L-Carnitine: a nutritional modulator of the glucocorticoid receptor

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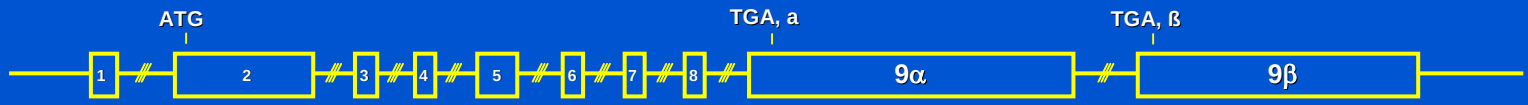
Glucocorticoids

- Steroid hormones secreted by the adrenal gland basally, and increasingly in response to stress
- Key regulators of carbohydrate, lipid and protein metabolisms
- Potent anti-inflammatory and immunomodulatory effects
- Essential for the normal functioning of the central nervous system, and the maintenance of cardiovascular, metabolic and immune homeostases
- Treatment of choice for many inflammatory, autoimmune and lymphoproliferative disorders
- Physiologic and pharmacologic actions mediated by intracellular receptors, the glucocorticoid receptors (GRs)

Human Glucocorticoid Receptor

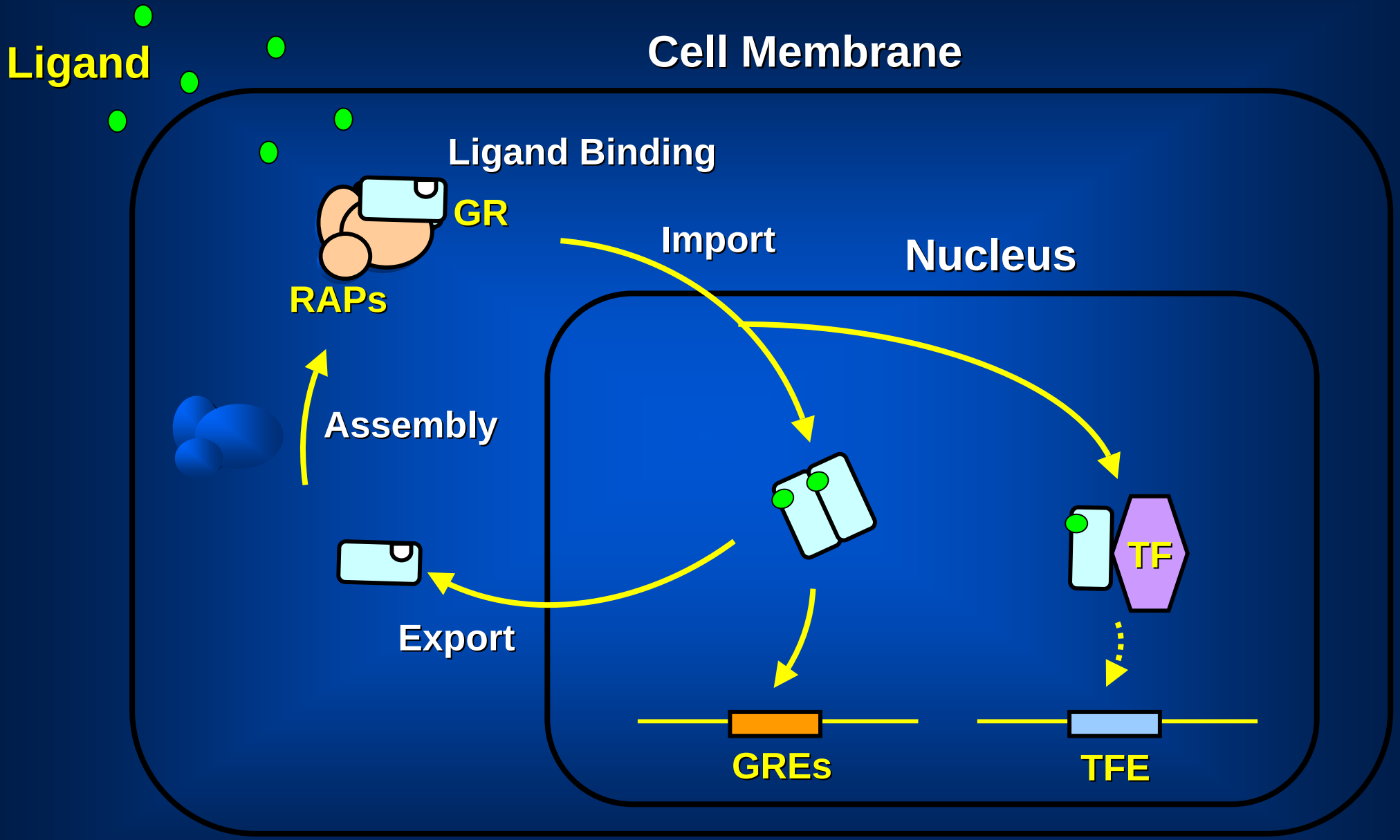


Chromosome 5

GRα cDNA

GRβ cDNA

GRα


Ligand
Cell Membrane
Ligand Binding
GR
RAPs
Import
Nucleus
Assembly
Export
TF
GREs
TFE

Glucocorticoid-like effects of L-Carnitine

- **Suppression of LPS-induced cytokine production in rodents**

  (Med. Inflamm. 2:S43-50, 1993; Br. J Cancer 72:1173-79, 1995)

- **Reduced *ex vivo* release of TNFα by *S. aureus-stimulated* human PMNCs**

  (Med. Inflamm. 2:S37-41, 1993)

- **Decreased serum TNFα levels in surgical and AIDS patients**

  (Med. Inflamm. 2:S33-36, 1993; Immunopharmacol. Immunotoxicol. 15:1-12, 1993)

- **Equal efficacy of L-Carnitine and betamethasone administered to pregnant rats in increasing the dipalmitoyl-phosphatidylcholine content of the fetal lung**

  (Pediatr. Res. 18:1246-52, 1984; J. Perinat. Med. 24:591-99, 1996)

