

# Dextran sulfate 5 in cell media and medicine

## What is dextran sulfate

Dextran sulfate is a derivative of dextran, which is a polysaccharide consisting of an  $\alpha$ -D-(1 – 6) linear glucan with a low content of sidechains linked to the 3-carbon of glucose. Dextran sulfate is normally produced by sulfation of selected dextran fractions. Dextran sulfates exist in different variation depending on a sulfation grade - high sulfate content (16-20% sulfur) and low sulfate content (8-13% sulfur) and molecular weight of dextran – low molecular weight dextran sulfates (LMW-DS, around 5 kDa) and higher molecular weight dextran sulfates (10-2000 kDa).

## The role of LMW dextran sulfate in cell culture media

Dextran sulfate of low molecular weight – such as DS5 - is a widely used anti-coagulation agent<sup>1</sup> for cells and antibodies and has been used for this purpose by large providers of cell culture media for over a decade.

DS5 added to cell media prevents clumping, and cell clumping is usually a problem for several reasons:

- It obstructs accurate cell counting, cell monitoring and control of the cellular surroundings<sup>2,3</sup>.
- The transport of nutrients to the cell and transport of products originating from the cells may be impaired by cell clumping<sup>2,3</sup>.
- Cell clustering affects growth behavior of cells dramatically<sup>2-4</sup>.
- Aggregation reduces the rate of proliferation<sup>2,3</sup>.
- Sheer forces cause a considerably higher death rate on aggregated cells compared to forces exerted on single cells<sup>3</sup>.

## Recent studies on anti-coagulant properties of DS5

- In a study from 2016, dextran sulfate 5 kDa was shown to diminish cell-aggregation during culture of Chinese hamster ovary (CHO) cells. A combination of 1.3 g/L DS and 8.0 mg/L r-trypsin was found to remove the cell aggregations completely<sup>2</sup>.
- In a study from 2016, dextran sulfate with various molecular weight (4kDa, 15 kDa and 40 kDa) was added in various concentrations (up to 1 g/L) was added to suspension culture of two different recombinant CHP cell lines producing monoclonal antibodies. Both cell lines improved growth and viability regardless of the molecular weight and concentration of dextran sulfate<sup>1</sup>.
- In another study from 2007, DS5 was shown to inhibit apoptosis and increase protein production in CHO cells 1.9-fold compared to controls<sup>4</sup>.

- The anti-coagulant properties of dextran sulfates and its biological use were described in the literature as early as in 1945 beginning with Dr. Ingelman at Uppsala University in Sweden and are still used in the modern biological research and pharmaceutical industry.

## Cell culture media additive users

The major driver for the increasing demand of cell culture media is the growing life science research.<sup>5</sup> The cell culture media of highest quality that prevents cell clumping and gives highest cell yield and survival has been in demand the last decade thanks to the rapid development of biopharmaceuticals, oncology, stem cell research and more<sup>6</sup>.

The US and Europe are the largest consumers of cell culture media<sup>5</sup> because of increased government initiatives to support research, an increasing number of diseases and therefore increasing the number of pharmaceutical manufacturers.

The Asia-Pacific region is the third largest user region for cell culture media<sup>5</sup>. Important factors are the high number of research organizations, and healthcare infrastructure under fast development.

The market in the Middle East and Africa are also expected to grow due to higher awareness among people, demands for better health care and constantly rising income<sup>5</sup>.

## How common is dextran 5 sulfate in cell culture media

Dextran sulfate 5 is a widely-known anticoagulant<sup>2-4,4</sup>. However, some of the major players in the cell culture media do not always reveal the full ingredient list of their cell culture media although they can still use dextran sulfate in the formula. Companies selling cell culture media have often an extensive portfolio of media with different additives, formulations and raw materials adapted to specific cell lines where DS5 is often involved.

In addition to cell culture media, DS is also included in several commercially available anti-clumping agents that can be added separately to the cell culture media by the customer in their own laboratory. Some examples are:

- Anticlumping Agent A, Lonza. The Anticlumping Agent A from Lonza contains a concentrated solution of dextran sulfate. Concentrations and molecular weights are not specified.
- DS 5 for cell media, TdB Labs. This anti-clumping supplement is produced using dextran derived from a natural source - the bacterium *Leuconostoc mesenteroides* B512F – and has a unique physical property of being a white powder. This allows it to be used in the conditions where color and transparency are extra important for cell culture setting, for instance for live microscopy of growing cells. In the cosmetic industry, it is highly preferred with white color of dextran sulfate instead of traditional off-white or yellow when using it as toxin-free green additive to improve viscosity.

## Low molecular weight dextran sulfate as a medicine

Low molecular weight dextran sulfate with molecular weight of 5 kDa (LMW-DS or DXS) is today available for clinical use and is registered as Orphan Drug by EMA<sup>7,8</sup>.

Recent studies on the instant blood-mediated inflammatory reaction (IBMIR) show that DS5 inhibits both complement and coagulation activation in clinical islet transplantation setting<sup>9,10</sup>. DS5 therefore being actively evaluated as a medicine against IBMIR in islet transplantation and being compared to

today's state-of-the-art treatment based on heparin<sup>11</sup>. DS5 has been also shown to interfere with activation of human DC at multiple levels by reducing immunostimulatory properties, secretion of proinflammatory cytokines, phagocytic activity, and prevents TLR signaling in response to endogenous and exogenous agonists by inhibiting phosphorylation of I $\kappa$ B- $\alpha$  and activation of NF- $\kappa$ B<sup>12</sup>.

In a study from 2017, Dextran Sulfate nanoparticles have been evaluated for clinical use in Rheumatoid Arthritis and have been shown to be potentially useful nanomedicines for Rheumatoid Arthritis imaging and therapy<sup>13</sup>.

## Summary

Application areas for Dextran Sulfate 5 are broad and growing thanks to the profound effect of dextran sulfates on biological systems. Dextran sulfate of low molecular weight is being used both as an additive in biochemistry and biotechnology settings, as well as in different clinical settings locally and systemically in combination with other agents and active substances. The concentration and dosage in all applications are crucial factors for success, and today some unique properties of DS5 are available on the market for the first time.

## References

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