



# A Quick Laboratory Method for Assessment of Blood Penetration and Splash Resistance of PPE Fabrics During the COVID-19 Pandemic Situation

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## Abstract

In the current outbreak of COVID-19, healthcare facilities are hit by a shortage of supply of Personal Protective Equipments (PPE) owing to extensive local and global demands and restrictions on their import or export. To circumvent this, trials with several indigenous materials suitable to qualify for PPEs and sterilization techniques for their reuse are being carried out. Prior to their commercialisation, it is imperative to evaluate the resistance of the PPE fabrics against penetration of synthetic blood under applied pressure, 40–300 mmHg as per test standards. Generally, two types of tests are recommended, Penetration Test and Splash Resistance Test, the former being more stringent. While the final certification of PPEs is carried out by authorised agencies, a first impression quick estimate of the choice of fabric can be made using a simple laboratory set-up. This study describes setups developed in the laboratory to carry out these tests. Evaluation of the fabrics, post-gamma irradiation, was also carried out. Microscopic examinations were performed to investigate radiation-induced structural changes in fabrics showing degraded performance. This set-up is useful for selection of fabrics and to assess the feasibility of reuse of PPEs, which is the need of the hour in this pandemic situation.

**Keywords** PPE fabrics · COVID-19 · Laboratory set-up · Synthetic blood penetration · Splash resistance · Radiation sterilization

## Introduction

In the current scenario of COVID-19 pandemic, a large number of Health Care Professionals (HCPs) are being engaged for medical, clinical, and logistics managements. The chances of accidental exposures have increased, with possibility of contact with pathogenic organisms. Major routes of pathogen entries are inhalation and/or ingestion of

aerosols, open skin lesions, subcutaneous injections through needle, and scratches with contaminated objects (Coelho and GarcíaDíez 2015; Wurtz et al. 2016). Some crucial viral infections like recent outbreaks of Ebola virus, norovirus, and Middle East Respiratory Syndrome (MERS) corona virus have already been reported for their association with occupational infections (Kilmarx, et al. 2014; Saegeman, et al. 2015; Hsieh 2015).

To avoid/minimize the risk of accidental exposure to pathogenic organisms, it is mandatory that the HCPs wear suitable Personal Protective Equipment (PPE) at workplaces when dealing with infected patients. Current scenario demands quality controlled, rapid production of Personal Protective Equipments (PPEs) like face masks, aprons, gloves etc. PPEs act as a protective barrier between the patients and the HCPs, by avoiding penetration of body fluid (blood, saliva, plasma, serum, urine, spit, etc.) which may ooze or spurt-out from the patient's body at a pressure ranging from 60 to 300 mm Hg (Jones et al. 2020). The spurted body fluids may carry various kinds of contagious microorganisms like viruses, bacteria, fungal-spores, and

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other parasites. Among these, the viruses are critical and seek special attention as they are small in size (Nanometre-range) and become the limiting criteria for the selection of appropriate fabric for the PPEs (Nikiforuk et al. 2017).

Selection of suitable PPE material is a crucial step for PPE manufacturing industries. ISO 16603 and ASTM F1670 are the most followed standards to categorise these materials, as per their performance, into different classes, which can be used in various medical/clinical scenarios (ISO 16603 2004; ASTM F1670, F1670M-17a. 2017). There are various methods available for testing of the resistance of the fabrics used for PPE kit preparations. Two standard methods, (1) Synthetic Blood Penetration Resistance Test, and (2) Splash Resistance Test (of synthetic blood), are widely accepted/practiced in the domain of PPE fabric testing (Rengasamy et al. 2015; Shimasaki et al. 2017). Synthetic Blood Penetration Resistance Test (SBPRT) is a standard, most crucial, and widely used criteria for qualification of the fabric. This involves assessing the resistance of the fabric against an applied fluid (synthetic blood) in the pressure range of 60–160 mmHg for varying time durations (5–30 min or more) (JIS, T. 2007; JIS, T. "8060 2007). Detection of penetrated fluid, downstream of the test sample, acts as qualifying indicator of the test.

