authenticity and the legitimacy of the experiments could be tested and retained. The results of most of the samples are nonetheless the same, the following is the generalized descriptive representation of the Table 2.

Outcomes from the first approach

As discussed earlier, we used two Petri dishes to prepare nutrient agar media for each sample. One was inoculated before sanitizing and the other was done after sanitizing. Then they were kept in an incubator upside down for 24-48 hours. These Petri dishes were taken out to observe a reduction in visible microbial activity that was skin transient and can thrive on nutrient agar. Unfortunately, the majority of the samples failed to show competency and there was not a significant reduction in the bacterial activity of both plates of each sample as the results from this method showed that all these samples had poor anti-bacterial efficacy as the Petri dishes showed almost the same amount of visible microbial growth before and after the use of sanitizer. Only a few showed a little bit of reduction in the bacterial activity but still, these were also not satisfactory which motivated the idea to try a different approach for affirmation and to evaluate if there was an external factor influencing the results of the experiments to be so dissatisfactory.

Outcomes from the second approach

After getting disappointing results, we decided to test all these 20 samples with a different approach; which was to divide the agar plate with an inoculation loop into four equal sections and to compare each sample's efficiency to something that was proven to be very effective in the elimination of bacteria on human skin. For comparison, we chose methylated spirit as it is an effective disinfectant because it contains 80% alcohol, and solutions that have alcohol % more than 70% are said to be very effective against bacteria and viruses. One section of the petri dish was left untouched/controlled to observe if there is any foreign contamination second section was inoculated after using methylated spirit on one hand the third section of the agar plate was inoculated before disinfecting. And the fourth section of each agar plate was dedicated to one sample inoculation was done by gently touching the agar plate by thumbs inside the vertical laminar flow cabinet. After inoculation, the Petri dishes were covered with their lids and sealed with scotch tape and their plates were kept inside an incubator at 37°C for 24 to 48 hours. The second procedure elaborated a single petri dish for each sample; thus, giving the same environment for each section that the media was divided into by using an inoculating loop which was heated red hot on a spirit lamp and left upside-down in an incubator at optimal temperature for 24-48 hours. These sections included

- Controlled/untouched
- Inoculated by thumbs directly before disinfecting with the sample of the publically available hand sanitizer.
- Inoculated after disinfecting with methylated spirit
- Inoculated after sanitizing with one sample from the samples collected from various public places of Islamabad.

Methylated spirit was chosen as a comparison because it has an alcohol content of 80% and solutions that have concentrations above 70% are proven to be very effective in killing bacteria. These plates were taken out after the required time to observe the efficiency of

each sanitizer sample as compared to another wide-range disinfectant, which in this case, tends to be the Methylated Spirit. We took out these plates to observe the anti-bacterial efficacy but we got disappointing results again. Here is the scenario we had observed in the majority of the Petri dishes.

- The controlled section of each plate was clean from any kind of bacterial influence which proved that there was no involvement of any external contamination that could have sabotaged the entire experiment. Hence the controlled environment showed no growth of bacteria.
- the Second section showed visible microbial activity as it was supposed to because of all the bacteria that is present on dirty human hands.
- Methylated section showed an excellent reduction in microbial activity as compared to the second section.
- Whereas sanitizer sample dedicated sections showed no significant efficiency in killing bacteria or even mitigating or resisting against the rapid growth rate of Bacteria. Some samples showed some level of resistance against the rapidly growing bacteria in the cultured media but it was minimal when compared with the strength and effectivity of methylated spirit.

S. No	Sample location	Approach 1	Approach 2
1	Shaheen Chemist G-11	Low	Low
2	Najeeb Pharmacy	Low	Low
3	Juice time F-11	Low	Low
4	Silk bank	Medium	Medium
6	Bahria University HQ	Low	Low
7	D. Watson	Low	Low
8	Gloria Jeans, Bahria Enclave	Medium	Medium
9	Chughtai Lab	Low	Low
10	Chikachino, Bahria Enclave	Low	Low
11	Burning Brownie, F-6	Low	Low
12	Alfalah bank, ATM	Low	Low
13	Silk bank, ATM	Low	Low
14	MCB, ATM	Low	Low
15	Tayto, F-6	Low	Low
16	United Bank Limited, ATM	Low	Low
17	Manolo Gelato	Low	Low
18	What a Paratha, F-11	Low	Low
19	Bahria University, OC	Low	Low
20	Bahria University, Chemistry lab	Excellent	Excellent

