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Guanylyl Cyclase C (GC-C) Inhibits Human Colon Carcinoma Cell Growth

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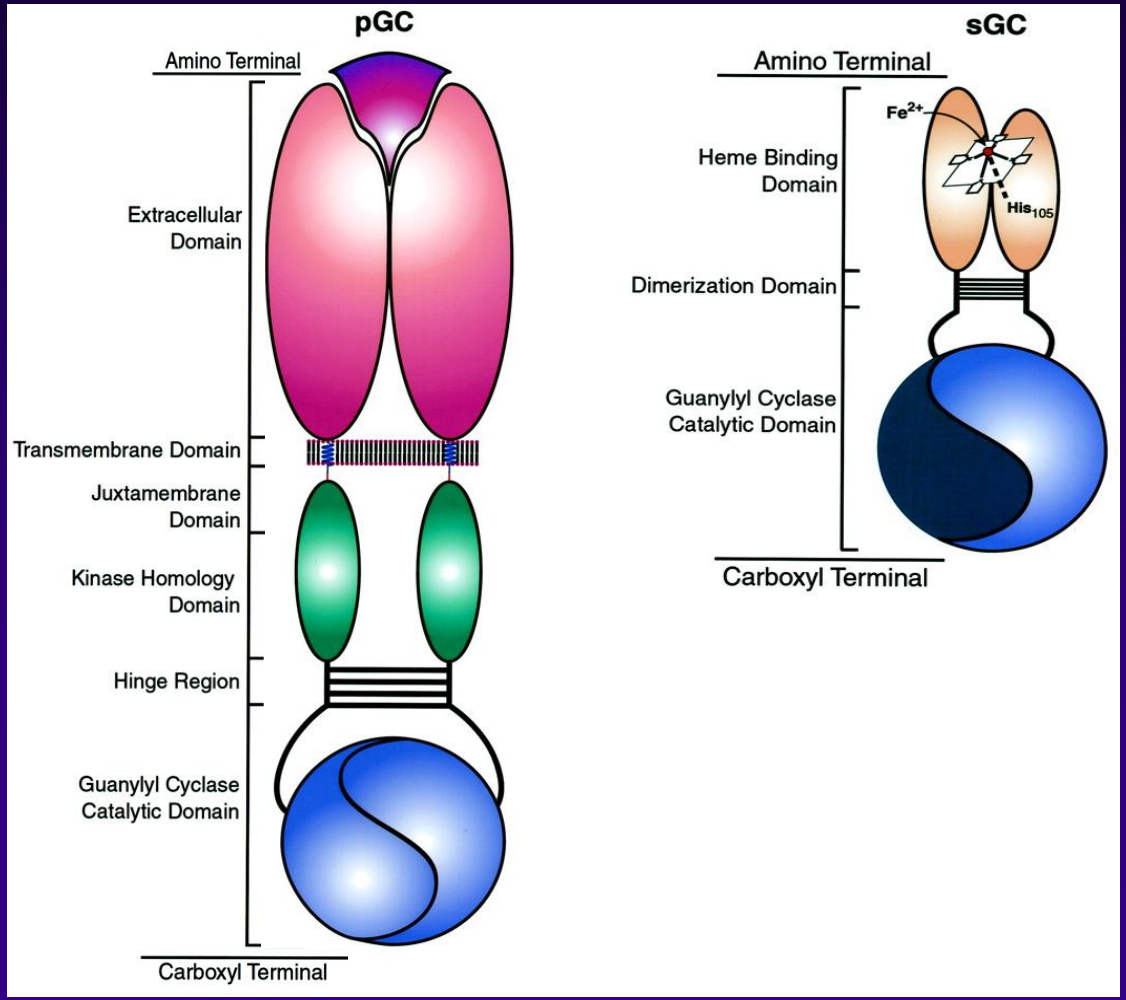
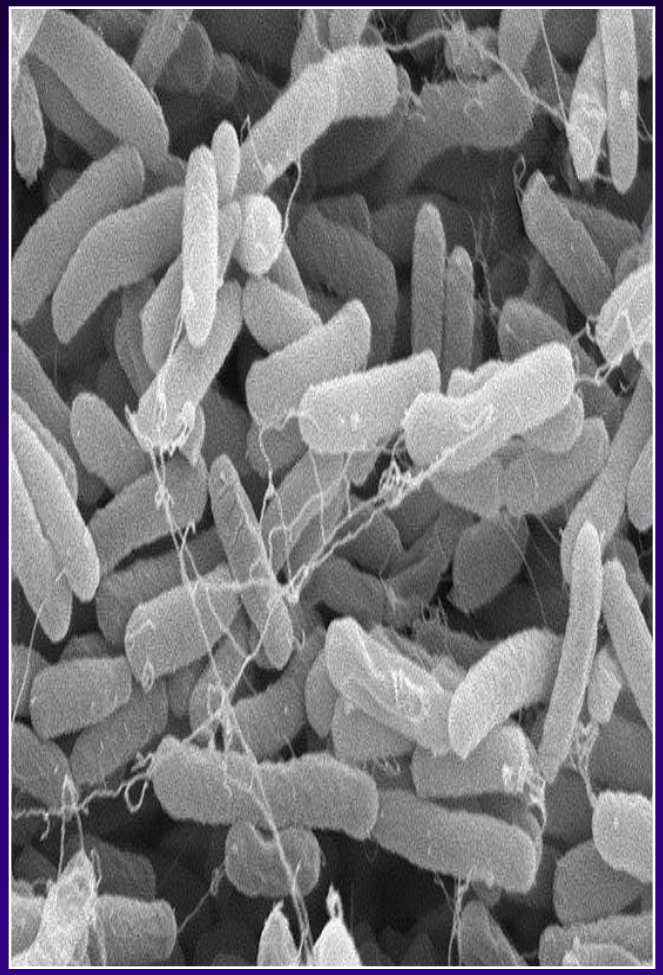
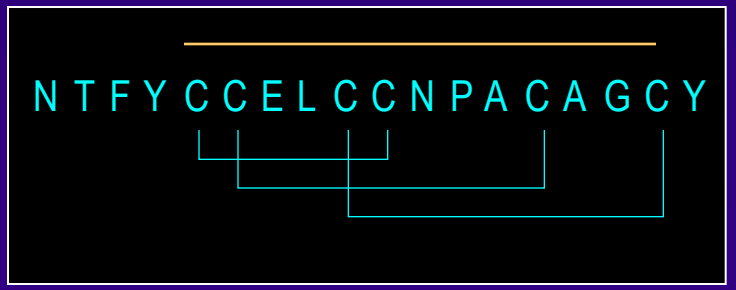
Guanylyl Cyclase Family



