Academic Center for Education, Culture and Research, Iranian Biological Resource Center

Nov/30/2020

To whom it may concern,

The following cell line was provided to the user, Dr. Samira Derakhshan on, Nov/2019:

Cell Name: OCC-11 (IBRC C10987)

Prior to the provision, the following validation of the above cell line was performed:

Mycoplasma infection

Detection by PCR method
The cells cultured without antibiotic for three passages. The extracted DNA from cultured cells and supernatants were used in multiplex PCR reaction. After PCR-amplification of Mycoplasma DNA, detection was done by the gel electrophoresis. The result was shown no amplification for usual Mycoplasma strains in cell culture including *Mycoplasma hyorhinis.*

Detection by the culture isolation method
The broth-agar microbiological culture method was used to detect Mycoplasma contamination. Cell culture supernatant was inoculated to rich liquid media. The culture was incubated for at least seven days to enrich for Mycoplasma. Subsequently, samples of the suspensions were passaged to the agar plates and were incubated at least for 7 days. As the Mycoplasma colonies were not seen by microscopy, the passage was repeated and the result was demonstrated no sign of Mycoplasma growth in culture (3, 4).

Bacterial and Fungal contamination detection

The cells cultured without antibiotic for three passages. Cultured cells and supernatant were inoculated to Thioglycolate and Soyabean Casein Digest broth media and incubated for 14 days in 22°C and 32°C separately. No sign of contamination and turbidity was seen in broth cultures (8).

Viral contamination detection

Polymerase chain reaction was carried out to detect HBV (Hepatitis B virus), EBV (Epstein- Barr Virus), HIV (Human immunodeficiency virus) and HCV (Hepatitis C virus) contamination. The results did not demonstrate contamination to mentioned viruses (7).

Species identification

Verification of presumptive species and verification together with the exclusion of contamination by other animal cells was performed by standard multiplex PCR method (Including: Mouse, Rat, Rabbit, Camel, Horse, Cow, Sheep, Cat, Dog, Guinea Pigs, Pig, Rhesus monkey, African green monkey, Chinese Hamster, Human, Chicken), The result was demonstrated that this cell line was derived from Human (2,6).

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