These tests demand high end/sophisticated, established infrastructure, involving high cost instruments/equipment. Besides, these facilities were non-functional or non-accessible during lockdown, when PPEs were high in demand. The proposed method is a simple laboratory set-up, which can be assembled with existing regular laboratory instruments/equipment in any tissue culturing biological laboratory. Most of the components/equipment used in this set-up such as syringes, manometer, and filtration assembly are commonly available in the market at low costs. In this pandemic crisis, small laboratory scale test set-ups can help to assess the choice of fabric having potential to qualify for PPE manufacturing and can be effectively used for performance evaluation of PPEs made from new materials, or those subjected to sterilization methods for their possible reuse.

Extensive use to PPE kits during pandemic demands high availability and local manufactures. Therefore, locally and globally, there have been an impetus for exploring feasibility of reuse of PPE kits after complete sterilization (World Health Organization 2020). Among all sterilization methods (dry heat, vaporized  $H_2O_2$ , plasma gas, etc.) radiation sterilization has its own distinct advantages as it can be carried out in a sealed package and without raising the temperature.  $^{60}Co$ -gamma-rays induced inactivation of SARS-CoV family viruses has been extensively reported. A dose of 30 kGy ( $^{60}Co$ -gamma-rays) is recommended for complete inactivation of RNA viruses (SARS-CoV, MERS-CoV), based on cell cultures or tissue-culture assays (Kumar et al. 2015).

This study includes, development of test set-up with existing laboratory equipment to evaluate performances of 11 PPE-fabrics against penetration of Synthetic Blood Equivalent (SB) at various pressures ranging from 40 to 300 mmHg applied fluid pressure, both before and after radiation sterilization (30 kGy, gamma radiation). Of the 11 fabrics, three were subjected to splash resistance test. As a mechanistic approach, radiation-induced structural changes in the fabrics were examined under fluorescence and bright field microscope.

## Materials and Methods

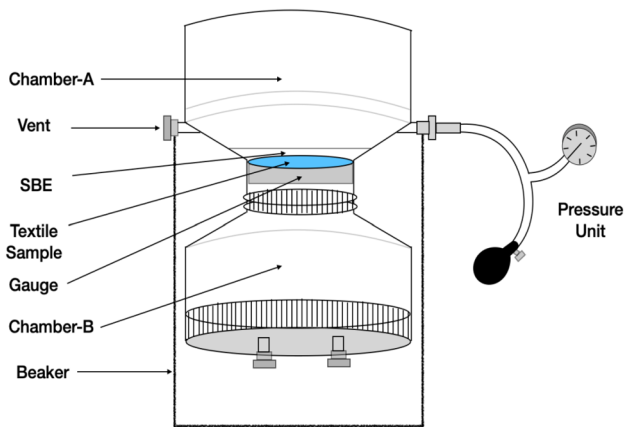
### Test Materials/Fabrics

Limited commercial information about composition of the material used in these fabrics was available to us; the maximum fraction of most fabrics is Polyethylene/Polystyrene/Polypropylene. All the fabrics were non-woven type with multiple internal layers and hot-spot press patterns. However, fractions and densities of Polyethylene/Polystyrene/Polypropylene were not same in all fabrics. Some samples of fabrics irradiated using gamma radiation to a dose of 30 kGy were also received to study the effect of gamma sterilization on the PPE material.

### Simulation of Test Set-Up for Synthetic Blood Penetration Resistance Test

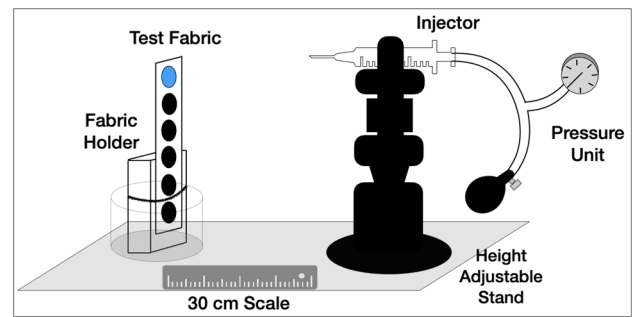
We have put together a working set-up with existing laboratory instruments while following the guidelines of ISO 16603 ASTM F1670 and JIS 8060 and 8122 with minor modifications without compromising on physical parameters of the test standards. The set-up was built with a conventional biological media filtration assembly, as shown in Fig. 1. It included two vented cylindrical chambers, which could be sealed airtight to sustain planned pressure levels. These two chambers were connected at the middle region of the assembly (which was designed to hold the filter) through a gauge. The circular piece of fabric, of diameter ~ 5 cm of the test material was placed over this filter holder. The upper surface of the fabric was layered with 20 ml of SB and this chamber was connected with a pressure unit of the sphygmomanometer to create and measure air pressure in the chamber. It was ensured that fluid column height of SB was very small (~ 1 cm) so that its gravitation pressure is negligible compared to the applied pressure.

