COMPARISON OF CYTOLOGICAL CHANGES ON PALMAR SURFACE PRIOR AND POST USAGE OF ALCOHOL BASED HAND SANITIZER IN HEALTH CARE PROFESSIONALS DURING COVID 19 PANDEMIC. A PILOT STUDY

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Abstract:

Background: After the outbreak of global pandemic, the novel coronavirus (COVID-19) increased the burden on health care professionals to take action to restrain the virus. WHO recommended hand sanitizers to prevent the spread of the disease. This Pilot study was carried out among 10 volunteered health care professionals to evaluate the effects of frequent use of alcohol based hand sanitizers on palmar surface through PAP test.

Aim: Comparison of cytological changes on palmar surface prior and post usage of alcohol based hand sanitizer in health care professionals during covid 19 pandemic.

Materials and Methods: PAP test was conducted on the smears taken from palmar surface of both the hands of 10 volunteered health care professionals and evaluated for clinical and cytological parameters.

Results and Conclusion: Health care professionals increased their use of hand sanitizers to prevent further spread of novel coronavirus (COVID-19) however use of hand sanitizers have adverse effects on the surface of the skin. The current trend in emerging pandemic like COVID 19, both the general public and among health care professionals, frequent washing with detergents, antimicrobial soaps and hand sanitizers needs careful reassessment in light of the damage done to skin. However further studies have be carried out for adequate protection and minimizing the risk of damage on the skin.

Keywords: Covid 19, Hand Sanitizers, Health Care Professionals

Introduction:

On 11th of March 2020, The World Health Organization (WHO) declared the novel coronavirus (COVID-19) outbreak a global pandemic. WHO Director General, Dr. Tedros Adhanom Ghebreyesus, said that the WHO is “deeply concerned both by the alarming levels of spread and severity and by the alarming levels of inaction," and he called on countries to take action to contain the virus.¹

The disease burden is expected to rise with ongoing community transmission through either direct contact with infected people or indirect contact with surfaces in the immediate environment. This requires frequent disinfection of the hands. Health Care Professionals (HCP) are at high risk of infection, contributing to further spread as they are in patient facing roles exposing to SARS-CoV-2.²

Frequent hand washing is essential to prevent the spread.³ however the requirement of frequent hand washing through soap and use of hand sanitizers may have adverse effects on the surface of the skin. The palmar surface being the most exposed to the disinfectant, this Pilot study was carried out among 10 volunteered health care professionals to evaluate the effects of frequent use of alcohol based hand sanitizers on palmar surface through PAP test.

Materials and Methods:

Informed Consent was taken from 10 volunteered HCP who were apparently healthy at the time of smear collection including no significant systemic conditions or any mucocutaneous lesions. 2 smears were taken from the palmar surface of both the hands. First smear was taken in the morning with no application of hand sanitizer for past 8 hours. Second smear was taken 20min post application of alcohol based sanitizer and both the smears were stained with PAP. Smears were evaluated for the following clinical parameters and cyto-logical parameters before and after the use of alcohol based hand sanitizer.

Clinical Parameters:

1. Age
2. Gender
3. Duration of Use Of Hand Sanitizer In Months
4. Frequency of Use Of Hand Sanitizer
5. Clinical Symptoms(Redness, Itching, Dryness, Burning)

Cytological Parameter:

1. Number of Cells Per High Power Field
2. Type of Strata Of The Epidermis
3. Cytological Alterations

Results:

Table 1 Clinical Parameter shows 80% female subjects and 20% male subjects with an average age of 40yrs. Average duration of hand sanitizer used was 7 months with frequency of 8times/ 6hrs. 40% of subjects showed clinical symptoms of Redness, Itching, Dryness or Burning sensation of hands. Table 2 and 3 showed cytological alterations including Pyknosis, increased micronuclei, karyorrhexis, Binucleation and nuclear vacuolation. PAP stained smears showed superficial and parabasal cells with more epithelial squames in post sanitized hands compared with before sanitization of hands. An average of 5-6squames were noted per high power field before the use of alcohol based hand sanitizer and 10-12 squames per high power field post alcohol based sanitization of hands.

Photographs:

Figure 1: Red cells: superficial cells, Green: Parabasal cells

Figure 2: Average of 5-6squames were noted per high power field before the use of hand sanitizer

Figure 3: Average of 10-12 squames were noted per high power field post alcohol based sanitization of hands

Figure 4: Nuclear atypia noted including pyknosis and increased micronuclei.

