



Chemically Defined Animal-free Highly Consistency



Providing an excellent solution for culture media to be

Chemically Defined Animal-free Highly Consistency



Potential problems with the unknown contaminants, reliability of supply, and performance variability between batches, undefined cell culture supplements have seen an intensification in industry demand for more well-defined, well-characterized, and easily controlled media.

Using the State-of-the-Art platform called Oryz^{HIEXP}, having a high capacity to express proteins or small peptides in the rice grain, we have achievements in developing and commercializing a series of animal-free cell culture supplements, such as OsrHSA, OsrhTF, OsrhFN, and recombinant growth factors. The use of these supplements enabled complex culture media to be chemically defined, animal-free, and highly consistency, as we provide animal-free, cGMP raw materials of high consistency and purity. Moving towards chemically defined media is favored by the regulatory authorities as comprehensive quality information must be available for all their constituents.

Animal-free Cell Culture Supplements



OsrHSA

Recombinant Human
Serum Albumin



OsrhTF

Recombinant Human
Transferrin



OsrhFN

Recombinant Human
Fibronectin



OsrhbFGF

Recombinant Human
Basic Fibroblast Growth
Factor



rhEGF

Recombinant Human
Epidermal Growth
Factor



rhIGF-1 Lr3

Recombinant Human
Insulin-like Growth
Factor-1 LR3



OsrhVEGF

Recombinant Human
Vascular Endothelial
Growth Factor



OsrhKGF

Recombinant Human
Keratinocyte Growth
Factor

Advantages

- Elimination of the potential risk of viral and other unknown contaminants;
- Avoid the quality changes from different batches, improve the repeatability of cell culture and experiment results;
- Regulatory compliant with well-defined components;
- Stable supply and manufacture capability at large scales to meet bulk processing needs ;
- Simplify the downstream bio-processing or biological research;
- Low prices allow for a reduction in cost without sacrificing quality.



Recombinant Human Serum Albumin

OsrHSA

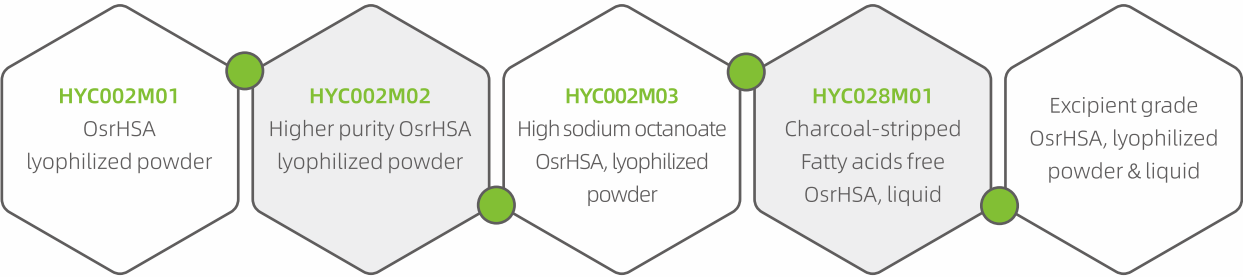


Table 1.1 Comparison of OsrHSA and pHSA

Char.	pHSA	OsrHSA
Amino Acids Sequence	Same	Same
N-terminus	DAHKSEV	DAHKSEV
C-terminus	KLVAASQAALGL	KLVAASQAALGL
Glycosylation	No	No
Molecular Mass(MALDI)	66.554KD	66.550KD
PI	4.8	4.8
Drug Binding	Similar	Similar
Thermal Stability	M/P 65°C	M/P 65°C
Lipid Conjugation	Identical	Identical
Crystal Structure	Identical	Identical

OsrHSA, a recombinant human serum albumin, genetically engineered and derived from a rice-based expression system, is a highly purified and completely animal-, virus-, and bacteria-free product that was developed as an alternative to plasma-derived HSA (pHSA), to which it is structurally, physiological and biochemical properties equivalent (Table 1.1). OsrHSA eliminates the risk of bacterial and viral contaminants. Additionally, the greater consistency of OsrHSA saves time while providing better performance.