Hypothesis

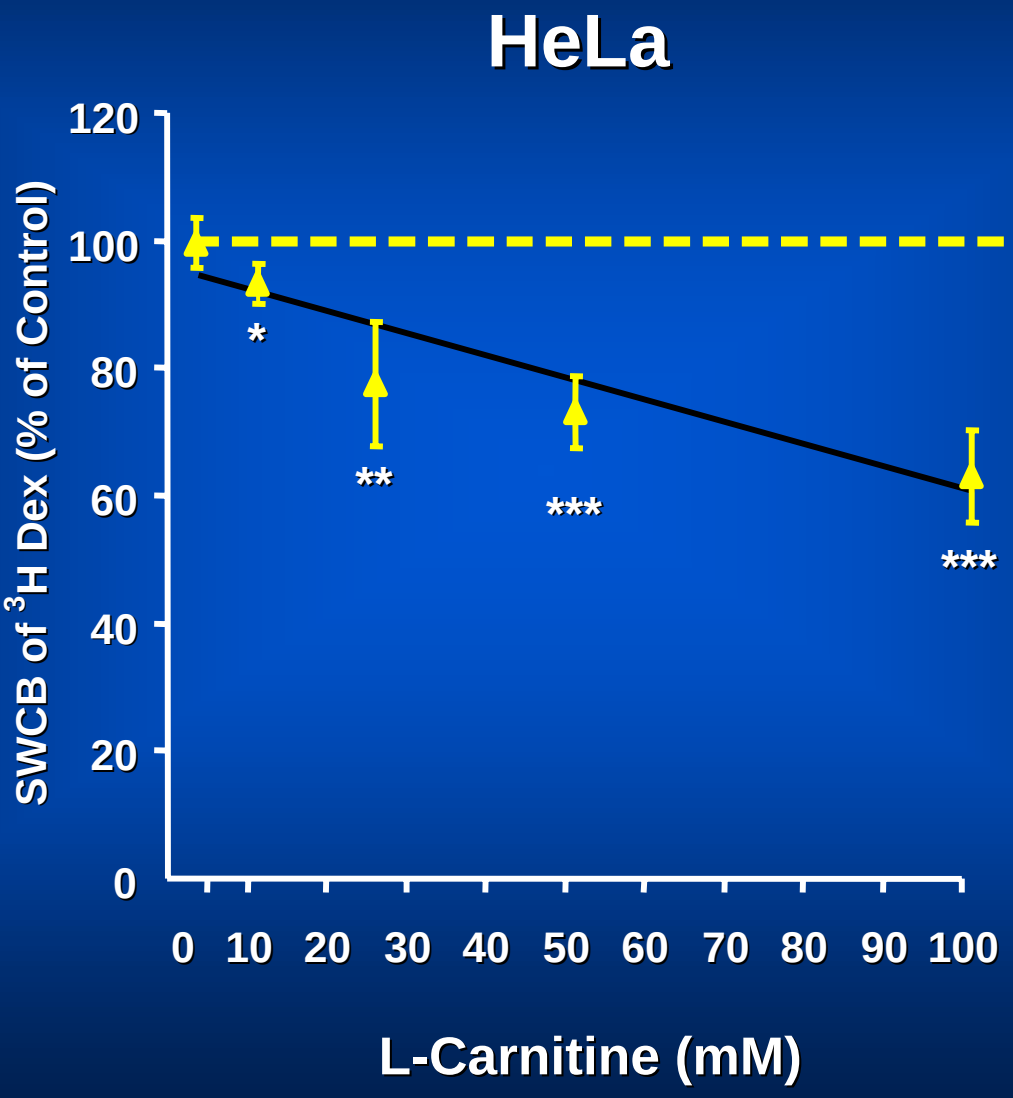
The Glucocorticoid-like effects of high L-Carnitine doses may be mediated via direct modulation of GRα function

Objective

To study *in vitro* the effect of L-Carnitine on:

- GRα
  - Binding capacity
  - Cell trafficking
  - Transcriptional activity
  - Biological activity