Table 2: *Illustrate Efficiency of collected sanitizers samples.*

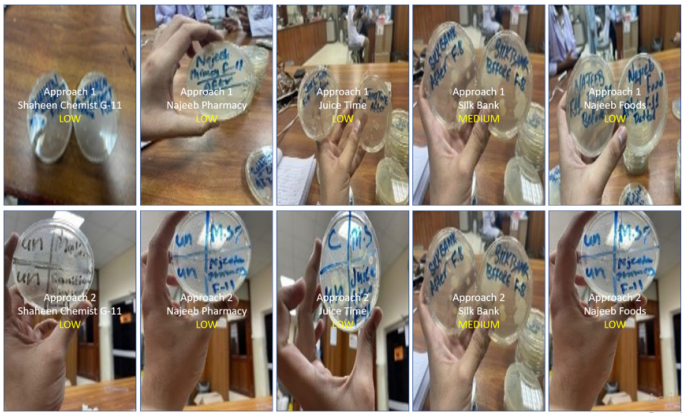

Figure 2: *Represents lab results sampling Sanitizers.*

Figure 3: *Represents lab results sampling Sanitizers.*

Figure 4: *Represents lab results sampling Sanitizers.*

Figure 5: *Represents lab results sampling Sanitizers.*

Discussion

While the dominant objective of this study was to understand the efficacy of the hand sanitizers present for the general public in the public areas, it is quite unfortunate to see that the results were not very comforting and satisfying. The publically available sanitizers failed to be proved efficient against the bacterial influence as all the results of the samples collected from different places brought forth their inefficiency when those were gone through repetitive series of experimentation done altogether. This objectifies the whole notion of satisfaction given to the general public that aggravated the use of hand sanitizers in the first place; causing a hoax in the understanding of the people as the use of hand sanitizers specifically meant a shield of protection from the bacterial influence to some extent which not only satisfies the user into thinking that they are somewhat protected against the germs but also makes them more vulnerable to the influence of the microorganisms making the public prone to get more diseases. Therefore, to expect any kind of protection and safety specifically in the tough times of Covid-19, we need to hop back to the conventional ways of cleansing that were normally practiced pre-pandemic, which included the use of hand wash soaps and then rinsing off the hands with a steady flow of water to thoroughly clean hands to remove the majority of contaminants. Not only is this well endorsed, but is also seen as a basic conventional method that has been practiced globally as a standard handwashing technique.

People also need to work upon the fact that the maintenance of hygiene in daily routine is not to be taken care of periodically just for the sake of a global pandemic, but should be regularly pursued as a daily routine so that the harmful influence of the external hazardous factors is minimized. This will not only ensure protection and safety from microbial contaminants but will also ensure a healthy lifestyle with a healthier immune system. Because, even though people try to regulate good hygienic practices in their daily lifestyle, they are most certainly only relying on hand sanitizers as it deems to be a satisfying shortcut towards good hygienic practice, but as the results have shown their way against the credibility of these hand sanitizers available at the public places, it is quite obvious that people would need to work upon the other attributes of good hygiene (e.g. bathing, showers, and hand washing Etc.) so that they could tackle any harmful external factor that could result in any infiltration of microbial contaminants inside the body.

Last but not the least, the center of discussion remains the fact that most of the sanitizers were not capable enough to mitigate the growth rate of the bacteria which kept on growing in the media that was present in the Petri dishes. Hence even though, we had these samples tested in a series of experimentation, the results moreover showed a similar and disappointing outcome. It was pretty shocking to see that the majority of the 20 samples proved to be an utter failure and were inefficient in showing any acknowledgeable antibacterial properties as both of the plates for each sample showed almost the same amount of bacterial activity which was visible with the naked eye.

Conclusion

Since the hand sanitizers were not up to par with the hygienic standards, as the sanitizers were not capable enough to mitigate or minimize the bacterial growth; it is completely appropriate to issue a proper check and balance on the quality of these publically given hand sanitizers.

Since one cannot rely on or trust the public places to have a good quality hand sanitizer in their publically available hand sanitizing bottles, therefore it is important to take the conventional hand cleansing techniques seriously and properly clean the hands under a steady flow of water with a good hand anti-bacterial hand wash to properly take care of hygiene.

Recommendations

- Those responsible for governing any public place should properly regulate QA/QC standards for the hand sanitizers that are put out by them for the public.
- Encouragement for transparency of the sources of hand sanitizers to the general public for proving the authenticity and legitimacy of the product.

- Strictly follow the given SOP's as per the government implementations (e.g., Avoidance of physical contact).
- Use anti-bacterial soaps to wash hands.
- Properly wash hands when returning home from any public place.
- Since one cannot suffice to achieve hundred percent results from any research study, therefore need for further experimentations to thoroughly observe the efficacy of publically available hand sanitizers remains necessary for more detailed results.

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