

iLite[®] IL-23

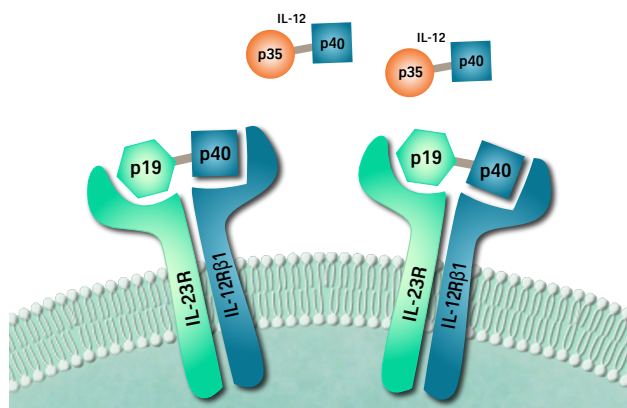
ASSAY READY CELLS

iLite IL-23 Assay Ready Cells are a highly specific genetically engineered reporter gene cell line, optimized to respond to IL-23 with specific, proportional expression of Firefly Luciferase without cross-reactivity to IL-12.

The cytokine IL-23 has been implicated as an inflammation mediator in several autoimmune diseases, and therapeutic agents targeting IL-23 and the related cytokine IL-12 are currently used clinically to treat psoriasis and psoriatic arthritis. In addition, related agents are in clinical testing for a variety of inflammatory disorders. Recent studies have also shown IL-23 to promote tumor growth, making it an interesting target also for cancer therapeutics.

The *iLite* IL-23 Assay Ready Cells can be used as both a sensitive and specific assay for the quantification of the potency of anti-IL-23 or anti-p40 drugs. Despite the common p40 subunit between IL-23 and IL-12, the *iLite* IL-23 Assay Ready Cells respond specifically to IL-23, allowing the use of the assay also when developing specific p19 inhibitors. When developing anti-p40 drugs, both IL-23 Assay Ready Cells and IL-12 Assay Ready Cells can be tested to determine which gives the best basis for an anti-p40 assay. In addition, this product also provides a very effective means of quantifying the neutralizing antibody response against IL-23 inhibitors in human serum in the absence of serum matrix effects.

- Easy and fast assay format, completed within 6 hours
- Highly sensitive, without cross-reactivity with IL-12
- Eliminates serum matrix effects & renders assay results independent of cell number
- Over 15x fold induction and large assay window



Schematic illustration of the over-expression of IL-23 receptor on the cell surface, which has no cross-reactivity with IL-12.

iLite[®] IL-23 Assay Ready Cells	
Product code	BM4023
Format	Assay Ready Cells
Related Products	Complementary Products
BM3044 <i>iLite[®]</i> TNF-alpha Assay Ready Cells	BM4012 <i>iLite[®]</i> IL-12 Assay Ready Cells
BM4050 <i>iLite[®]</i> GM-CSF Assay Ready Cells	
BM3049 <i>iLite[®]</i> Type I IFN Assay Ready Cells	
Application	The <i>iLite</i> IL-23 Assay Ready Cells can be used for the quantification of IL-23 or p40 inhibitor activity, quantification of functional IL-23 or the neutralizing antibody response against such inhibitors in human serum. The following application notes are available: <ul style="list-style-type: none"> • Quantification of IL-23 inhibitor activity using <i>iLite</i> IL-23 Assay Ready Cells • Determination of neutralizing antibodies against IL-23 inhibitors using <i>iLite</i> IL-23 Assay Ready Cells • Quantification of functional IL-23
Incubation time	Drug Assays 30 min + 5 hours NAb Assays 30 min + 30 min +5 hours
Detection system	Luminescence
Availability	Research Use Only (RUO)*

**These products are intended for professional research use only. The data and results originating from using the products, should not be used either in diagnostic procedures or in human therapeutic applications.*

In accepting delivery of iLite[®] Assay Ready Cells the recipient agrees not to sub-culture these cells, attempt to sub-culture them or to give them to a third party, and recipient is only to use them directly in assays. The iLite[®] cell-based products are covered by patents which are the property of Svar Life Science AB and any attempt to reproduce the delivered iLite[®] Assay Ready Cells would constitute an infringement.

Svar Life Science AB

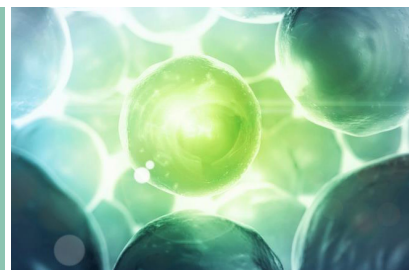
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iLite[®] Reporter Gene Assay for Quantification of the Activity of Anti-IL-23 and Anti-IL-23 Neutralizing Antibodies

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Introduction

Interleukin 23 (IL-23) is a heterodimeric pro-inflammatory cytokine that shares a common p40 subunit and a common receptor chain with IL-12. Both cytokines exert however distinct non-redundant biological functions. Conventional assays for IL-23 activity are based on the ability of IL-23 to support the proliferation of cell lines such as the IL-2 dependent human T-cell line Kit 225, which has been reported to partially lose dependence on IL-23, rendering the routine use of such assays problematic.

