ActiSM medium

HYCLONE MEDIA AND SUPPLEMENTS

HyClone™ ActiSM™ cell culture medium has been formulated to provide high yields of recombinant proteins in cell culture processes when using Chinese hamster ovary (CHO) cell lines (Fig 1). The medium is chemically defined, animal-derived component-free (ADCF), and optimized for high-yield protein production. ActiSM medium does not contain any growth factors (such as insulin), peptides, hydrolysates, phenol red, or 2-mercaptoethanol, ensuring batch-to-batch consistency and increased cell culture process efficiency.

ActiSM medium is intended for use in combination with HyClone ActiPro™ medium and Cell Boost™ 7a and Cell Boost 7b supplements as part of the ActiPro system of media and supplements designed with adaptation and production in mind. The ActiPro system is suitable for general biomanufacturing with CHO cell lines such as CHO-GS, CHO-K1, CHO-DG44, and CHO-S.

Key features of ActiSM medium:

- Chemically defined ADCF formulation
- Does not contain hypoxanthine, thymidine, and glycine
- Recommended as the first step when adapting cells cultured in other lean media
- Accompanied with detailed protocols for use to maximize output

Specifications

ActiSM is a lean, chemically defined medium that does not contain glycine, hypoxanthine, or thymidine. This medium was developed to be used as the first step in adapting cells to the ActiPro system of media and supplements. Once cells have undergone the adaptation process, we recommend using ActiPro medium for production, in combination with Cell Boost 7a and 7b supplements.

Shelf-life

Please refer to the label for the expiry date of your ActiSM medium.

Storage

ActiSM powder medium should be stored in a dry environment, protected from light at 2°C to 8°C. Reconstituted ActiSM medium should be protected from light and stored at 2°C to 8°C.

Fig 1. ActiSM medium is designed to be used as the first step in adapting cells to the ActiPro system of media and supplements for high-yield production of recombinant proteins in CHO cell culture processes.

Suggested preparation

To ensure proper reconstitution, please follow the protocol for your ActiSM medium.

Required equipment and materials include:

a. Mixing vessel

b. Stirrer

Note: a magnetic stirrer can be used for small-scale reconstitutions up to 5 L. An overhead or bottom-mounted impeller is recommended for larger volumes.

c. Calibrated pH meter

d. Calibrated osmometer

e. Highly purified water such as water for injection (WFI)

Reconstitution of ActiSM powder medium

1. Fill a clean mixing vessel to 96% of the final volume with high-quality purified water, such as WFI, at ambient temperature (18°C to 25°C) and start mixing. For example, to prepare a 1 L solution of ActiSM medium, take approximately 960 mL of water.

   Note: the mixing should be vigorous enough to quickly draw the powder below the liquid surface, but not too fast to cause formation of air bubbles and excessive foaming.

2. Add 20.56 g/L ActiSM powder slowly to the vessel, avoiding formation of clumps. Mix until no clumps of powder remain. The solution will remain cloudy in this step, but should be clear of any clumps or dry powder residues.
3. Slowly add 4.8 mL/L of a 5 N NaOH solution or 2.4 mL/L of a 10 N NaOH solution and mix until solution is clear. Should be less than 5 min depending on batch size. After this step, the solution will be clear.
4. Add 1.8 g/L sodium bicarbonate ($NaHCO_3$) into the vessel and mix until dissolved.
5. Adjust the pH to between 6.90 and 7.35 by drop-wise addition of 5 N or 10 N NaOH or HCl. After adjusting, continue mixing for an additional 20 min to ensure that all components are completely dissolved.
   Note: the pH will gradually increase with longer mixing times. Use caution when adjusting pH. Over adjusting can cause the osmolality to be out of specification.
6. Adjust to the final volume with high-quality purified water, such as WFI, and mix for an additional 10 min.
7. Measure and record the final pH and osmolality.
   Expected values:
   - pH 6.90 to 7.55
   - Osmolality 285 to 345 mOsmol/kg
8. Sterilize immediately by membrane filtration. Use a low-binding filter membrane type, such as PVDF, PES or cellulose acetate. The medium is a clear yellow/brown liquid.
9. Store the reconstituted medium protected from light at 2°C to 8°C until use.

Preparation note

ActiSM powder medium can be conveniently reconstituted using single-use mixers, such as the Xcellerex™ XDM mixers (Fig 2).

Fig 2. XDM Quad Mixing System, with a powerful motor and magnetically locked impeller, effectively mixes even highly viscous materials.

General culture recommendations

1. Cultures should be incubated at 37°C in a 7.5% CO_2 environment.
2. Maintain adapted cells by establishing a mid-logarithmic growth phase subculturing schedule.
3. Suggested seeding density of cultures 3.0 × 10^5 cells/mL; viability should be > 90%.