The E. coli Heat-Stable Enterotoxin (ST) Binds GC-C

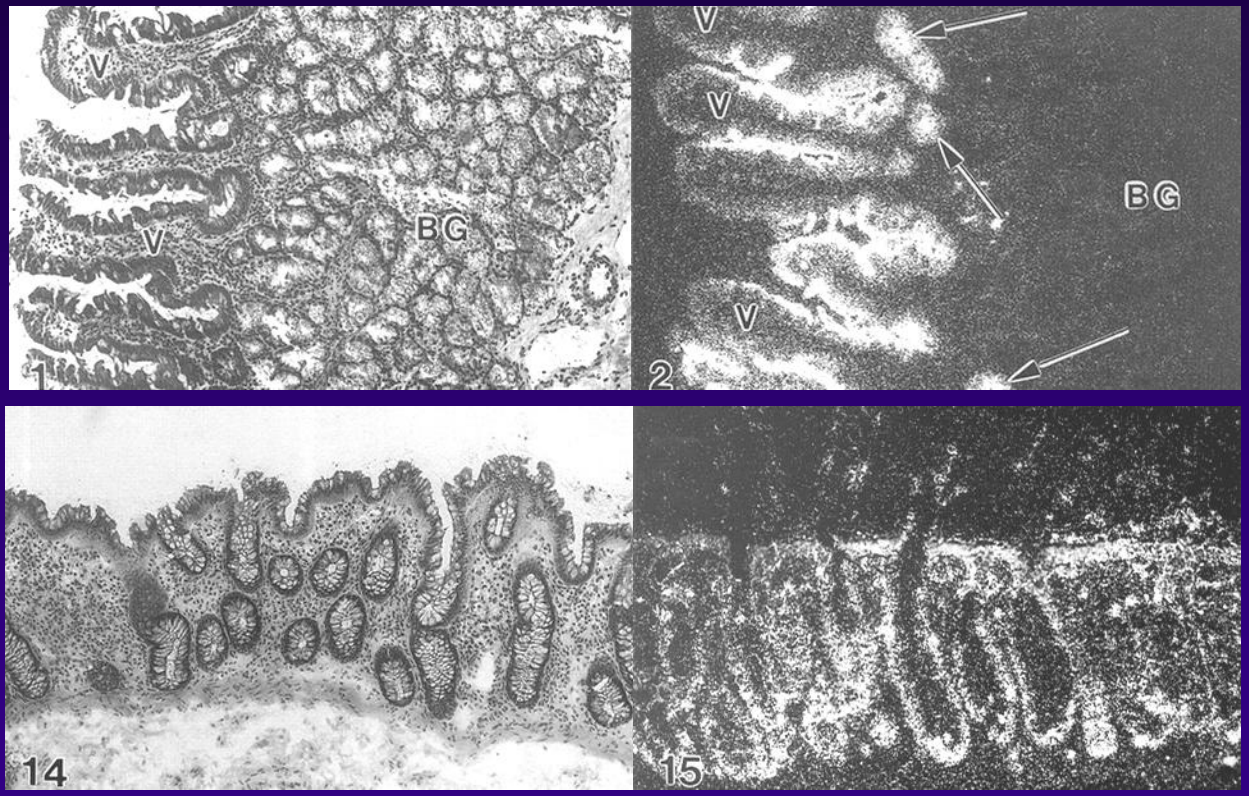




GC-C is Localized to Intestinal Epithelial Cells

H&E

^{125}I-ST Binding

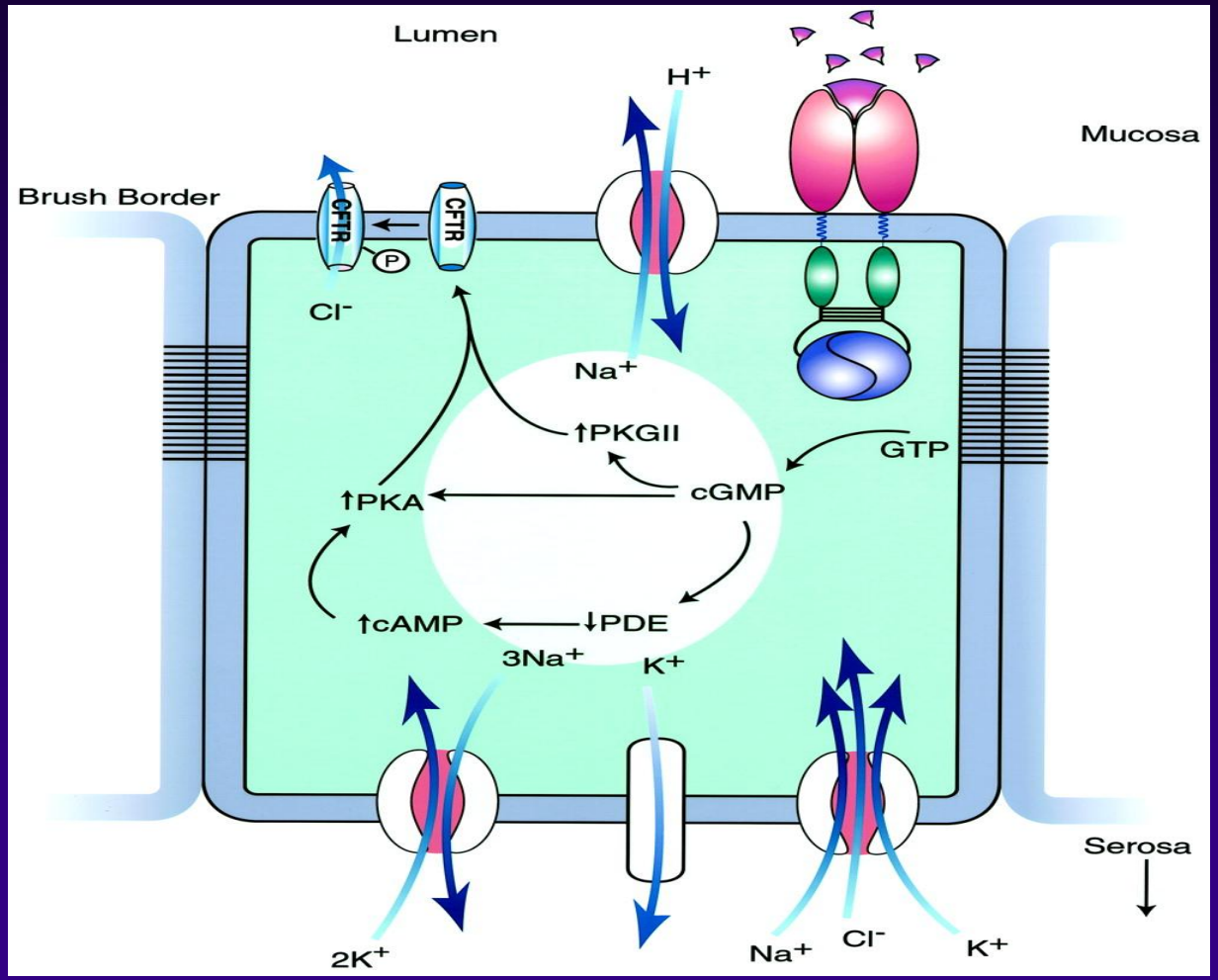


Small Intestine

Large Intestine

Krause et al (1994) J Anat 184:412

GC-C Signaling Cascade



Does GC-C Mediate More Than Fluid Transport in Intestine?

- *Does GC-C regulate intestinal epithelial cell proliferation?*
- *What are the molecular mechanisms by which GC-C regulates intestinal cell proliferation?*