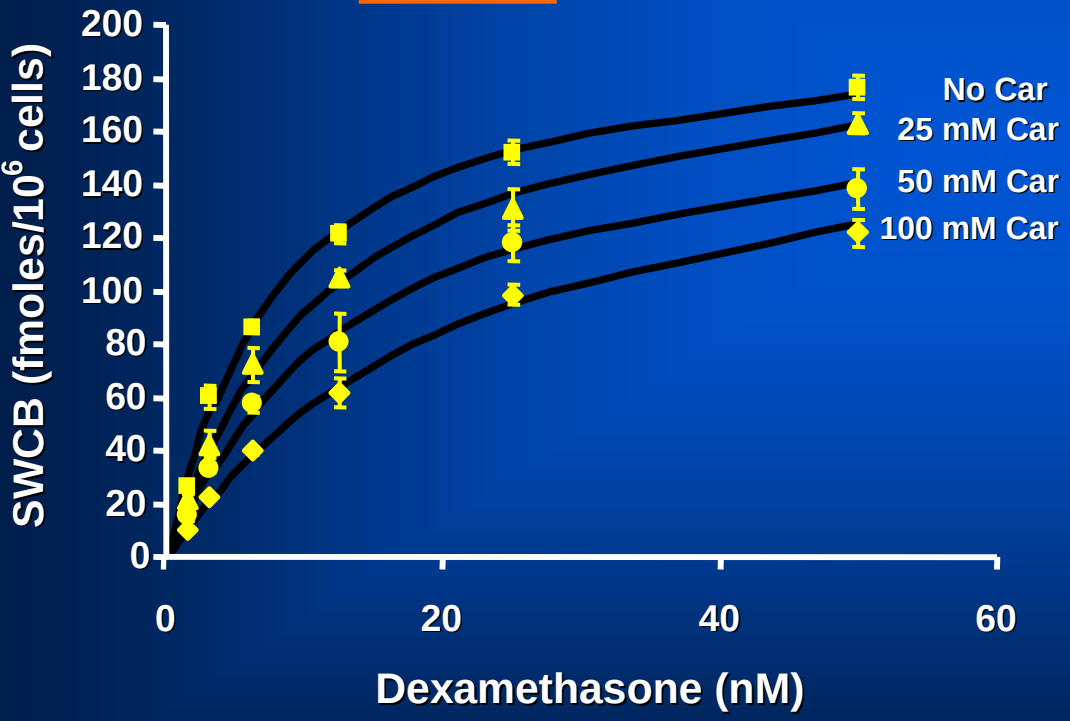
Competitive Binding

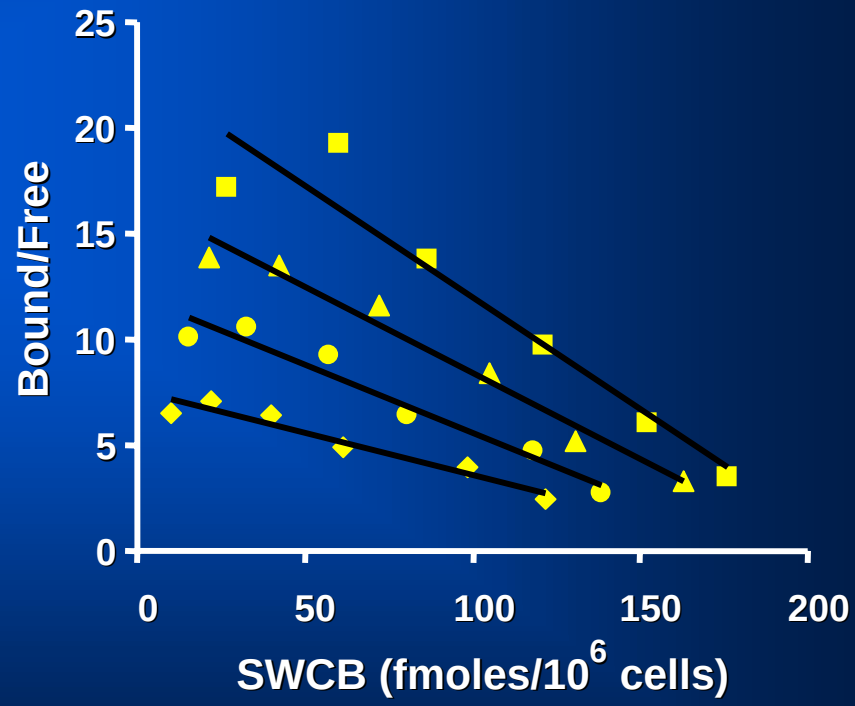


Saturation Binding

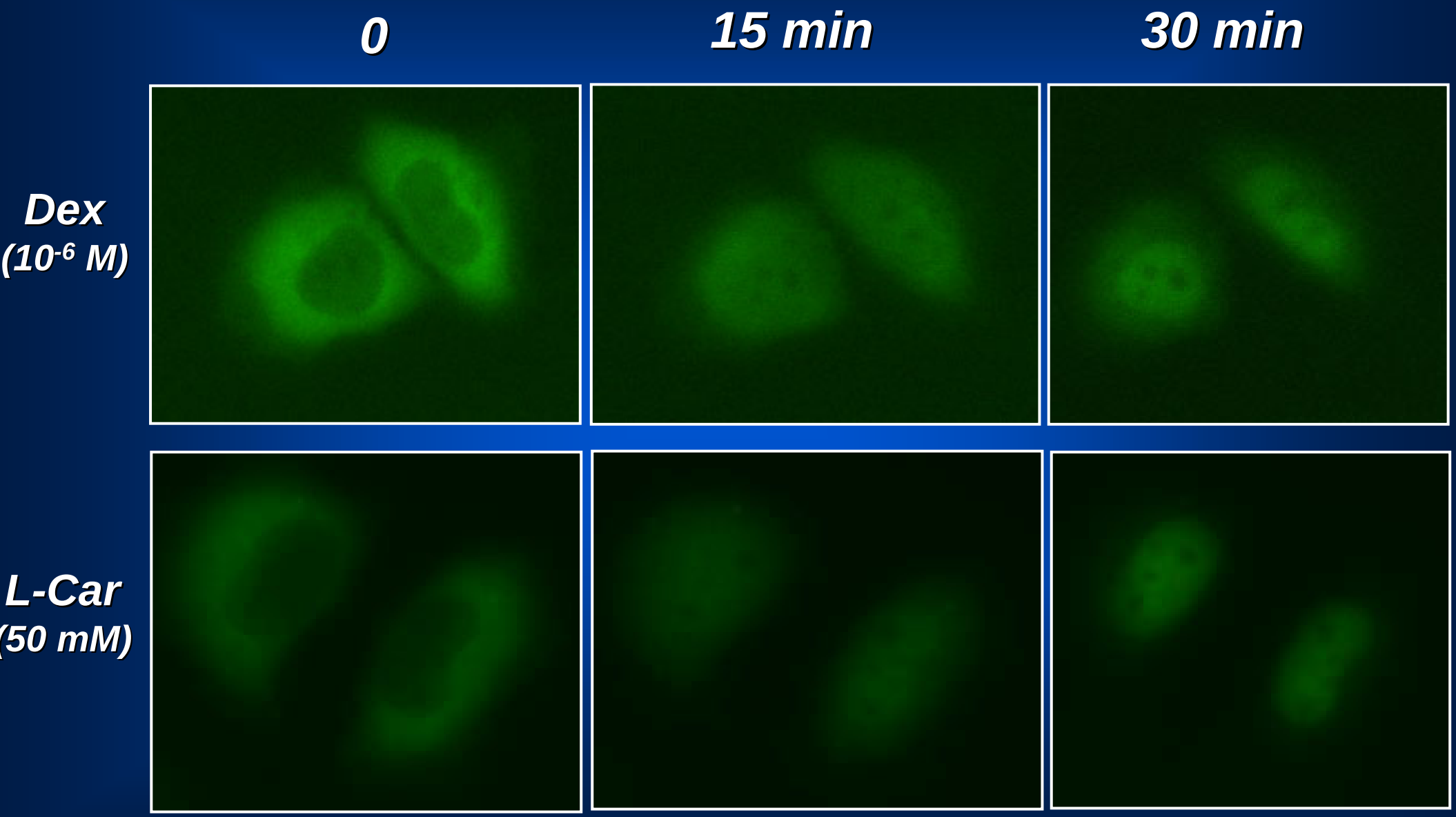
HeLa

No Car:	B_{max} = 202.1 +/- 6.46	K_d = 8 +/- 0.633
25 mM Car:	B_{max} = 201.7 +/- 4.46	K_d = 11.8 +/- 0.696
50 mM Car:	B_{max} = 180.5 +/- 7.72	K_d = 14.0 +/- 0.919
100 mM Car:	B_{max} = 183.2 +/- 11.9	K_d = 23.0 +/- 2.100