Process conditions

ActiSM medium is recommended for use in a CO_2 atmosphere. Equilibration of ActiSM medium in 7.5% CO_2 will result in a starting pH of 7.15 ± 0.05. During the cultivation, pH control can be managed by varying the CO_2 concentration or by addition of base such as $NaHCO_3$ or NaOH. The culture temperature should be adjusted according to the requirements of the specific clone or target product. ActiSM medium has demonstrated excellent results both under constant temperature conditions and in biphasic processes comprising a shift to a lower temperature.

Direct adaptation

Transfer cells grown in your current serum-free medium directly into ActiSM medium at 3.0 × 10^5 cells/mL. Passage cells every third to fourth day. Adaptation of most cell lines is complete once cells have reached a constant growth rate. As ActiSM medium does not contain insulin, please contact your sales representative for advice on how to handle insulin-dependent clones.

Sequential adaptation

Sequential adaptation can be accomplished by beginning with a 50/50 mixture of existing medium to test medium and culturing as above, reducing the concentration of the existing medium with each passage. Generally cells will adapt within 3 to 4 passages. If sluggish growth is observed, stay at the current medium mixture until the culture has recovered. A back-up culture is recommended until adaptation is complete.

Cryopreservation

Adapted cells can be cryopreserved in ActiSM medium supplemented with 5% to 10% DMSO. We recommend freezing the cells at a minimum cell density of 1 × 10^7 cells/mL.

Custom production

Formulations and delivery systems can be customized to your specific process requirements or optimized to maximize process yields.

Rapid Response Production (RRP)

Our RRP program manufactures up to 200 L of your custom prototype formulation within seven working days of your request. Use our RRP service to expedite the development and testing of custom media for your biopharmaceutical manufacturing process.

Related products

Product specifications for ActiSM medium and related products are listed in Table 1.

Table 1. Product specifications for ActiSM medium and related products

Specification	ActiSM	ActiPro	Cell Boost 7a	Cell Boost 7b
L-glutamine	-	-	-	-
Glucose	Y	Y	Y	-
Phenol red	-	-	-	-
Proteins	-	-	-	-
Hydrolysates	-	-	-	-
2-mercaptoethanol	-	-	-	-
Poloxamer 188	Y	Y	Y	-

ActiPro medium

ActiPro is a rich, chemically defined, and ADCF medium that does not contain hypoxanthine or thymidine and has been formulated for use in high-yield batch or fed-batch processes. Once your process has been adapted for use with the ActiSM medium, we recommend utilizing ActiPro medium for production.

Cell Boost supplements

Cell Boost 7a and Cell Boost 7b are highly concentrated, chemically defined ADCF supplements that help increase cell culture performance and product yield. Cell Boost 7a and 7b are intended for use together in defined concentrations.

The supplements should be added to the cultivation vessel as individual solutions and should not be mixed in advance as this will cause precipitation. The recommended ratio of Cell Boost 7a to 7b is 10 to 1 (v/v). The total amount of feed added and the specific feeding regime will need to be adjusted according to the nutritional requirements of each specific clone.

Cell Boost 7a

Cell Boost 7a has a pH close to neutral and contains amino acids, vitamins, salts, trace elements, and glucose.

Cell Boost 7b

Cell Boost 7b has an alkaline pH and is a concentrated solution of amino acids.

Technical support

Our cell culture medium specialists and technical support functions are happy to discuss your needs in getting the most out of your culture. In addition, we have an extensive service offering to help with, for example, process development, optimization, and scale-up. Please contact your local sales representative to learn more about the services we offer.

To find a certificate or a MSDS for a specific product, please visit cytiva.com/certificates.

Ordering information

Product	Quantity	Product code
HyClone ActiSM powder medium	5 L*	SH31038.01
	10 L†	SH31038.02
	25 L†	SH31038.05
HyClone ActiSM liquid medium	500 mL*	SH31040.01
	1000 mL*	SH31040.02
	1 L†	SH31040.03

Product	Quantity	Product code
HyClone ActiPro powder medium	5 L*	SH31037.01
	10 L†	SH31037.02
	25 L†	SH31037.05
HyClone ActiPro liquid medium	500 mL*	SH31039.01
	1000 mL*	SH31039.02
	1 L†	SH31039.03
HyClone Cell Boost 7a powder supplement	1 L†	SH31026.07
	5 L*	SH31026.01
	10 L†	SH31026.02
	25 L†	SH31026.03
HyClone Cell Boost 7b powder supplement	0.5 L*	SH31027.01
	1 L†	SH31027.07
	5 L†	SH31027.02
	10 L†	SH31027.04
L-glutamine 200 mM	100 mL*	SH30034.01
	500 mL*	SH30034.02
	500 g†	SH30336.03

Note: powder product quantity is shown as the final volume of after powder reconstitution.
* Stock items.
† Item is made to order. Lead times and minimum order quantities apply.

cytiva.com/hyclone

For local office contact information, visit cytiva.com/contact

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