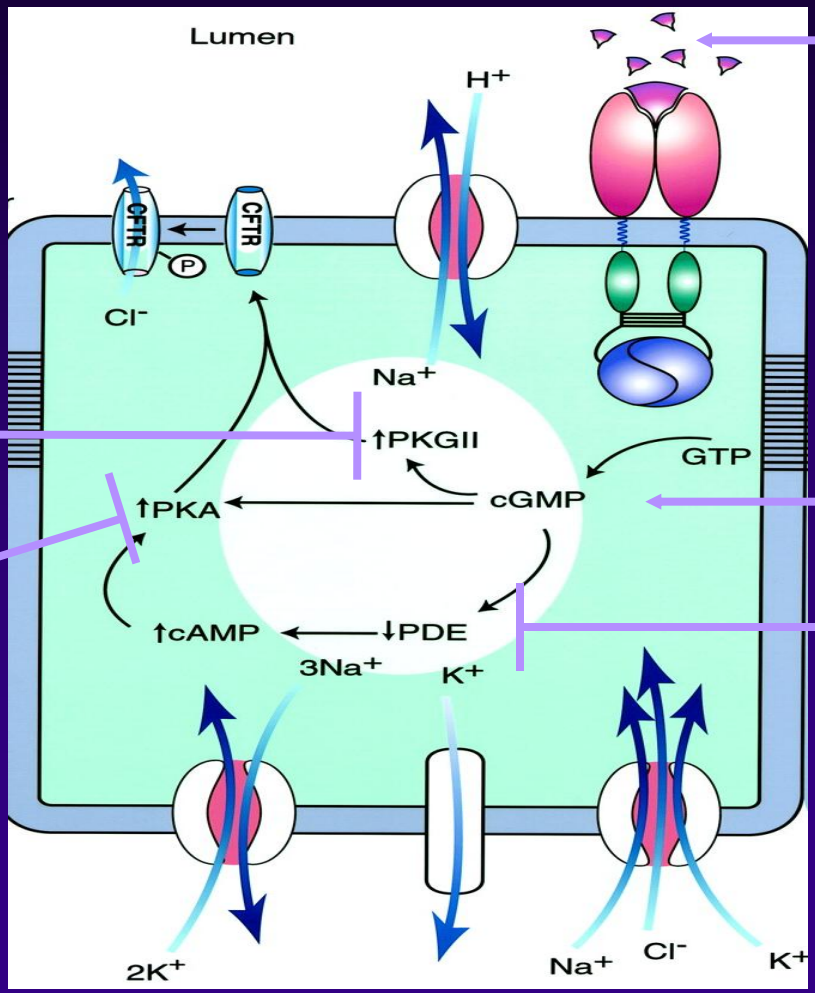
Protocol Design & Materials

Cell Lines:
T84, Caco-2, SW480

Pro-Proliferative Agents:
FBS, L-Glutamine

KT5823
RP8pCPT-cGMP

KT5720
Rp-cAMPs

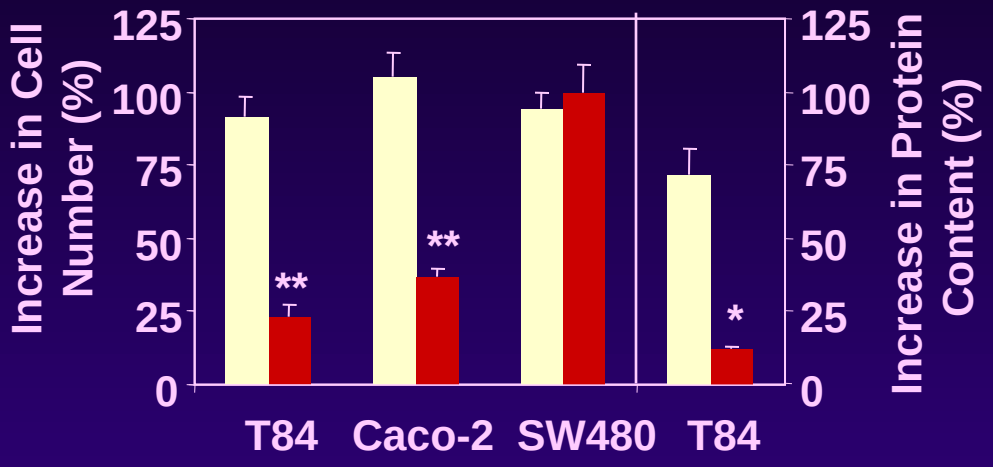