Different various of OsrHSA offer you more options



Sodium octanoate is a stabilizer in commercial HSA, which is also a kind of fatty acid. Higher sodium octanoate OsrHSA can endure higher temperature and pH.

As a long-established ingredient of cell culture media, HSA is well recognized for its ability to facilitate growth of many cell types. Cell culture can face many challenges in regard to reproducibly optimizing, controlling and scaling cell isolation, expansion, transformation, cryopreservation, and final formulation. OsrHSA is suitable as a cell culture media supplement to promote cell growth (Figure 1.1 & 1.2). OsrHSA is shown consistently to perform better than pHSA and serum products and possesses numerous other supply chain reliability, regulatory, and safety benefits.

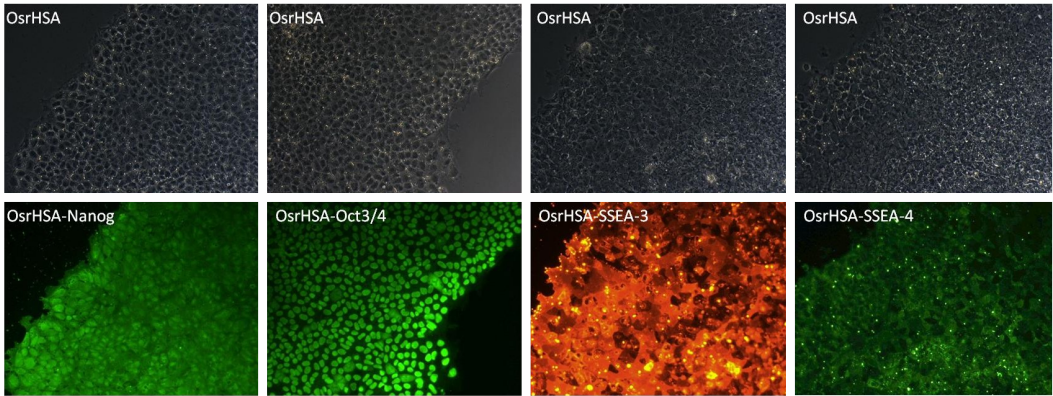


Figure 1.1 The immunofluorescent staining showed OsrHSA was effective in keeping undifferentiation and keeping viability of iPS cells.

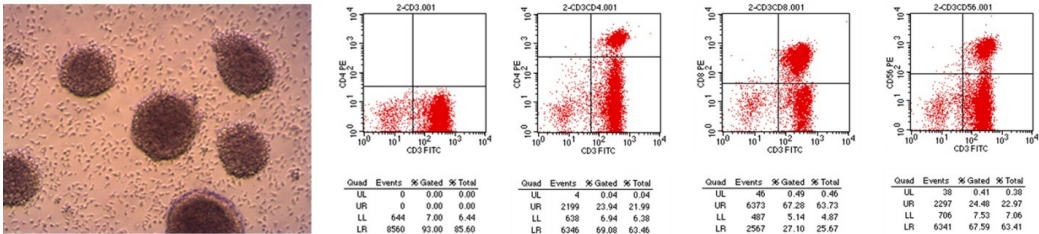


Figure 1.2 OsrHSA was used in PBMC culture to induce the CIK cells. After 14 days, the total cell amount reach to 6.6X10⁹, and the CD3+CD56+ rate is more than 22.97%. It showed that OsrHSA has effective action in promoting CIK cell growth.



Recombinant Human Transferrin

OsrhTF

Serum-free cell culture requires a form of iron delivery. Transferrin is the preferred delivery form of iron because cells can process transferrin-bound iron in a physiologically appropriate manner through transferrin receptors on the cell surface.

OsrhTF is a recombinant human transferrin derived from rice platform, which can provide bulk and stable supply, mainly used in serum-free cell culture to promote iron transport and cell growth. OsrhTF is animal component free, and high purity and can avoid the risk of contamination from human or animal-derived viruses and mycoplasma.

Moreover, compared with other recombinant transferrin, the cell culture data has shown our OsrhTF has equivalent or better effect on cell proliferation (Figure 2.1 & 2.2).