Figure 5: Nuclear vacoulation noted

Figure 6a and 6b: karyorrhexis seen

Figure 7: Binucleation noted
Table 1: Clinical Parameter:

| Case Number | Age  | Gender | Duration Of Use Of Hand Sanitizer In Months | Frequency Of Use Of Hand Sanitizer | Clinical Symptoms(Redness, Itching, Dryness, Burning) Seen In Percentage |
|-------------|------|--------|--------------------------------------------|-----------------------------------|------------------------------------------------------------------------|
| 1           | 32   | Female | 8                                          | 10                                | Present                                                                |
| 2           | 38   | Female | 6                                          | 6                                 | Absent                                                                 |
| 3           | 39   | Female | 8                                          | 10                                | Absent                                                                 |
| 4           | 45   | Female | 8                                          | 8                                 | Absent                                                                 |
| 5           | 50   | Male   | 8                                          | 8                                 | Absent                                                                 |
| 6           | 38   | Female | 7                                          | 6                                 | Absent                                                                 |
| 7           | 35   | Female | 8                                          | 8                                 | Present                                                                |
| 8           | 35   | Female | 8                                          | 7                                 | Present                                                                |
| 9           | 45   | Female | 8                                          | 8                                 | Absent                                                                 |
| 10          | 42   | Male   | 7                                          | 8                                 | Present                                                                |

Table 2: Cytological Parameter:

| Case Number | Number Of Cells Per High Power Field | Type Of Strata Of The Epidermis | Cytological Alterations |
|-------------|--------------------------------------|---------------------------------|-------------------------|
|             | Before Use Of Hand Sanitizer | After The Use Of Alcohol Based Hand Sanitizer | Before Use Of Hand Sanitizer | After The Use Of Alcohol Based Hand Sanitizer |
| 1           | 6-7 | 10-12 | Superficial Cells | Superficial Cells And Parabasal Cells | Pyknosis | Pyknosis, Increased Micronuclei, Karyorrhexis |
| 2           | 3-4 | 7-8   | Superficial Cells | Superficial Cells | Pyknosis | Pyknosis, Increased Micronuclei |
| 3           | 5-6 | 10-12 | Superficial Cells | Superficial Cells | Pyknosis | Karyorrhexisbinucleation |
| 4           | 2-3 | 6-7   | Superficial Cells | Superficial Cells | Absent  | Pyknosis |
| 5           | 5-6 | 10-12 | Superficial Cells | Superficial Cells | Pyknosis | Nuclear Vacuolation |
| 6           | 5-6 | 10-12 | Superficial Cells | Superficial Cells | Absent  | Karyorrhexis |
| 7           | 6-7 | 10-12 | Superficial Cells | Superficial Cells And Parabasal Cells | Pyknosis | Pyknosis, Increased Micronuclei, Karyorrhexis |
| 8           | 6-7 | 10-12 | Superficial Cells | Superficial Cells And Parabasal Cells | Pyknosis | Pyknosis, Increased Micronuclei, Karyorrhexis |
| 9           | 5-6 | 7-8   | Superficial Cells | Superficial Cells | Absent  | Increased Micronuclei |
| 10          | 5-6 | 7-8   | Superficial Cells | Superficial Cells And Parabasal Cells | Pyknosis | Pyknosis, Increased Micronuclei, Nuclear Vacuolation |

Table 3: Differences in the Cytology of Before and After the Use of Alcohol Based Hand Sanitized Smears:

|                  | Before Hand Sanitization | After Hand Sanitization |
|------------------|--------------------------|-------------------------|
| With Clinical Symptoms | 5-6 squames were noted per high power field with no nuclear atypia | 10-12 squames per high power field with moderate nuclear atypia |
| Without Clinical Symptoms | 5-6 squames were noted per high power field with no nuclear atypia | 10-12 squames per high power field with mild nuclear atypia |

Discussion:

