

Recombinant Human Serum Albumin: Oryzogen phyto-rHSA

Introduction: Human Serum Albumin (HSA) has varied medical, pharmaceutical and biotech uses, such as a 1) vaccine excipient 2) cell culture media additive to promote growth and survival of cultures, some clinically important 3) as part of medical devices, such as protein coatings 4) as a treatment for burn injuries, shock, hypoproteinemia, cirrhosis of the liver, and in *in vitro* methods for fertility.^{1,2} Because of its availability and properties, new uses for HSA are in development, such as in drug delivery.^{1,2} The annual global albumin market is \$5 billion and steady growth could double the size within a decade.³ HSA market segments include forms derived from human blood (pHSA) and recombinant forms (rHSA).

Challenge: A low-cost supply of animal-free rHSA would provide a safer alternative to that derived from plasma for both consumers and investors. While pHSA has been generally safe, contaminating blood pathogens remain an on-going issue, with greatest concern on the spread and severity of new emerging infectious diseases.^{1,2} Also, while pHSA is generally cost effective, when supplies have become limited, shortfalls were followed by price spikes and counterfeit product in some markets.^{1,2} Particularly for high-value products that require HSA in their formulation, as for example in media used for growing cultures for cell therapies, a low cost source of HSA free of animal pathogens protects the high cost of investment for the research, development and manufacture of product.¹

Solution Description: The best-in-class solution to challenges in the HSA market would provide for a stable source of rHSA from an animal-free source to avoid potential for animal pathogens. Product should be well-characterized and production scalable for meeting market demand at a reasonable cost. Certified manufacturing that easily achieves a desirable low-endotoxin product also would assist cost and facilitate scaling to reliably meet demand. Long shelf-life, proven safety, proven structural identity to pHSA and in the case of therapies, proven efficacy would demonstrate universality as a pHSA replacement.

Proposed Solution: Recombinant HSA from rice (Oryzogen branded phyto-rHSA) developed by Healthgen, Wuhan, China, and distributed by eEnzyme, Gaithersburg, Maryland provides a formulation of rHSA under ISO9001:2008 certified and GMP compliant manufacturing. It is an animal-free source that can meet large scale requirements with a planned annual capability of 40 metric tons by the end of 2020. The expression system, rice endosperm, has low level of endotoxin naturally which reduces difficulty of manufacturing the low-endotoxin product. Product suitable for cell media comes as a lyophilized powder with long shelf life, certified for 4 years from date of manufacture, again facilitating cost and supply. Research has shown Oryzogen phyto-rHSA has the same structure as pHSA, is suitable with stringent cell lines such as T cells, and is nontoxic in animal studies.^{1,4} Further research has shown low immunotoxicity and low immunogenicity for residual non-HSA proteins of the purified product.^{4,5} Forms suitable for the clinic and as an excipient are in development. A Phase I trial provided initial positive data for its safety in humans.

Technical Specifications: Lyophilized material suitable for cell media and like products. The Certificate of Analysis specifies, manufacture date, expiration date (4 years from manufacture date), freeze-dried powder, light yellow to white powder, reconstitutes in water in under 15 minutes, $\geq 99\%$ pure by SDS PAGE, total protein $\geq 90\%$, pH 5.0-7.5, endotoxin ≤ 0.125 EU/mg. Certificate of Origin specifies China. Structures as determined by x-ray crystallography have shown pHSA and phyto-rHSA to be equivalent¹. While HSA is considered non-glycosylated, minor glycosylation patterns have been analyzed and like other rHSA forms, show variance with pHSA.⁴ Oryzogen phyto-rHSA is expressed from rice endosperm, which is naturally low in endotoxin and lipid. Expression is scalable by growth of recombinant transgenic rice. Although phyto-rHSA has low lipid content, the lipid profile has been analyzed.¹ Technical specification on clinical grade material available upon request and comes in liquid formulation. Clinical trials and development of clinical grade

material is on-going. Clinical grade is provided as a 20% solution, 99.99% pure by SDS PAGE and HPLC analysis, endotoxin ≤ 0.01 EU/mg.

Target Market: Present market uses for Oryzogen phyto-rHSA are in high value products that must avoid animal-pathogens such as cell media, in vitro fertilization media, as an excipient and in use with medical devices. Future markets include human therapies. Although pHSA presently generally continues at lower cost for human therapies, Oryzogen phyto-rHSA will provide cost containment during short-falls of pHSA supply or in critical-need situations.

Summary: Oryzogen phyto-rHSA has shown value as a pHSA replacement to meet cost, quality and pathogen-free requirements. It will fill supply as demand accelerates for future HSA uses, and particularly when a highly-qualified pathogen-free source is needed. Other proteins expressed on the same platform, such as phyto-Fibronectin, will likewise meet the high future demands within the developing biotech marketplace.

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