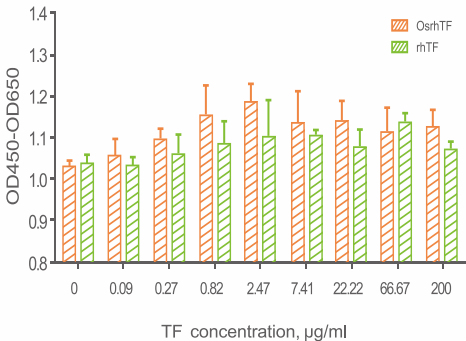


Figure 2.1 In reduced serum cell culture, OsrhTF performs better in different concentrations, compared with other commercially rhTF.

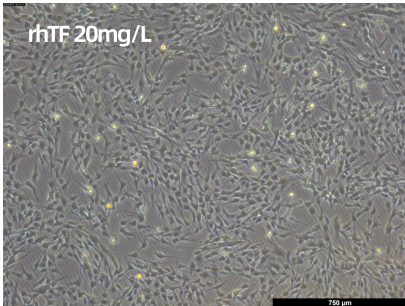
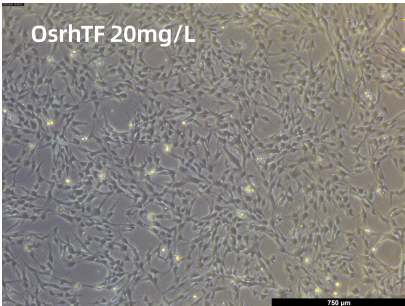
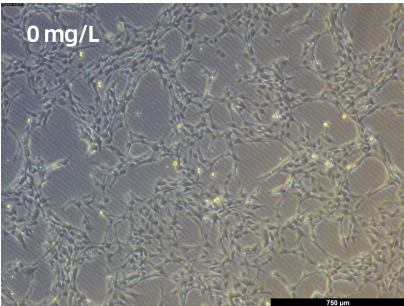


Figure 2.2 OsrhTF can significantly promote cell proliferation, and the effect is comparable to that of other commercially rhTF.

Fibronectin (FN) is a particularly important and well-studied component of the extracellular matrix, and is known to play a key role in cell adhesion, growth, spreading, migration, differentiation and proliferation. OsrhFN is a recombinant fibronectin expressed in the rice platform, which is animal component-free and has high purity (Figure 3.1), and has been demonstrated has the same physical and chemical with the plasma derived FN.

OsrhFN is a more defined and consistent cell culture substrate to replace Matrigel, rat tail collagen, blood-derived fibronectin, etc., improve cell adhesion rate, enhance cell metabolism and shorten cell growth cycle. In addition, it can also be directly added to the medium, which can replace or reduce the amount of serum, providing a safety solution.



Recombinant Human Fibronectin OsrhFN

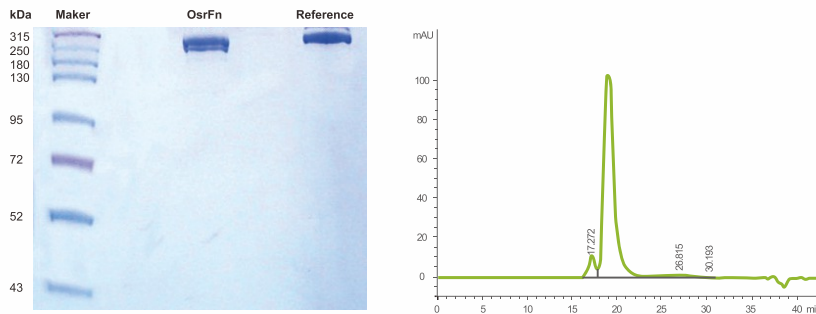


Figure 3.1 In the SDS-PAGE and SEC-HPLC purity analysis, the purity of OsrhFN is more than 95%.