For the production of PPEs, commercially available blood repellent fabrics are chosen, performances of these fabrics are being evaluated by employing a liquid, resembling physical characteristics of blood. Surface tension of blood and



**Fig. 1** Laboratory simulated set-up for Synthetic Blood Penetration Resistance Test on PPE fabric

other body fluid ranges from 42 to 60 mN/m (except saliva). To simulate similar wetting characteristics, synthetic blood was prepared with polysucrose and ionic salts of sodium and calcium disodium in water to obtain the surface tension of  $50 \pm 10$  mN/m (Portnoff et al. 2021). A colouring agent was added for better detection of SB penetrated through the fabrics. The whole set-up was kept in a 2.5 L beaker to support it and collect the dripping fluid, in the vertical set-up, as shown in Fig. 4. The pressure was exerted manually from chamber A with the help of a rubber bulb. Setup was first tested with non-porous nitrile glove material for its pressure holding ability and leakages, if any. It was confirmed that, pressure once exerted persists without any leakage up to tested pressure of 300 mmHg. Our set-up qualified for precise pressure creation and holding criteria without any leakage.



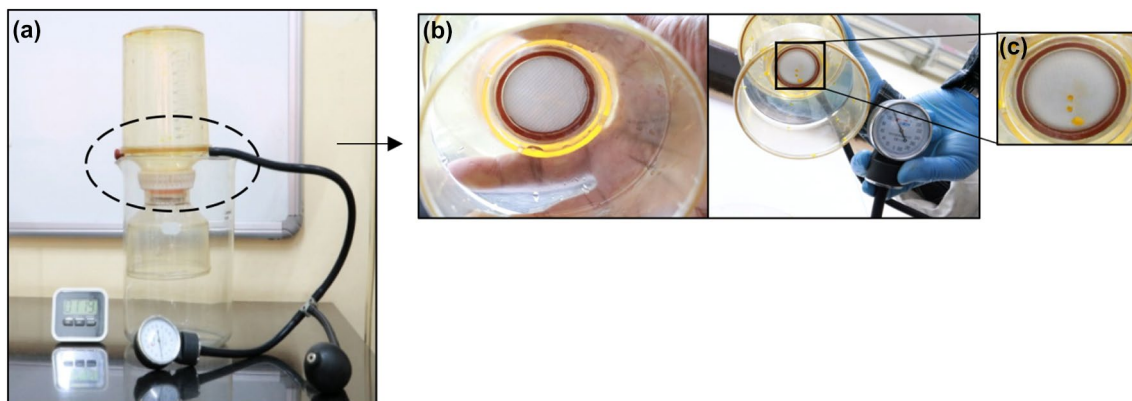
**Fig. 2** Laboratory simulated set-up for Splash Resistance Test on PPE fabric

### Simulation of Test Set-Up for Splash Resistance Test

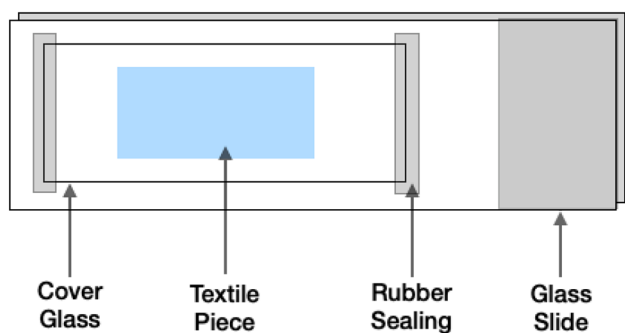
This test set-up comprised of a circular fabric holder and an injector placed at a distance of 30 cm as shown in Fig. 2. The fabric holder was mounted on a lead block for stability and placed in a beaker to collect the dripping liquid. The injector was placed at an appropriate height with the help of a height adjustable stand. In one run, 8 ml of SB fluid was injected on the fabric sample and the penetrated fluid was detected on the other side of the fabric.