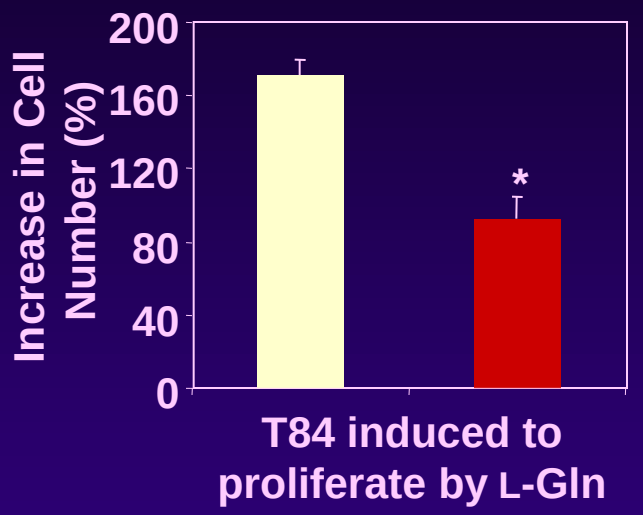
ST
Uroguanylin

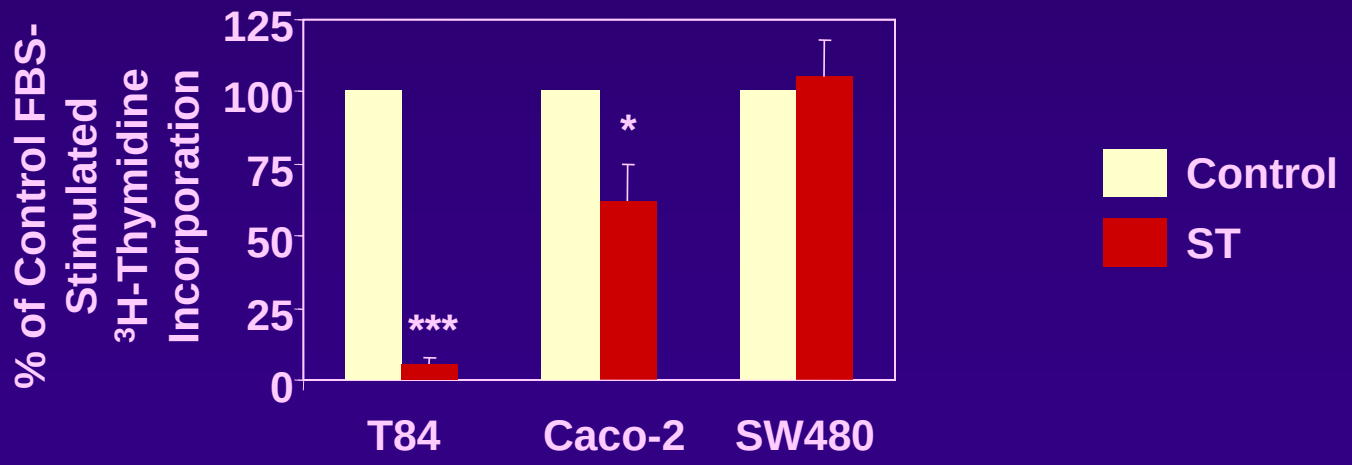
8-Br-cGMP

Milrinone

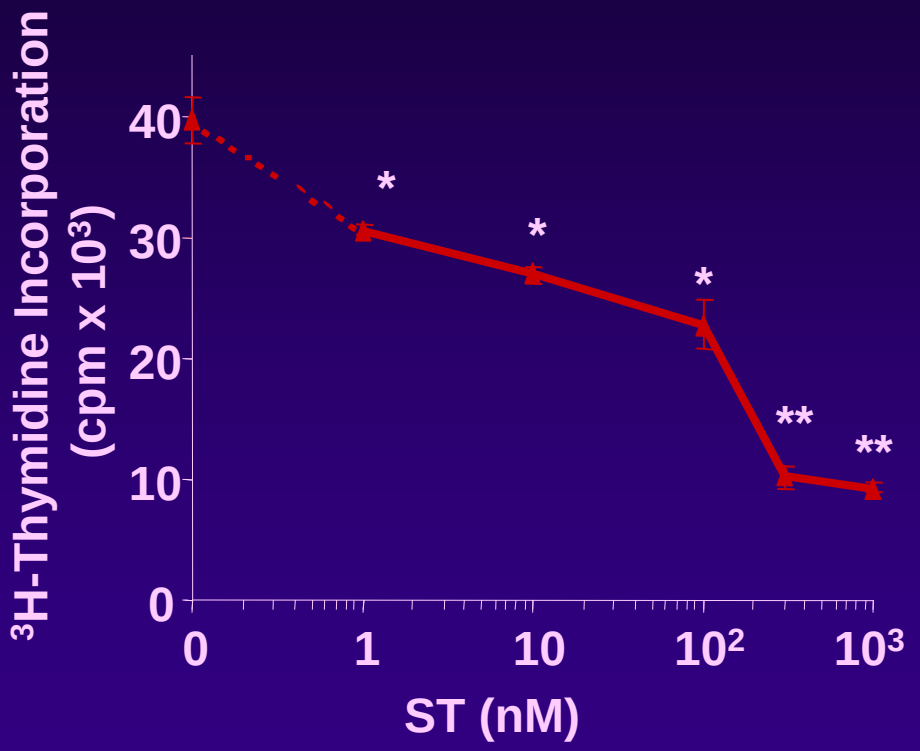
ST Inhibits Intestinal Cell Proliferation