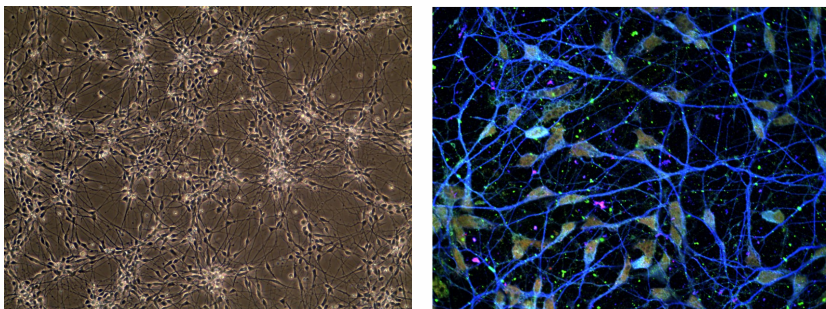


Figure 3.2 The morphology and immunofluorescent staining showed that OsrhFN was effective for keeping viability of neuron cells.



Recombinant
Human Basic Fibroblast
Growth Factor
OsrhbFGF

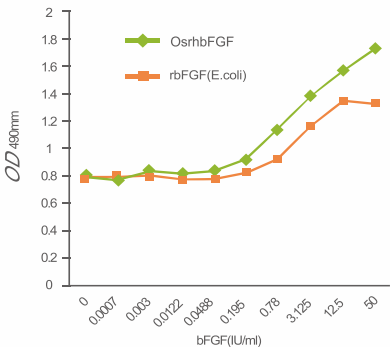


Figure 4.1 OsrhbFGF has a higher bioactivity than the commercial rbFGF derived from E.coli.

Human basic fibroblast growth factor, also known as bFGF, FGF2 or FGF- β , is a member of the fibroblast growth factor family. It is a single-chain polypeptide growth factor that plays a significant role in the process of promoting cell proliferation, inhibiting cell apoptosis of a wide variety of cell types. Using high-purity, high-bioactivity proteins is extremely helpful for the role of growth factor. OsrhbFGF is derived from rice platform, which is animal component-free with high purity to minimize interference from other contaminants. Rice platform makes the manufacture capacity at grams scale to meet bulk needs, providing consistent performance in cell culture media with low cost and time consuming.

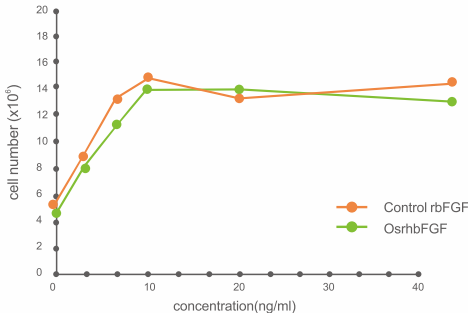
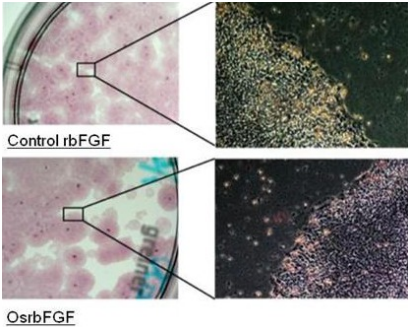


Figure 4.2 OsrhbFGF sample showed almost equal performance for feeder-free culture of iPS cells compared to control rbFGF from E.coli.



Recombinant Human Epidermal Growth Factor rhEGF

By binding with high affinity to epidermal growth factor receptor (EGFR) on the cell surface, EGF results in a variety of biochemical changes within the cell, protein synthesis and regulating the gene expression, including the gene for cell proliferation. rhEGF is developed as a supplement for animal-free cell culture, including a variety of fibroblast and epithelial cell types. It is guaranteed identical to human native EGF with high batch-to-batch consistency and optimal bioactivity.

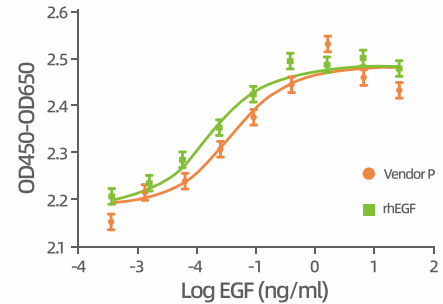


Figure 5.1 The stimulating cell proliferation of rhEGF measured using the Balb/c 3T3 cell line. The ED50 of rhEGF is less than 1ng/ml. Moreover, rhEGF performs better than other vendor's.