### Detection of Synthetic Blood Penetrating Through the Fabric

For both the above-mentioned tests, the penetrated SB was detected at 3 levels; (1) visual detection, (2) detection by absorbent paper—after applying the planned fluid pressure, absorbent paper was swiped on to the downstream surface, SB spot created if any, was examined visually and (3) detection by magnifying lens—third level of detection was carried out with magnifying hand-lens (for both penetrated droplets and spots created on absorbent paper) (Fig. 3).



**Fig. 3** a Complete test set-up for SBPRT, (b) Lower side of the fabric mounted on a holder, and (c) Visually observable droplets that penetrated through the fabric



**Fig. 4** Preparation of slide for microscopic examination—PPE fabric sample sandwiched between glass slide and cover glass with the ends sealed with rubber cement

**Table 1** Time duration of the gradual pressure increase applied to the fabric samples and its JIS classification based on penetration resistance (Adapted from JIST 8122)

Time (min)	Pressure (kPa)	Visual check	JIS T 8122 classification
$0 \leq t \leq 5$	0	Leak	< Class 1
$5 < t \leq 10$	1.75	Leak	Class 1
$10 < t \leq 15$	3.5	Leak	Class 2
$15 < t \leq 20$	7	Leak	Class 3
$20 < t \leq 25$	14	Leak	Class 4
$25 < t \leq 30$	20	Leak	Class 5
$30 < t$	20	Non-leak	Class 6

### Radiation Sterilization of PPE Fabrics

For experimentation a piece of 6 × 6 inch. of fabric was cut from the PPEs and irradiated with the dose of 30 kGy at a dose rate of 6.2 kGy/h in the gamma chamber (GC-5000) at BARC.

### Microscopic Examinations of Radiation-Induced Structural Changes in Fabrics

Fabrics were cut into pieces of 2 cm × 1 cm dimensions (from the piece of 6 × 6 inch.), sandwiched between the glass slide and a cover glass and ends were sealed with rubber cement (Fig. 4) to prepare them for examination under a microscope. Total 6 samples were observed and analysed, 3 before and 3 after radiation sterilization (30 kGy). Structural changes/examination of voids, were carried out at 100× and 400× magnifications (Figs. 11 and 12).

## Results and Discussion

### Performance Evaluations by Synthetic Blood Penetration Resistance Test

The fabric samples A–K were subjected to SBPR test in a pressure range of 0–300 mmHg or up to the breaking/leaking pressure point. The fabrics were first exposed to the fluid for 5 min at atmospheric pressure. Pressure was applied in an increasing order in step of 20 mmHg for 5 min at each pressure point.

Japanese Industrial Standard (JIS) has classified protective clothings/fabrics in different classes based on performance of the fabrics against contact of the blood (synthetic blood) and other body fluids applied under pressure JIST 8122, has ranked the penetration resistance of the fabric material in 7 classes (from < class 1 to class 6), as described in Table 1 (Shimasaki et al. 2016). The results of the test with respect to the sustained pressure, the corresponding JIST 8122 classification and their utility classes are given in Table 2. The effect of radiation in degrading/deterioration of Fabrics A and B is also evident from this Table 2. For Fabric A, the pressure holding ability decreased from 80 to 60 mmHg and Fabric B lost its ability to hold 40 mmHg fluid pressure post-irradiation. The Fabrics C, I, J and K did not reach the breaking point even up to a pressure of 300 mm Hg, before and after radiation sterilization. It was observed that Fabrics D, E, F, G and H were able to sustain at atmospheric pressure but could not resist at 40 mmHg fluid pressure, started leaking heavily, both before and after radiation sterilization. For all fabrics, the test was repeated three times to confirm the results.