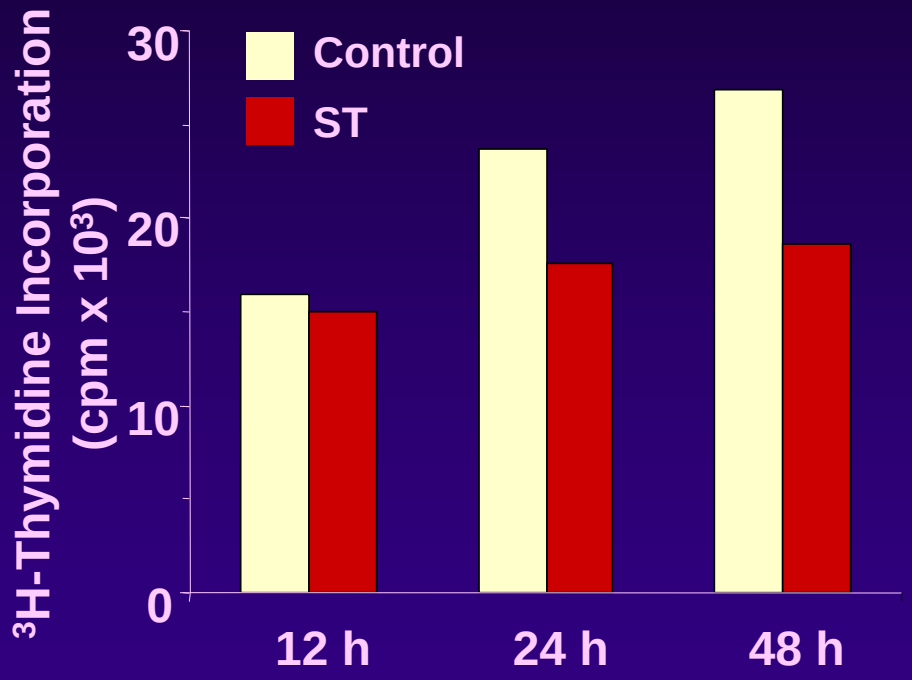




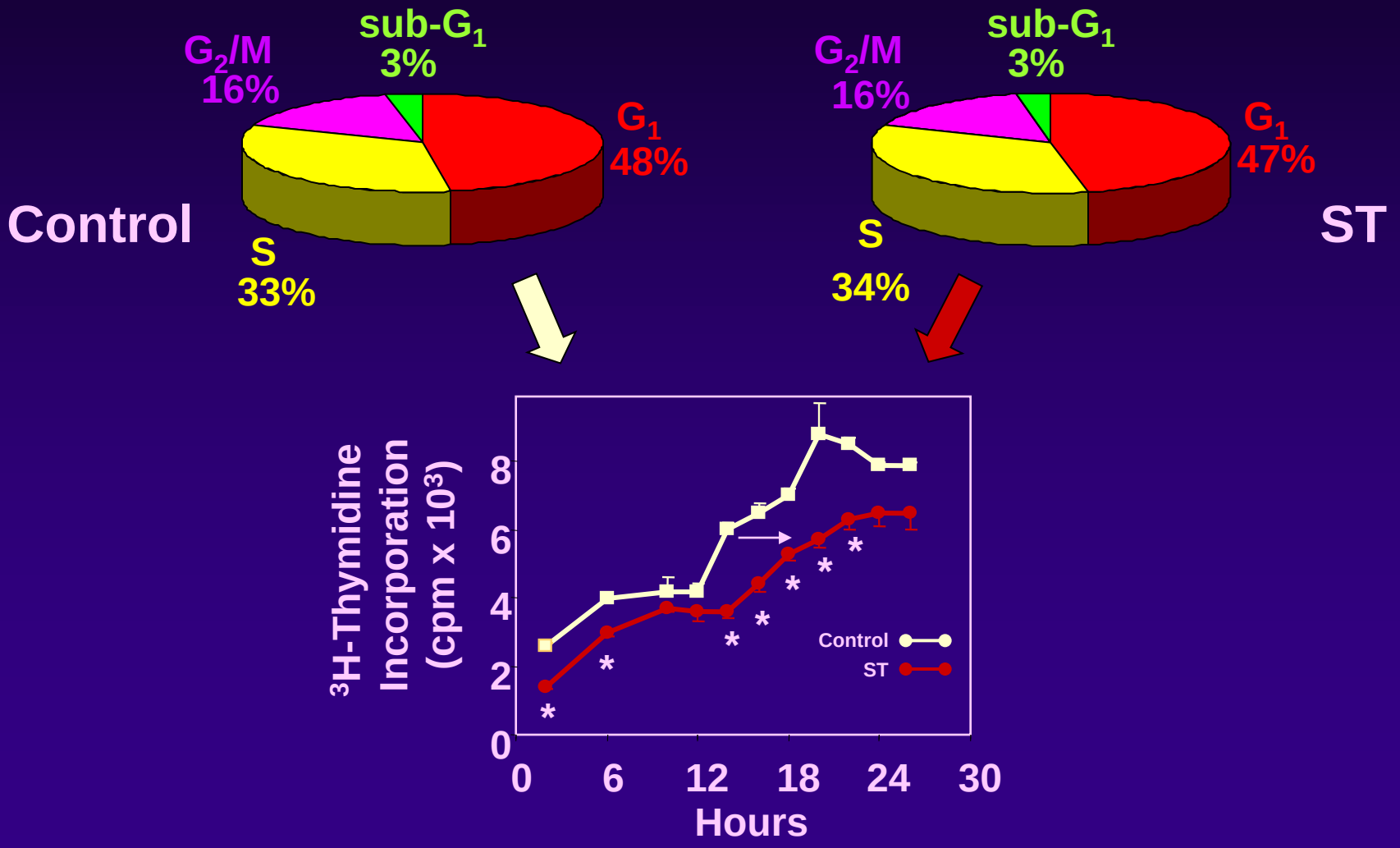


ST Inhibition is Dose- and Time-Dependent

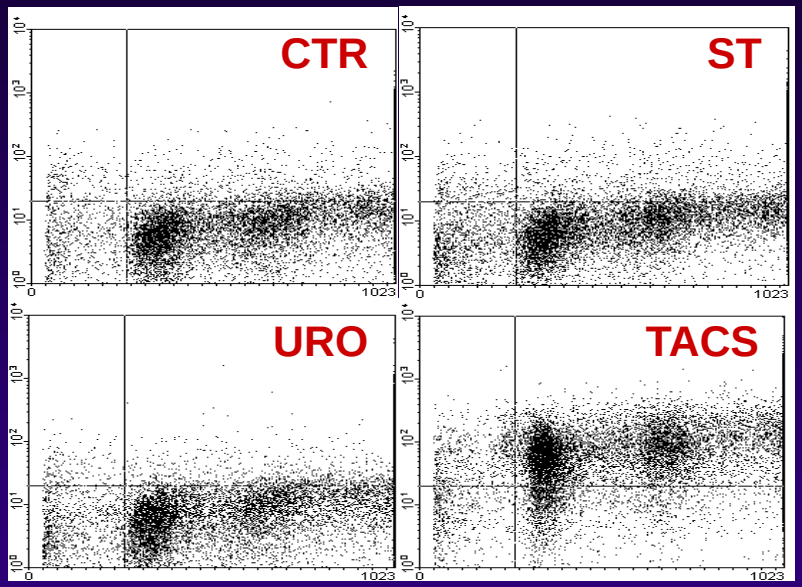


