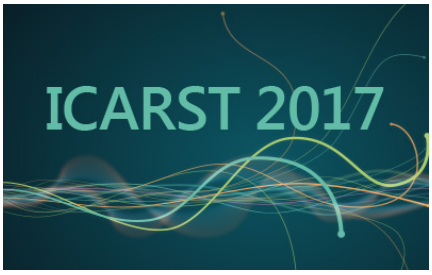


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# A Comparative Study of Radiation Sterilization of Cell Culture Media and Filtration Sterilization Method in Cell Culture Laboratory

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Cell culture media is uses in biopharmaceutical processes to stimulate the natural environment of the cell. Media used in cell culture have a balanced salt solution; the most commonly used is sodium bicarbonate with a pH of 7.2–7.4 at 37°C that the optimal growth of cells. Filtration is a safe method used for media sterilization in cell culture laboratory; it helps to removes the microorganisms, but is unable to separate microorganisms that have the same size. The most common types used in tissue culture has a pore size 0.2 $\mu$m. The aim of this study is using $\gamma$-radiation to sterilize media in cell culture laboratory compared to filtration sterilization method.

For media preparation, RPMI-1640 media in powder form, containing vitamins, minerals, amino acids and red phenol, was added to $NaHCO_3$ and dissolved in distilled water, and a pH meter was used for pH determination. A $^{60}$Co $\gamma$-irradiator was used for dose irradiation of 5 kGy and 20 kGy at 1.26 kGy/h dose rate. The irradiation was carried out at the Chemistry and Physics Institute of the Sudan Atomic Energy Commission.

Testing of bacteria included total viable aerobic count and total coliform count. Cell viability testing was made by adding 0.5 m$\ell$ of lymphocytes to 4.5 m$\ell$ from RPMI media, 1.2% penicillin/streptomycin, 1% fetal bovine serum, 30 $\mu$m$\ell$ Phytohaemagglutinin glutamine, and then incubated for 24 hours in $CO_2$ incubator. Cells were then counted using jumper.

A $\gamma$-radiation dose of about 20 kGy was found to be enough to destroy most type of bacteria rather than 5.0 kGy, but not suitable for sterile media cell culture in laboratory because increasing the probability of poor cell growth. While a dose of 5 kGy does not completely sterilize the media and cell growth observed. Physiology of media environment is summarized as follows for Lymphocytes growth and pH control for $\gamma$ radiation dose and filtration for sample media:

| ——————| ——————| ——————-| ——————-| ——————| ——————|

| Media | Cell Death | Cell Growth | pH (pre) | pH (post) | Media |

| Sterilization | (24 Hours) | (48 hours) | | | Colour |

| 5 kGy | 50% | 50% | 7.2 | 7.2 | change |

| 20 kGy | 70% | 10% | 7.2 | 8.0 | colourless |

| Filtration | 10% | > 60% | 7.2 | 7.3 | no change |

|——————|——————|——————-|——————|——————|——————|

Media can be sterilized by $\gamma$-radiation with doses larger than 5 kGy, but red phenol that indicator pH of media was degraded with increases the $\gamma$-dose. This result suggests that $\gamma$-doses larger than 5 kGy and less than 20 kGy should be tested to achieve the dose range accepted for cell culture media sterilization without affecting cell integrity. Then, radiation sterilization among RPMI media may useful for liquid RPMI media without red phenol, used in stem cell. Further research on red phenol degradation by $\gamma$-radiation, and amino acids concentrations post-$\gamma$ irradiation is strongly recommended, and might help in sterilization and modifications of media for various cell culture applications using $\gamma$-rays.

## Country/Organization invited to participate

Sudan

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