GRα-GFP translocation



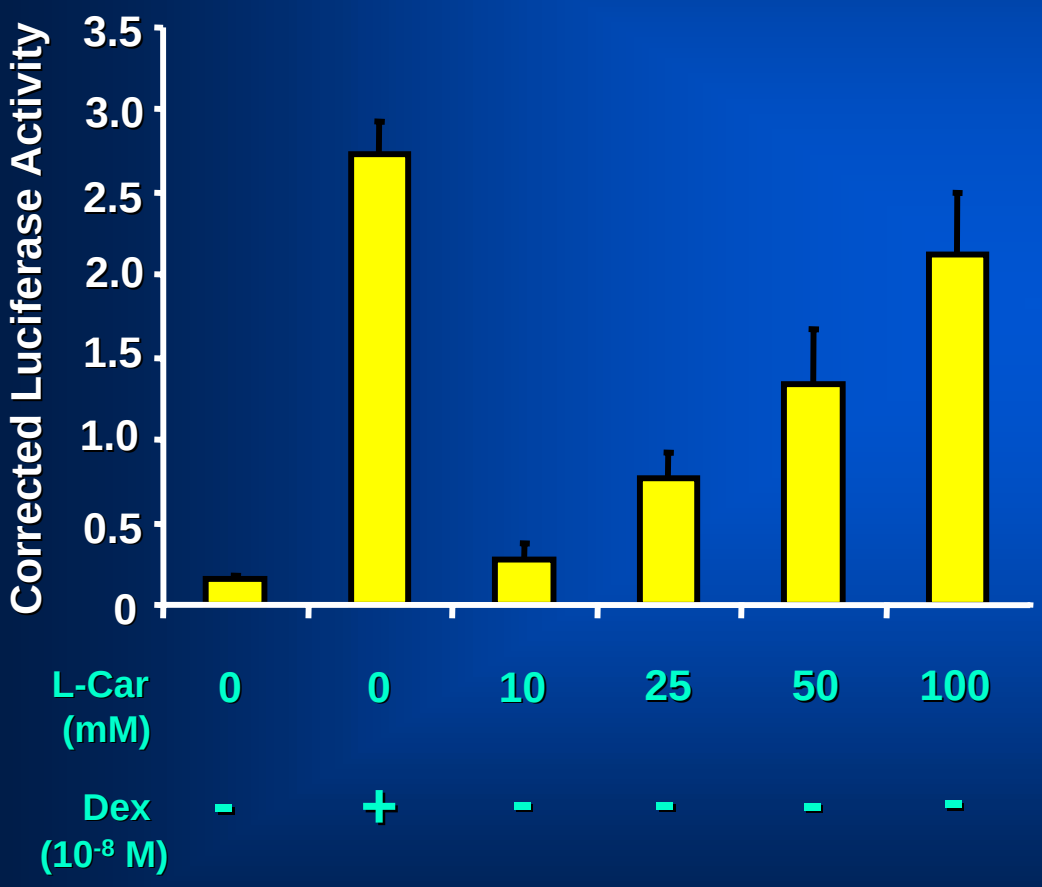
Reporter Plasmids

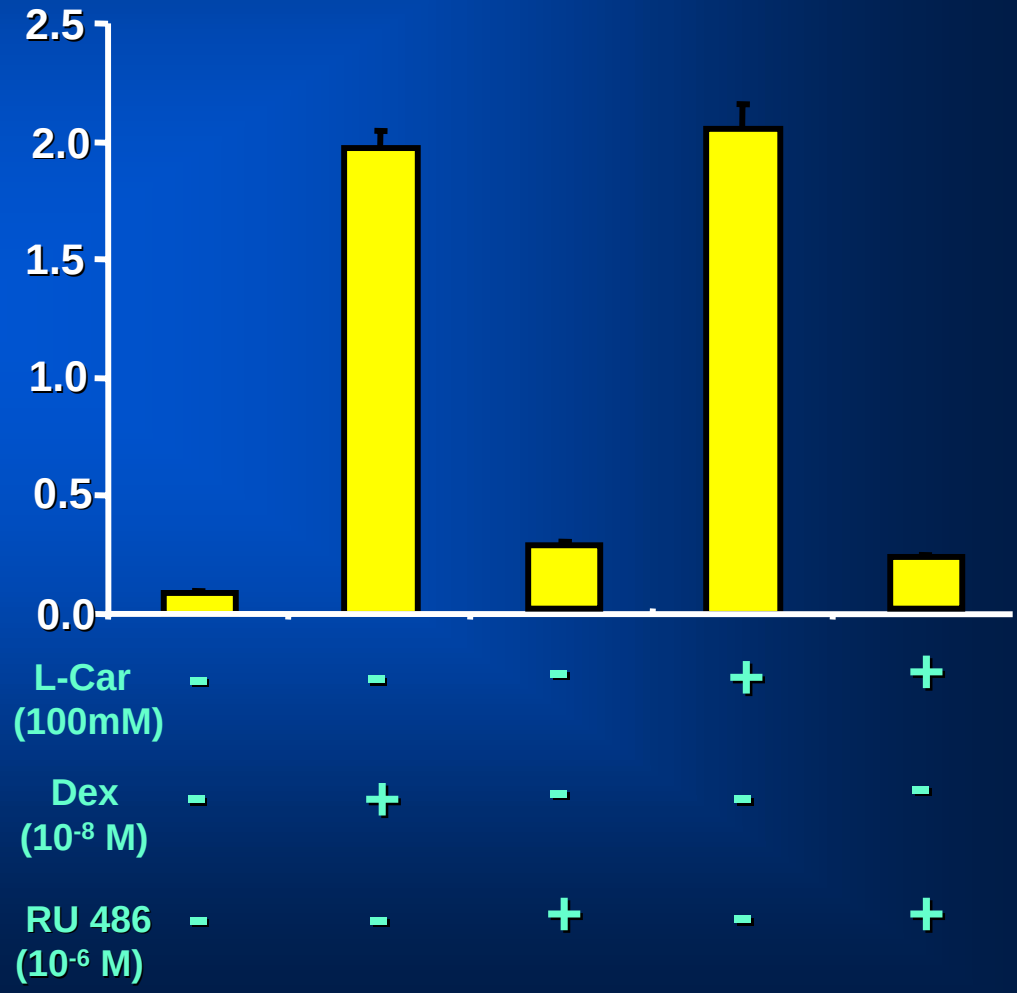
MMTV:LUC

SV40:β-Gal

Promoter transcription

HeLa: MMTV





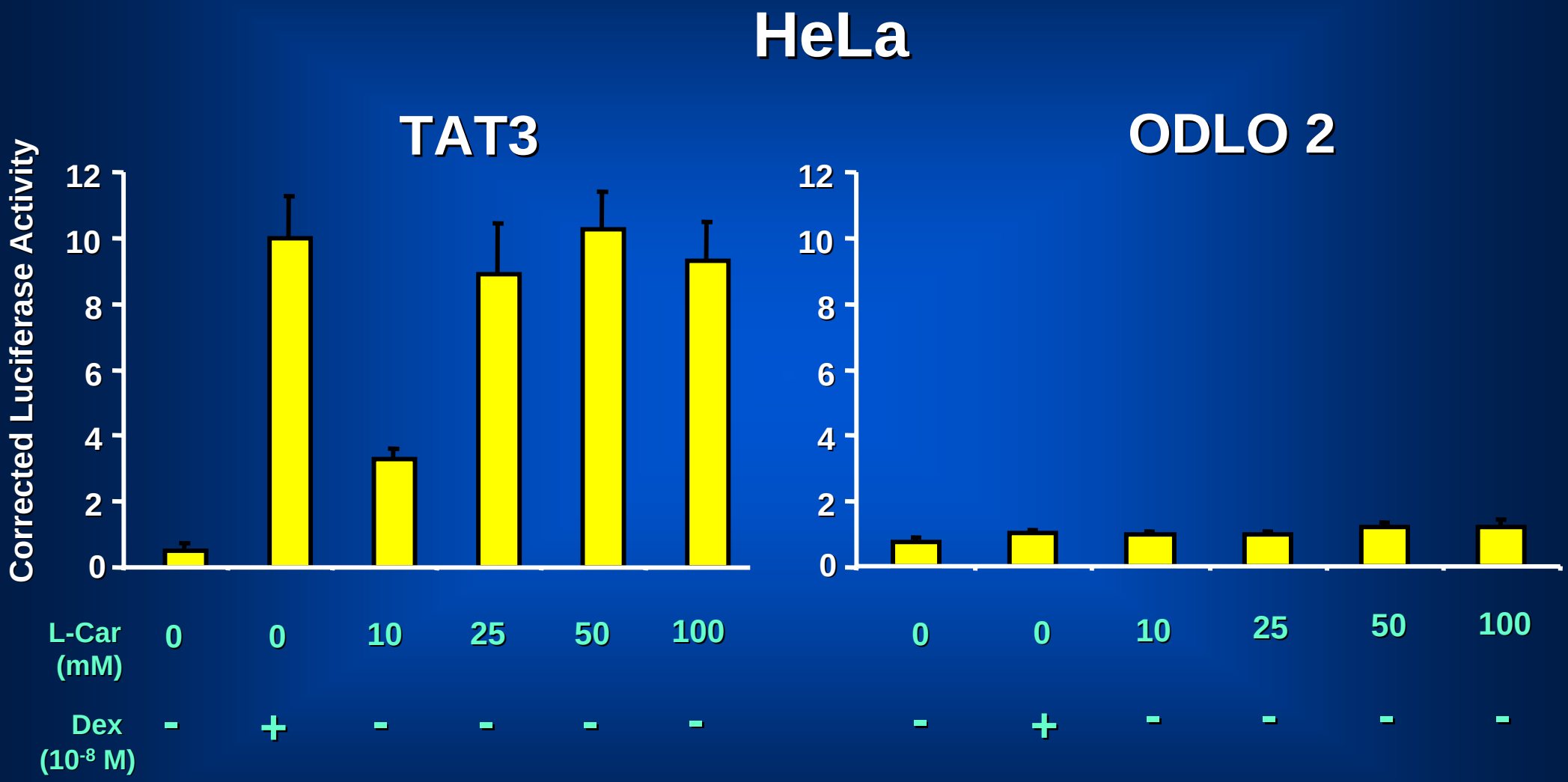
Reporter Plasmids

TAT3:LUC

pODLO2:LUC

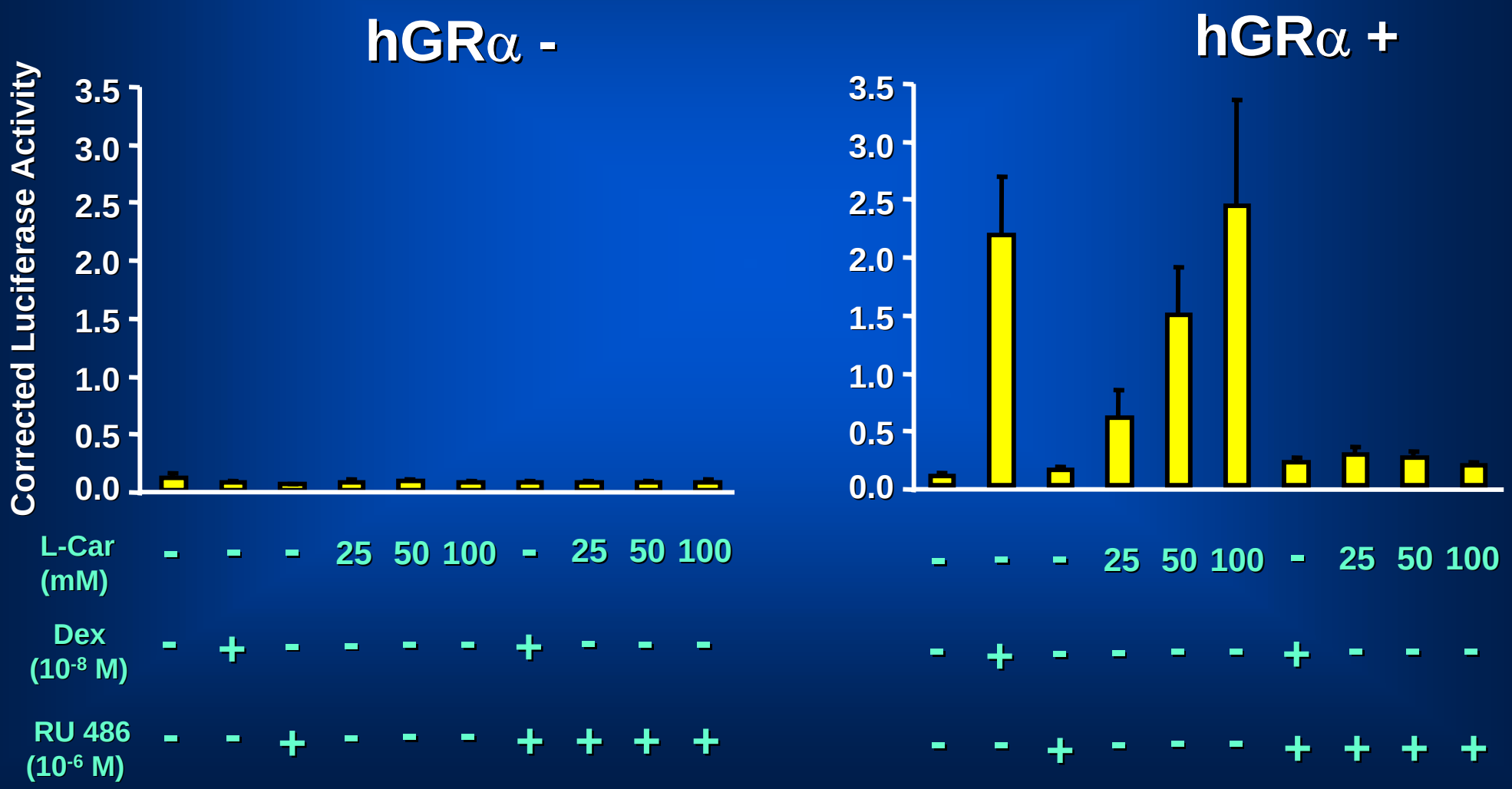
SV40:β-Gal

Promoter transcription



Promoter transcription

CV-1: MMTV