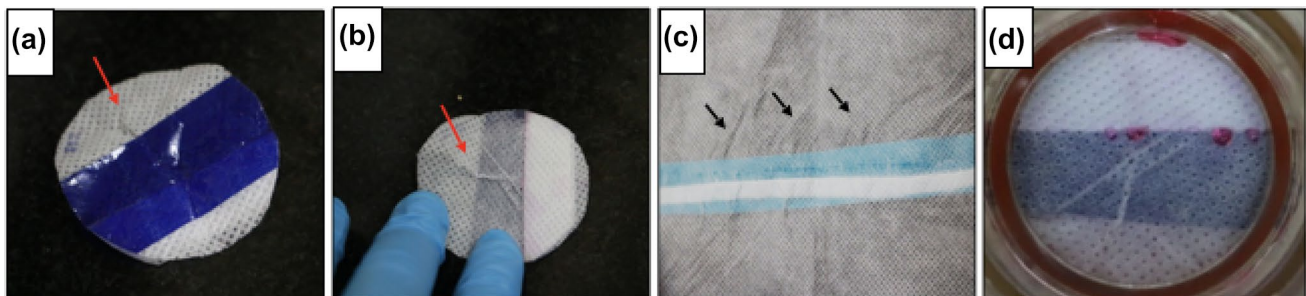
For Fabrics I and J, resistances were also tested at the fabric joints (sewed and taped). It was observed that, at improperly taped joints, some creases trapped inside the taping line acted as a passage for fluid to reach to the sewed line and penetrate through it (Fig. 5). However, in case of samples with properly taped joints, the highest pressure was sustained by the fabric. It is emphasized that quality of taping plays a crucial role irrespective of performance of the fabric as a whole.

### Method of Detection of Fluid Penetration Through the Fabric

Fluid penetration through the fabric was detected at three levels, first level of detection was visual through naked eye, following ISO 16603 protocols. To enhance sensitivity of the detection, swipe test was performed, wherein leaked surface was swiped with absorbent paper and spot created on it, if any, was observed to confirm leakage (adapted from Shimashaki et al. 2017) (Shimasaki et al. 2017). The test

**Table 2** Illustration of maximum tolerance pressure of Fabrics A–K before and after radiation sterilization and its JIST8122 classification and utility

Fabric sample	Tolerance pressure (mmHg)		JIST8122 classification	Performance of the material
	Before radiation sterilization	After radiation sterilization		
A	80	60	Class 3	Moderate
B	40	<40	Class 1	Low
C	>300	>300	Class 6	High
D	<40	<40	Class 1	Low
E	<40	<40	Class 1	Low
F	<40	<40	Class 1	Low
G	<40	<40	Class 1	Low
H	<40	<40	Class 1	Low
I	>300	>300	Class 6	High
I (joint)	<40 (improperly taped) >300 (properly taped)	<40 (improperly taped) >300 (properly taped)	Class 1	Low
J	>300	>300	Class 6	High
J (joint)	<40 (improperly taped) >300 (properly taped)	<40 (improperly taped) >300 (properly taped)	Class 1	Low
K	>300	>300	Class 6	High

**Fig. 5** Fabric sample showing creases at the taped joints: (a) Front view of the joint, (b) Back view of the joint, (c) Close view of the joint, (d) SB penetrated through the joint at 40 mmHg applied fluid pressure

was added assuming that micro amount of fluid penetrating through the fabric may not be visible by naked eye but can be detected by swipe test. The presence or absence of spots on absorbent paper was further confirmed by observation under a magnifying hand-lens. These levels of detection helped us to define cut off pressure holding point for each tested fabric.

Figure 6 shows visual observations of the samples showing the leakages through the fabric at varying pressure points, both before and after irradiation. On the other hand, Fig. 7, shows the three levels of detection adopted to confirm the leakage at the breaking point pressure.