The IL-23 Gene reporter cell line

In order to reduce assay time from 2 days or more required for a conventional IL-23 bioassay, to 6 hours or less, and to obviate non-specific activation by other cytokines or growth factors with overlapping biological activity, a reporter gene assay was established using the avian B-cell line DT-40 that does not require IL-23 or other human cytokines in order to proliferate and that is unresponsive to the growth factors present in human serum.

Thus, DT-40 cells were co-transfected with the IL-23R and IL-12Rb1 receptor chains together with a STAT5 responsive Firefly luciferase (FL) reporter gene construct, and the Renilla luciferase (RL) normalization gene under the control of a constitutive promoter (Figure 1).

Assay performance

The IL-23 responsive cells (DT4023L) exhibit a 30-fold or greater increase in IL-23 induced FL activity 5 hours after treatment with increasing concentrations of IL-23 (Figure 2). The IL-23 responsive cells provide a highly sensitive assay for the quantification of IL-23 activity with an EC50 of 1.0 ng/ml or less and a lower limit of quantification of 100 pg/ml (Figure 2). The assay is also highly selective with no detectable cross-reactivity with IL-12 (Figure 2) even though both cytokines share a common p40 chain and a common receptor chain.

Assay normalization

Both FL and RL activity are read sequentially in the same well of a micro-titer plate using Dual Glo™ (Promega).

This allows IL-23 induced FL activity to be normalized with respect to the constitutive expression of RL activity thus rendering the assay independent of cell number and providing a very effective means for compensating for serum matrix effects.

Applications

The IL-23 responsive reporter gene cell line (DT4023L) can be used as both a sensitive and specific assay for the quantification of IL-23 activity or for the quantification of the potency of anti-IL-23 or anti-p40 monoclonal antibodies (Figure 3).

This cell line also provides a very effective means of quantifying the neutralizing antibody response against such products in human serum in the absence of serum matrix effects.

Conclusion

- IL-23 responsive cell line: quantification of IL-23, anti-IL-23 or anti-p40 activity within 6 hours
- IL-23 response without any response from IL-12
- Normalized readout: eliminates serum matrix effects & renders assay results independent of cell number
- Assay-Ready Cells available for 96 well plates
- Detailed assay description is available.

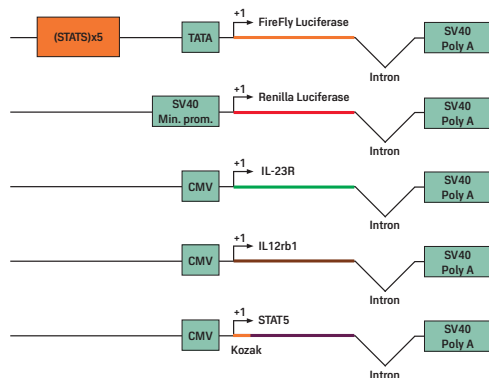


Figure 1. Molecular constructs of the IL-23 Reporter Gene Cell Line.

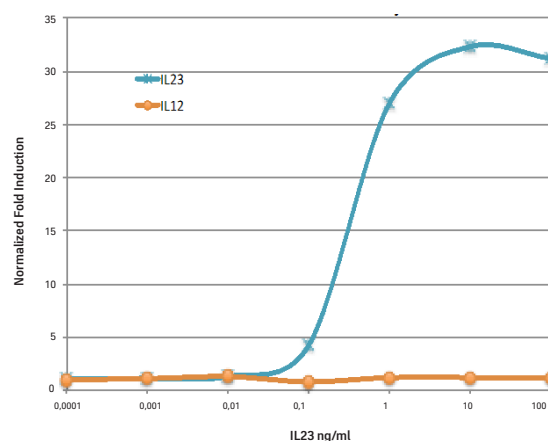


Figure 2. Response curve for the *iLite* IL-23 responsive Reporter Gene cell line, showing no cross-reactivity with IL-12.

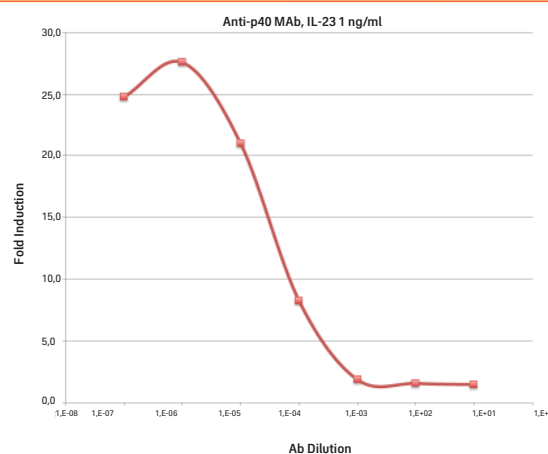


Figure 3. Quantification of an anti-p40 monoclonal antibody, using the *iLite* IL-23 responsive reporter cell line.