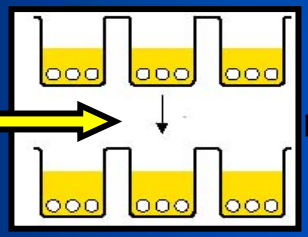


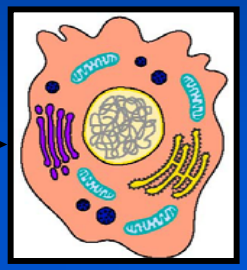
Cytotoxicity assay

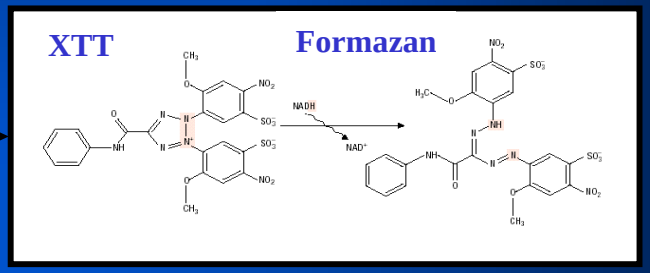
24 h

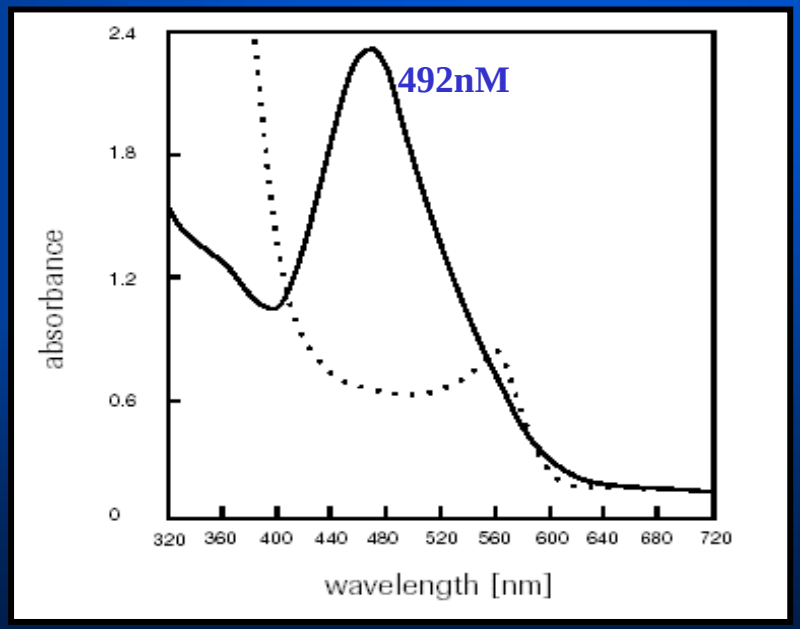
XTT

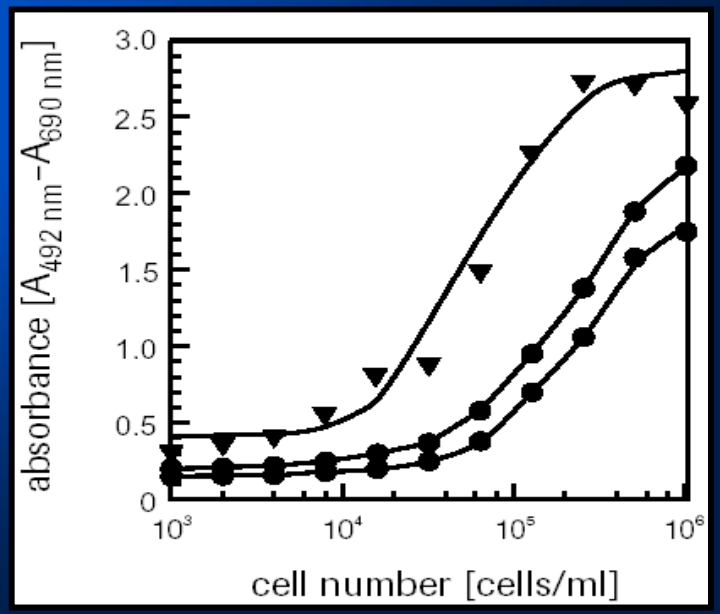
18 h



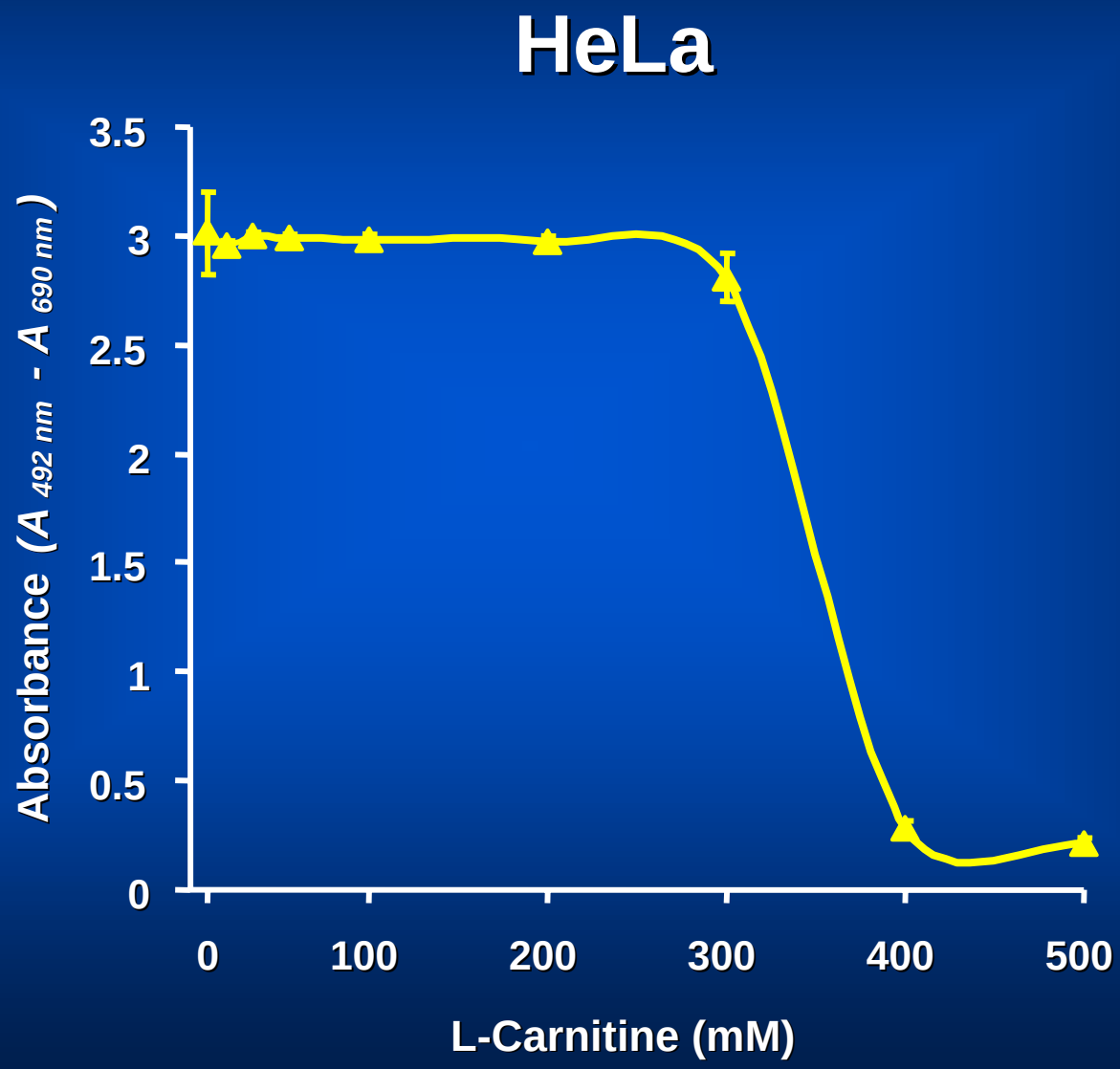






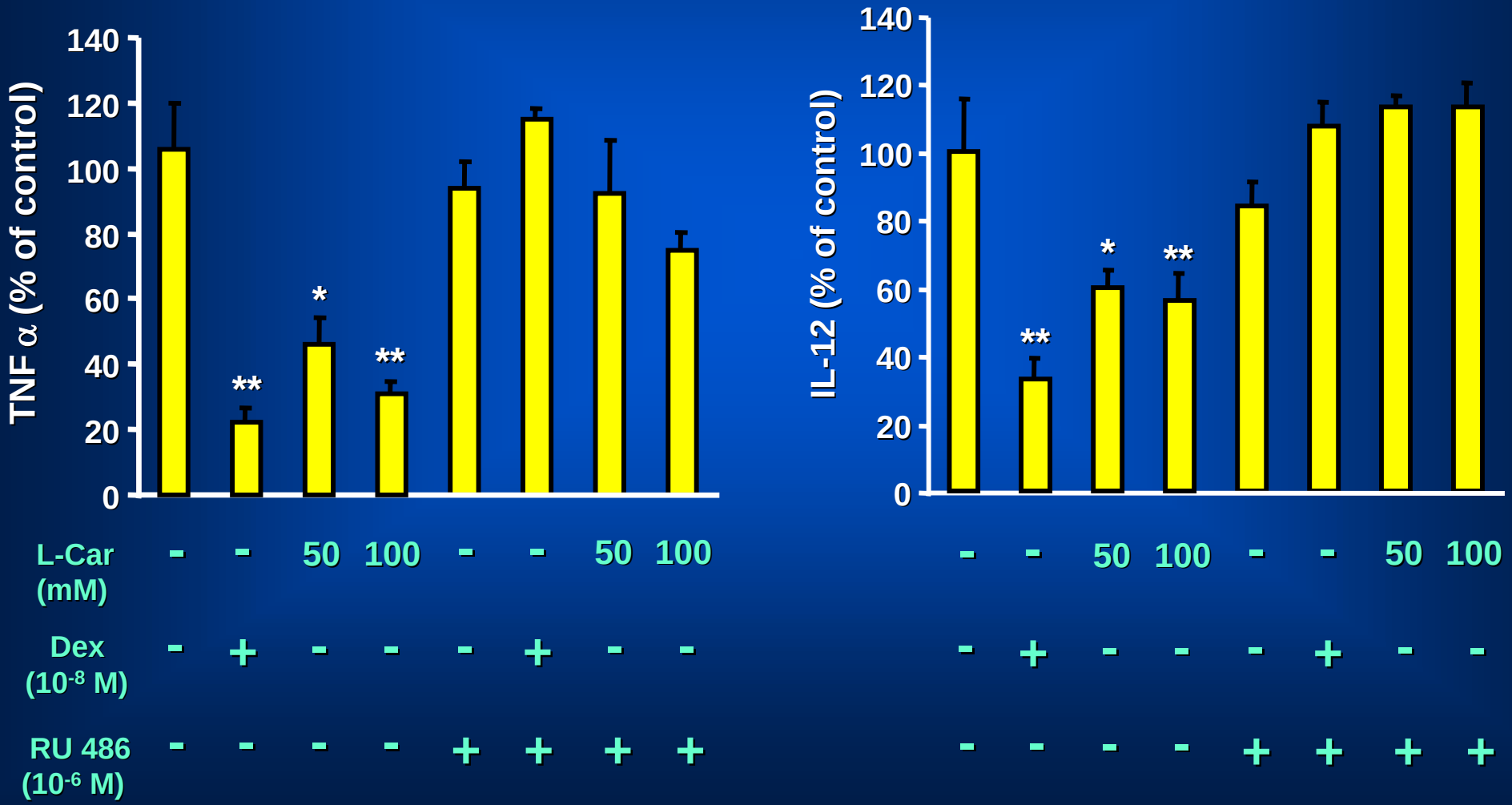


Cytotoxicity assay



LPS-stimulated cytokine secretion