### Fabric Evaluations by Splash Resistance Test

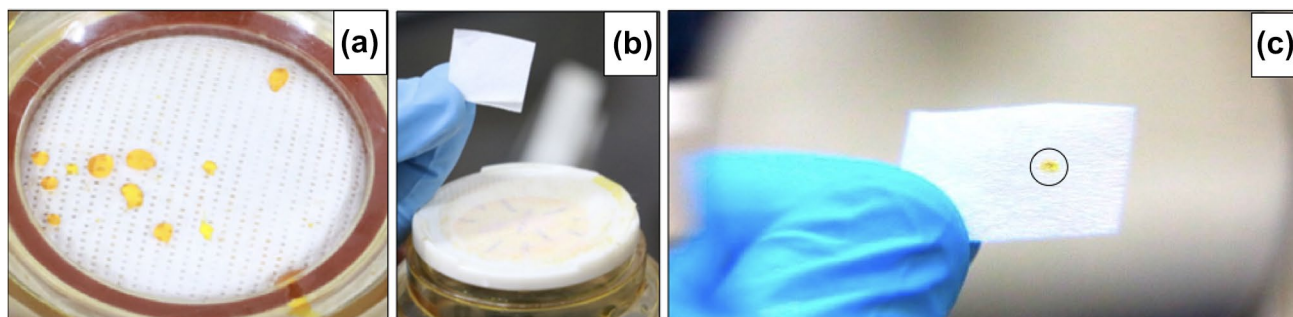
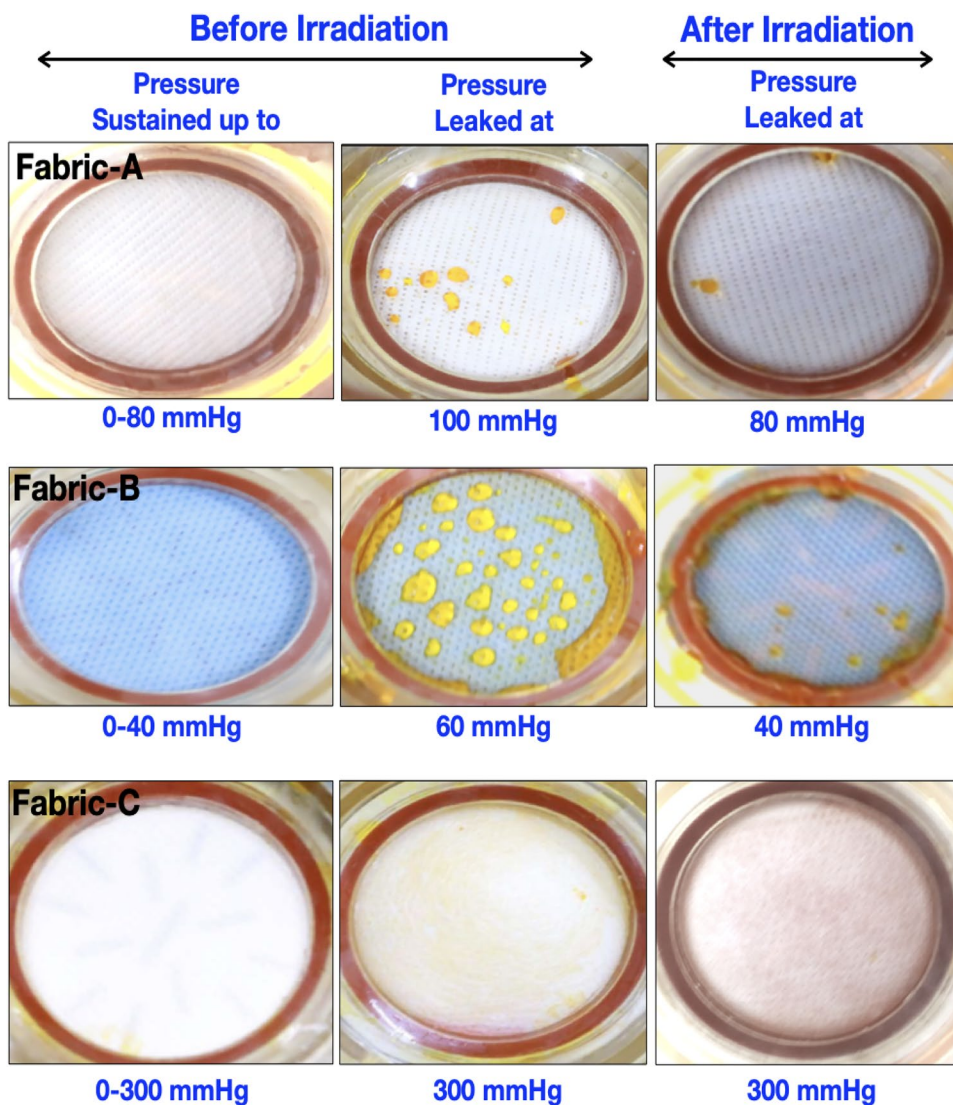
Performances of Fabrics A, B and C were further evaluated by Synthetic Blood Splash Resistance Test using the test set-up developed (Fig. 8). A ~2 cm diameter of fabric sample was placed in the fabric holder and 8 ml of SB was splashed

on it at a pressure range of 280–300 mmHg from a distance of 30 cm. This was repeated three times on each sample so that effectively 24 ml of SB was splashed on each tested fabric sample. As per ISO reference protocol, 2 ml of SB should be splashed on the test fabric, with 60–160 mmHg pressure ranges from a distance of 30 cm (ISO 16603 2004). No fluid penetration was detected for all the three fabrics (Fig. 9). Since the fabrics did not leak at the higher-pressure ranges and for large volumes over longer duration, there was no need to evaluate their performances at lower pressure ranges. All three levels of detections were followed as described earlier and are depicted in Fig. 10.