The human skin constitutes the first line of defence against microorganisms and protecting against mechanical impacts and preventing excessive water loss from the body. It is composed of 3 main layers comprising of superficial epidermis (50–100 μm), a middle dermis (=2 mm), and an innermost hypodermis (1–2 mm). The uppermost epidermal layer, the stratum corneum (SC) functions as vital barrier, contains layers of corneocytes that are terminally differentiated from keratinocytes. The lipids that are derived from the process of exocytosis of lamellar bodies during terminal differentiation of keratinocytes will fill up the intercellular spaces between the corneocytes maintaining the cutaneous barrier function. Skin hosts a wide array of beneficial bacteria including *Staphylococcus epidermis*, *Staphylococcus aureus*, *Micrococcus* spp., *Propionibacterium* spp. and *Corynebacterium* spp. The skin flora is disrupted, by long term usage of frequent hand washing and microflora may become virulent. Constant skin regeneration is carried out by maintaining the balance in the microbiota. The above process takes about 28 days, from the time of mitotic division of basal epithelium to desquamation. Colonized microbes are removed by sloughing off dead keratinocytes in the SC. HCP are at greater risk in transmission of SARS-CoV-2 virus as they are in patient facing roles. To prevent further transmission of the virus, WHO recommends alcohol based hand sanitizer, containing isopropanol, ethanol or n-propanol with proven advantages of their rapid action and also a broad spectrum microbicidal activity. Thus providing protection against bacteria and viruses. Sanitizer containing at least 60% alcohol is more effective than hand
washing with anti microbial soaps as they inactivate and destroy the microbes. Proper hand hygiene by washing hands or using alcohol based sanitizer has become the most critical measure to prevent direct or indirect transmission of the COVID-19 as it reduces viable SARS-CoV-2 virus numbers on the contaminated hands.  

Alcohol based sanitizer usage causes mild to severe skin reactions like irritant contact dermatitis and allergic contact dermatitis by damaging the skin through denaturation of the stratum corneum proteins, alteration of intercellular lipids, decrease in corneocyte cohesion and reduction of stratum corneum water-binding capacity and frequent colonization by bacteria.  

Cells Palmar surface of the hand has twice as many cell layers and the cells are >30 times thicker than on the rest of the skin palsms are quite permeable to water. Long-term changes in skin pH associated with handwashing may pose a concern since some of the antibacterial characteristics of skin are associated with its normally acidic pH, subsequent changes in resident flora such as propionibacter. Literature shows repeated use of two washing agents, all skin function tests including stratum corneum capacitative resistance, lipids, trans epidermal water loss, pH, laser Doppler flow, and skin reddening were noticeably changed after a single wash. Further damage was noted after 1 week.  

Another study on irritant skin reactions induced by 3 surfactants, damage lasted for several days and complete skin repair was not achieved for 17 days. Nurses who frequently washed their hands showed declining skin health and damaged hands were twice as likely to be colonized with S. hominis, S. aureus, gram-negative bacteria, enterococci, and Candida spp. Each wash of skin, undergoes profound changes. Most of them transient. However, health care professionals, who require frequent hand washing can have long term changes resulting in chronic damage, irritant contact dermatitis and eczema, and concomitant changes in flora.  

Exfoliative cytology is the microscopic examination of shed, desquamated cells from body surfaces or cells harvested by rubbing or brushing a lesional tissue surface. PAP/Papanicolaou stain is most commonly used for exfoliative cytology as Papanicolaou analyzes squamous epithelial cell morphology with improved cellular transparency and by producing wider ranges of polychromasia, optimizing cell type differentiation and diagnostic confidence. It stains highly keratinized cells yellow, superficial cells red and parabasal cells green (Fig 1).  

In our study, cytological examination showed increased number of exfoliated cells after using alcohol based hand sanitizer compared to before using hand sanitizer (Fig 2 and 3). Pyknosis (Fig 4), nuclear vacuolization (Fig 5) (degenerative changes), karyorrhexis (Fig 6a and 6b) and Binucleated cell (Fig 7) features suggestive of cell death. Cell Death is comprised of a continuum of effects, culminating in chromatin condensation nuclear (pyknosis), nuclear fragmentation (karyorrhexis), dissolution of a cell nucleus (karyolysis) Numerous toxicants have been shown to cause both apoptosis and necrosis, with necrosis associated with higher doses and more severe toxicity.  

**Conclusion:**  
The current trend in emerging pandemic like COVID 19, both the general public and among HCP, frequent washing with detergents, antimicrobial soaps and hand sanitizers needs careful reassessment in light of the damage done to skin. However further studies have be carried out for adequate protection and minimizing the risk of damage on the skin.

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