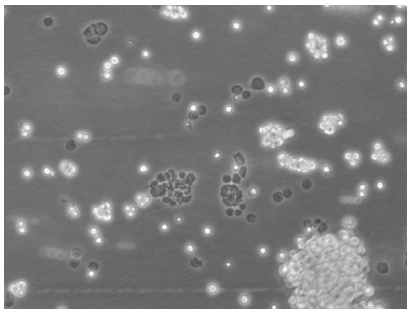
Cell line profile

Cell line profile B95-8 (ECACC catalogue no. 85011419)

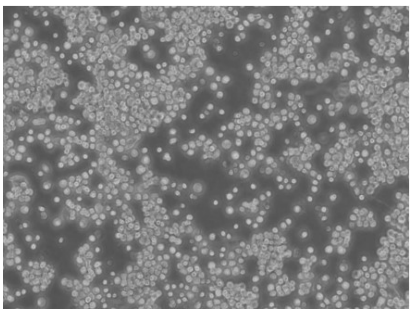
Cell line history

The B95-8 cell line's name originates from the B95 strain of Epstein Barr Virus (EBV). The viral strain was isolated from a case of transfusion induced mononucleosis and subsequently used in vitro to immortalise monkey blood leukocytes (Miller et al, PNAS, 1972) to generate the cell line.

Through the use of DNA profiling, it has now been shown that the B95-8 cell line was most likely originally derived from the Cotton-top Tamarin Monkey Peripheral Blood Lymphocyte as opposed to the commonly assumed origin of a marmoset. All 172,282 base pairs of B95-8's nucleotide sequence have been successfully mapped using the dideoxynucleotide/M13 sequencing procedure (Baer et al, Nature, 1984). This procedure uses DNA polymerase to synthesise DNA from a single stranded template, halting when one of four dideoxynucleotides (for each base) is incorporated. This incorporation produces an output which can be read, and after many cycles, gives the complete sequence of DNA (Yu, DNA Sequencing Methods, 2001). B95-8 is known for producing large quantities of EBV, however studies have shown that the DNA characterising this production was not present in the original strain. This therefore means that a mutation must have taken place shortly after the creation of the B95-8 cell line causing the production of EBV (Skare et al, JoV, 1982). EBV is a common human virus which spreads through bodily fluid (in particular saliva) and causes infections such as Mononucleosis (Mono) (Sumaya & Ench, Pediatrics, 2003).



48 hours post resuscitation



Prior to cryopreservation

Key characteristics

B95-8 cells originate from the blood of a cotton-top tamarin. The morphology of the cell line is predominantly a suspension of singular, ovular cells or clusters of small clumps, however roughly 10% of the cells remain adherent. The cells are lymphoblasts – immature cells which will become lymphocytes after antigenetic stimulation (Miller-Keane, 2003). Under normal cell culture conditions (37°C, 10% Serum) the cell line proliferates and does not produce optimum virus titres, however, reducing the temperature to 32°C and lowering the serum concentration results in highly efficient pathogenic virus production (Bolton & Spurr, 1996). B95-8 cells have also been reported to produce high titres of EBV when exposed to other EBV-inducing agents (Shaw *et al*, JoV, 1987).

Applications

The B95-8 cells are used as a source of EBV for the transformation and immortalisation of human B lymphoctyes into continuous cell lines (Romano et al, Nucleic Acids Research, 2009) (Shope et al, PNAS, 1973) (Bolton & Spurr, 1996). Being able to propagate cells indefinitely from specific donors ensures an indefinite supply of genomic material without the need to resample from the original donor. This technique has been used at ECACC to generate tens of thousands of patient specific lymphoblastoid cell lines over the last 30 years and was fundamental to the success of the UK human genome mapping project. An example of their use was in a genomewide scan for autism involving a resource of over 2000 cell lines generated by ECACC (Palferman et al. 2001). Cell lines immortalised by EBV transformation have been used to generate all the cell lines in the ECACC HLA Typed Collection and the Human Random Control DNA Panels.

Culture tips

B95-8 should be cultured in RPMI 1640 + 2mM Glutamine + 10% Foetal Bovine Serum (FBS).The culture should be maintained between 3-9 x 105 cells/ml at 37°C + 5% CO2. Due to its partial adherence, trypsin may be required to fully remove all of the cells from the flasks. During cryopreservation, the quantity of cells should be kept at a minimum of 8 x 106 cells/ampoule to ensure enough cells survive to resuscitate the cell line. These cells should be handled under laboratory containment level 2.

Key references

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Related cell lines	ECACC catalogue number	Description
B95a	01092505	Derivative of B95-8 adapted for more adherent growth. Much more vulnerable to the measles virus so useful in the isolation of measles. Culture in DMEM +2mM Glutamine + 5% FBS.