### Microscopic Examination of Fabrics

To observe structural changes induced in the fabrics by radiation sterilization, microscopic examinations were carried out. It was observed that post-irradiation, number and size of voids increased in the mesh region of the Fabrics A and

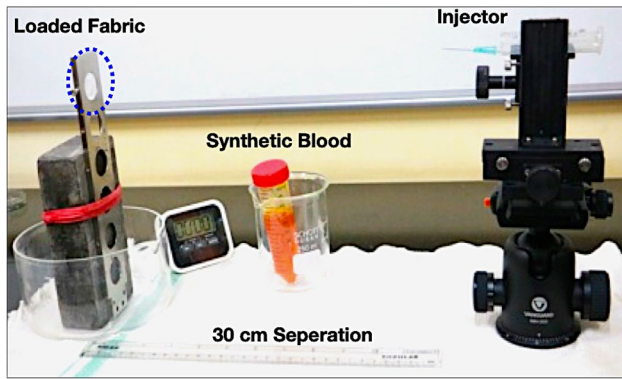
**Fig. 6** Visual examination of fluid penetration through test fabric at various pressure ranges. Left column shows no observed leakage, middle column shows the breaking pressure point at which fluid penetrated through the fabric and the last column shows breaking pressure points after radiation sterilization



**Fig. 7** Illustration of three levels of detection of fluid penetration through the fabric (a) naked eye, (b) swipe test with absorbent paper, (c) magnification with hand-lens

B, however, not much detectable changes were observed in pressed regions (Image of Fabric B is shown in Fig. 11, The image of Fabric A was also found to be similar and hence

not shown). This increased number and size of voids was responsible for decreased fluid pressure holding ability. No structural changes were observed (both in mesh and pressed

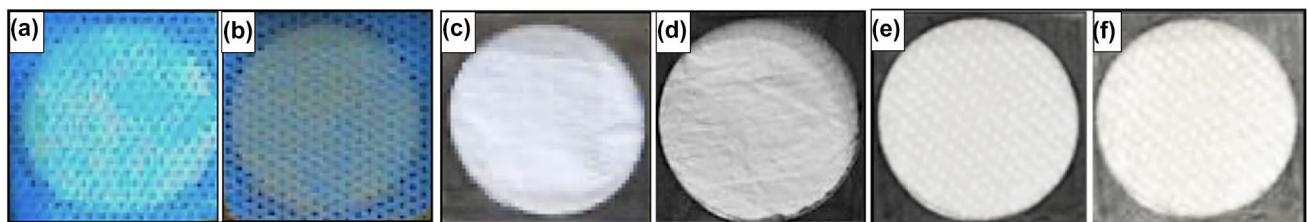


**Fig. 8** Test set-up for splash test showing the test fabric holder and the synthetic blood injector

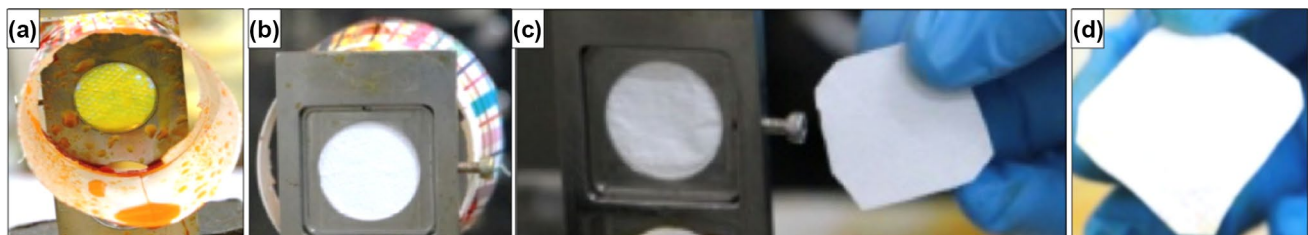
regions) in Fabric C, before and after radiation sterilization as illustrated in Fig. 12. Information gathered by microscopic examination of Fabrics A, B and C was in accordance with our SBPRT findings and it provided supplementary structural and mechanistic support to our observations.

