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Serum-free T-Cell Expansion Medium Set

Product:

Product	Catalogue-No.	Size	Description
Nobimpex 003	N103-1000	1000 ml	Serum-free T-Cell Culture Medium. Without L-glutamine, be used to promote the optimal expansion of T-Cells.
Nobimpex 004	N104-0050	50 ml	Serum-free T-Cell Growth-Promoting Solution. With L-glutamine, For the first five days culture and Expansion of T Cells in adoptive cellular immunotherapy.

Description:

Serum-free T-Cell Expansion Medium has been developed to promote the optimal expansion of T-Cells of human origin. This medium supports high viable cell densities. The elimination of serum reduces performance variability in the medium and eliminates safety risks associated with possible adventitious agents in serum.

Introduction:

The immune system is composed of many different types of white blood cells the most common of which is the lymphocyte. A specialized type of lymphocyte called a T-cell is responsible for orchestrating the cellular arm of the immune response against cancer or infectious diseases. There are a variety of medical conditions in which patients' T-cells are present in low numbers or are not functioning properly. These clinical conditions place patients at high risk for infections and cancer. Adoptive immunotherapy is the ex-vivo manipulation and expansion of antigen specific T-Cells for subsequent administration into patients. The effectiveness of T-cell mediated immunotherapy depends upon a number of conditions after ex vivo expansion such as the fold expansion, the functionality, polyclonality, and antigen-specificity of the T-cells, etc. And the early expansion of cell is very important to meet those goals. To this end, we have developed a set of serum-free medium, Serum-free T-Cell Growth-Promoting Solution(Nobimpex 004) for the optimal expansion of T-cells during the first five days, and Serum-free T-Cell Culture Medium((Nobimpex 003) be used in another days.

Storage conditions:

The medium set is stable when stored at 2-8 °C and protected from light, until the date indicated on the label.

Composition:

The Serum-free medium set are proprietary formulation. And both the medium do not contain antibiotics or cytokines. Human serum albumin and human transferrin are the only human origin materials and are non-reactive (donor level) for anti-HIV 1 & 2, anti-HCV and HBsAg. Handle as if potentially infectious.

Preparation of complete media:

The medium set is supplied as a sterile (1X) liquid and must be supplemented with stimulatory antibodies, cytokines and/or antibiotics, if desired. Serum-free T-Cell Expansion Medium(Nobimpex 003) must also be supplemented with L-glutamine, add 20ml of 200mM L-glutamine solution or 0.584 g powder (irradiated) per liter of medium.

We recommend that Nobimpex 004 medium be used in two times(25 ml each) during the first five days Expansion of T cells , and Nobimpex 003 medium be added as well in another days .

Procedure:

1. Add certain amount of stimulatory antibodies and cytokines in 25 ml Nobimpex 004 medium.
2. Prepare either fresh or frozen PBMNCs (peripheral blood mononuclear cells) as directed by the supplier or in accordance with established protocols.
3. Count cells using a hemacytometer.
4. Transfer the proper number of cells to the desired culture vessel containing medium supplemented with cytokines and stimulatory antibodies (and antibiotics if desired).
5. Place the culture vessel in a humidified incubator at 37°C and 5% CO2.
6. Day 3, add other 25ml the Nobimpex 004 medium supplemented with cytokines and stimulatory antibodies (and antibiotics if desired).
7. Day 5 and beyond, add Nobimpex 003 medium which also supplemented with cytokines and stimulatory antibodies (and antibiotics if desired) with optimal amount.

Product Profile:

Nobimpex's Serum-free T-Cell Expansion Medium set(Nobimpex 003&004) demonstrated rigorous expansion of T-cells from PBMNCs. This product was compared with several other commercially available serum-free expansion media for their ability to expand T-cells in T75 culture flasks. For these small-scale experiments, 200,000 PBMNCs/ml were incubated for up to 7 days in Serum-free medium or other commercial product containing 100 IU/ml IL-2 and anti-CD3 (OKT3, 20ng/ml) antibody.

The expanded T-Cell population from these experiment was then used in 51chromium release assays to test for functional cytolytic potential. Briefly, target cells (K562, a human chronic myelogenous leukemia cell line), were labeled with 51 Chromium and linked to anti-CD3 (OKT3) via Fc receptor. When mixed with effector or cytolytic T-cells, the target cells undergo apoptosis or lysis and release 51 Chromium. The amount of 51 chromium release into the supernatant is proportional to the number of targets killed and the number of functional cytolytic T-Cells.

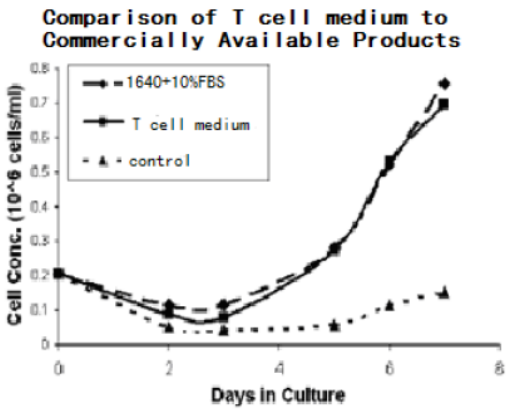
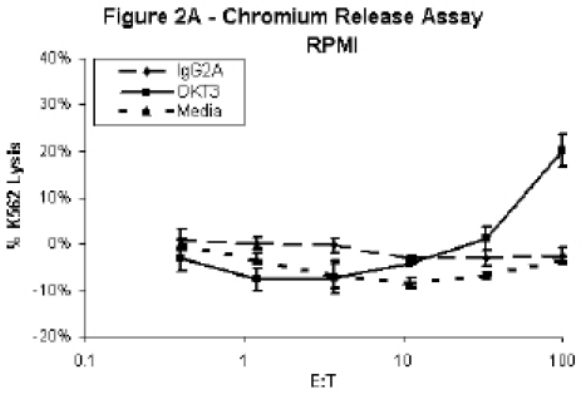


Figure 1. Peripheral Blood mononuclear cells wereincubated in either RPMI 1640 + 10% FBS, Control Medium or Serum-freeT-Cell Expansion Medium. Cells were stimulated using OKT3 anti-CD3 (20ng/ml) in the presence of 100 U/ml IL-2.



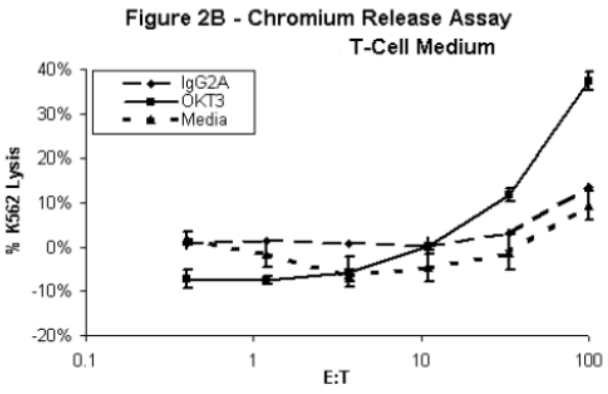


Figure 2A/B. In order to measure cytolytic potential from the harvested T-cells in Figure 1, a re-targetedlytic assay was used. Briefly, anti-OKT3 was linked tochromium-labeled K562 target cells via F_c receptor.Harvested T-cells from either control medium (Fig. 2A,RPMI 1640 or Fig 2B, Stemline T-Cell Expansion Medium) were then incubated at various effector-to-target ratios and the level of polyclonal cytolytic activity was determined as percent K562 lysis.