Insulin-like growth factor-1 (IGF-1) is a hormone similar in molecular structure to insulin. IGF-1 is one of the most potent natural activators of the AKT signaling pathway, which stimulates cell growth and proliferation. rhIGF-1 LR3 is an IGF-1 analog with the substitution of an Arg for the Glu at position 3 (hence R3), and a 13 amino acid extension peptide at the N-terminus. The rhIGF-1 LR3 has much higher stability and biological activity, providing a consistent, compliant and reliable alternative to recombinant insulin.

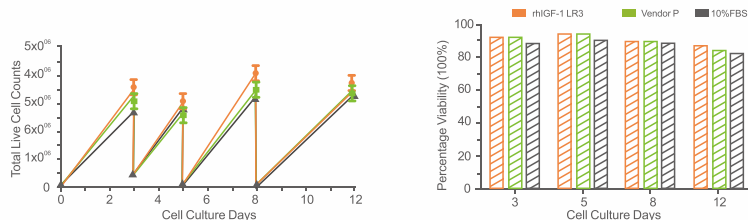


Figure 5.1 Comparison of stimulating cell proliferation activity of rhIGF-1 LR3 with another vendor, the result shows rhIGF-1 has equivalent effect.



Recombinant Human Insulin-like Growth Factor-1 LR3 rhIGF-1 LR3



Recombinant Human Vascular Endothelial Growth Factor **OsrhVEGF**

Vascular endothelial growth factor (VEGF), also known as vascular permeability factor (VPF), can highly specific stimulate vascular permeability increasing and cell external matrix degeneration, vascular endothelial cell migration, proliferation and vascularization.

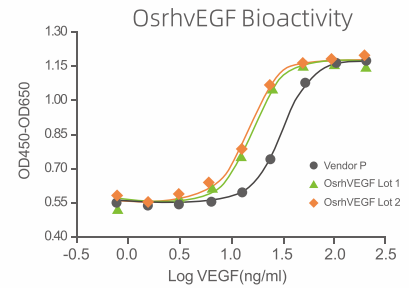


Figure 7.1 The cell proliferation assay using human umbilical vein endothelial cell (HUVEC) shows that OsrhVEGF has higher bioactivity and excellent batch consistency.



Recombinant Human Keratinocyte Growth Factor **OsrhKGF**

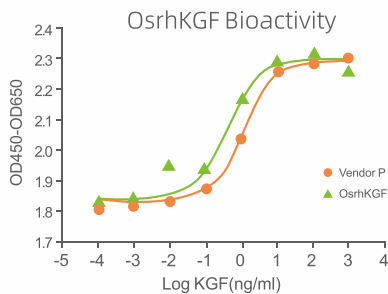


Figure 8.1 The cell proliferation assay using breast cancer cell (MCF-7 cell) shows that OsrhKGF has higher bioactivity.

Keratinocyte growth factor (KGF), also known as FGF-7, is a member of FGF family. Its mitotic activity is mainly manifested in keratinocytes, which can specifically stimulate the proliferation, migration and differentiation of epithelial cells.

OsrhVEGF and OsrhKGF, expressed in the rice platform and purified by chromatography, are animal component-free and have high biological activity. The rice platform makes the manufacture capability at gram scales to meet the bulk bioprocessing needs.



Wuhan Healthgen Biotechnology Corp. (Healthgen Biotech) develops and manufactures products for the biopharmaceutical industry under GMP compliant quality management system. We focus on the timely delivery of high quality, consistent, animal-free and cost-effective products by using the State-of-the-Art platform called Oryz^{HiExp}, having high capacity to express proteins or small peptides in the rice grain. We will continue to develop new products and refine our supplements to improve the safety of biopharmaceutical to ensure business continuity for our customers.



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