### Conclusion

Simple test set-ups have been put together using existing laboratory instruments to evaluate the blood penetration resistance of various fabrics of PPEs. These can help in a quick screening and selection of fabric materials without compromising on parameters of the tests. These set-ups can be adopted in any research/medical facility for rapid

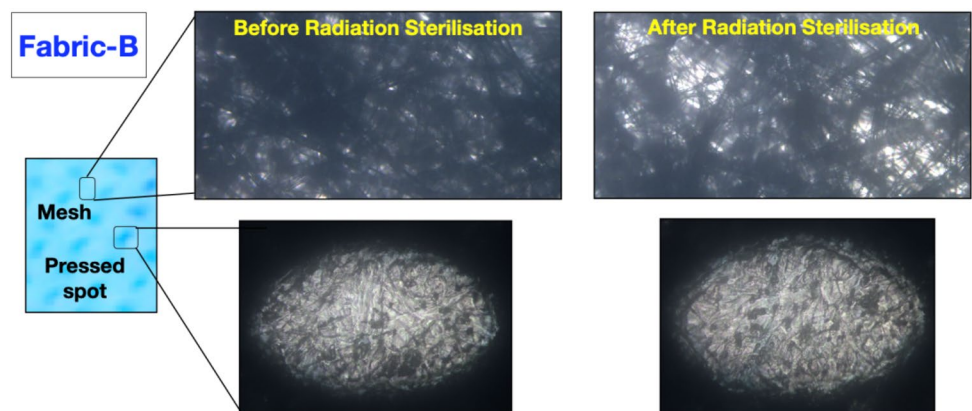


**Fig. 9** Back view of fabrics A, B and C before and after splash test. All fabrics resisted penetration of fluid splashed at pressures of 280–300 mmHg

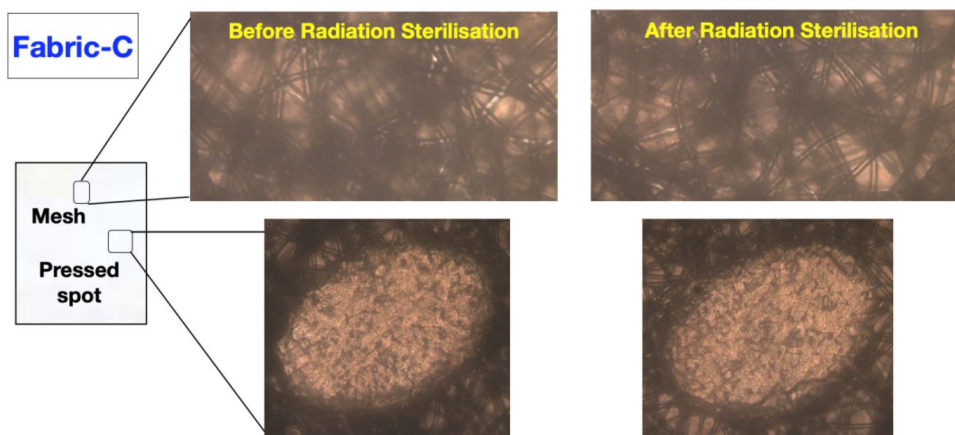


**Fig. 10** The detection levels for splash test: (a) Front view of fabric after splash, (b) Back view of fabric after splash, (c) Detection of leakage by swipe test with absorbent paper, (d) Magnified view of the absorbent paper after swipe test

**Fig. 11** Microscopic (bright field) examination of Fabric B, at 100× magnification revealed that radiation sterilization increases the number and size of voids in the mesh region of the fabric, however, no changes/deteriorations were observed in the pressed region, before and after radiation sterilization



**Fig. 12** Microscopic (bright field) examination of Fabric C at 100× magnification showing no structural changes in mesh and pressed regions, before and after radiation sterilization



performance evaluation of PPE-fabrics during any emergency situation like the current COVID-19 pandemic. Such a set-up will help local manufacturers to produce PPEs in adverse situations without compromising on the safety parameters.

Pressure holding ability of the fabric is an intrinsic property of the material that cannot be changed. However, when the PPEs are tailored, the quality of the sewing and taping at the joints is utmost important. As observed in this study, the pressure holding ability of the poor joints was considerably reduced, much below the holding ability of the same fabric. An appropriate tailoring and taping to achieve proper sealing of the joints can improve the performances of these PPEs.

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## Declarations

**Conflict of interest** Authors declare that, they do not have any conflict of interests.

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