Human elutriated monocytes

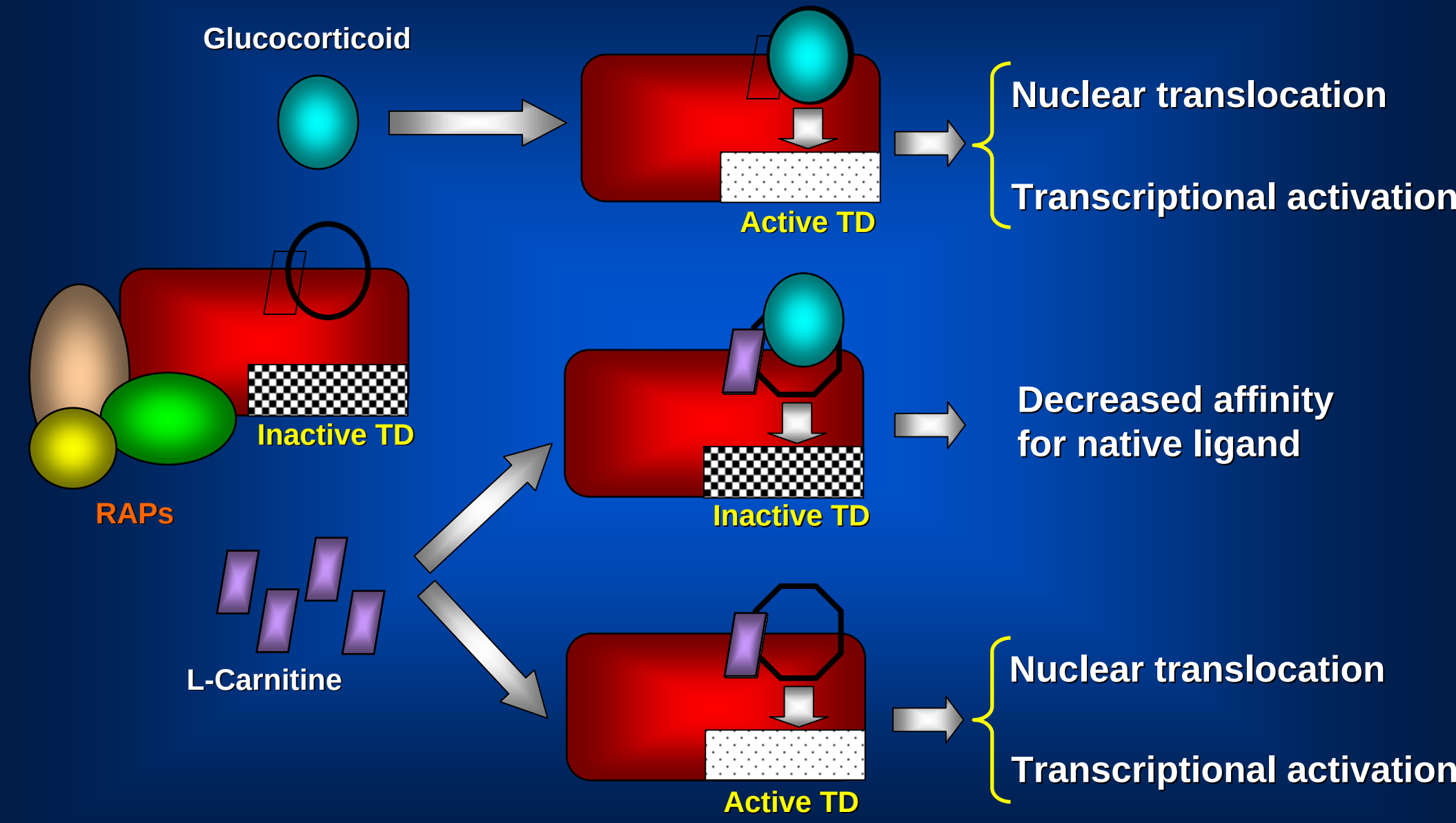


Summary

***In vitro*, high but non-cytotoxic concentrations of L-Carnitine are able to:**

- ***Compete with dexamethasone for binding to the GRα, reducing the affinity of this receptor for its native ligand***
- ***Trigger nuclear translocation of GRα***
- ***Stimulate the transcription of glucocorticoid-responsive promoters through GRα transactivation***
- ***Suppress the release of TNFα and IL-12 from human elutriated monocytes in a GRα-dependent fashion***

Proposed model of GR modulation by L-Carnitine



Future Research

- *Molecular and structural mechanisms of L-Carnitine → GRα interaction*
- *Tissue specificity/selectivity of GRα modulation by L-Carnitine (muscle, bone, adipose tissue, neurons)*
- *Effect of L-Carnitine metabolites (Acetyl-L-Carnitine, Propionyl-L-Carnitine, etc.) on GRα function*
- *Effect of L-Carnitine and its metabolites on the activity of other steroid receptors (PR, MR, AR, etc.)*
- *Studies in animal models of chronic inflammatory and autoimmune disorders*
- *Clinical trials*
- *Drug designing*

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