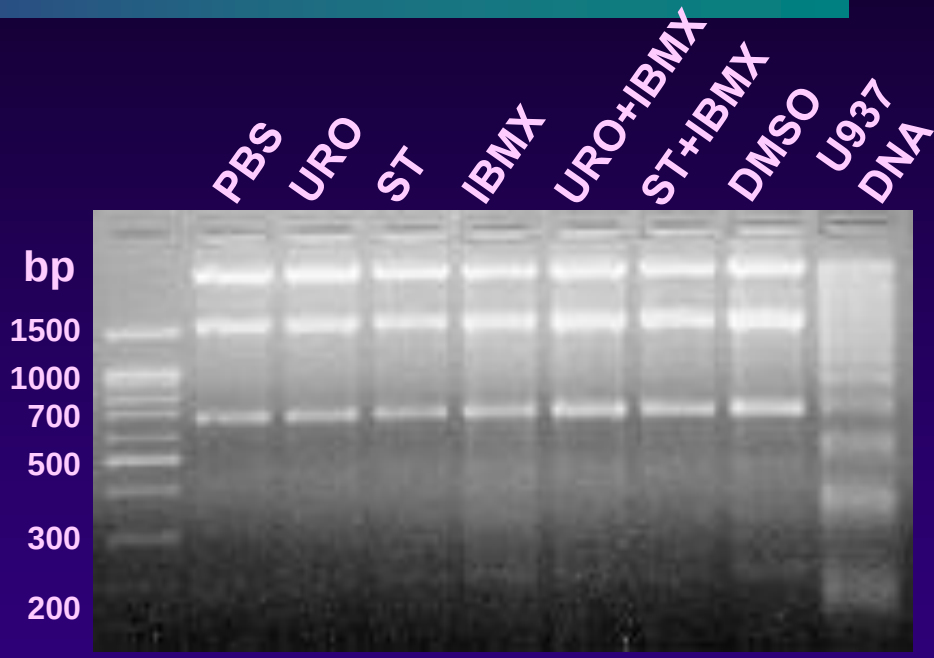


ST Delays, But Does Not Arrest, the Cell Cycle



GC-C Agonists Do Not Induce Apoptosis or Necrosis

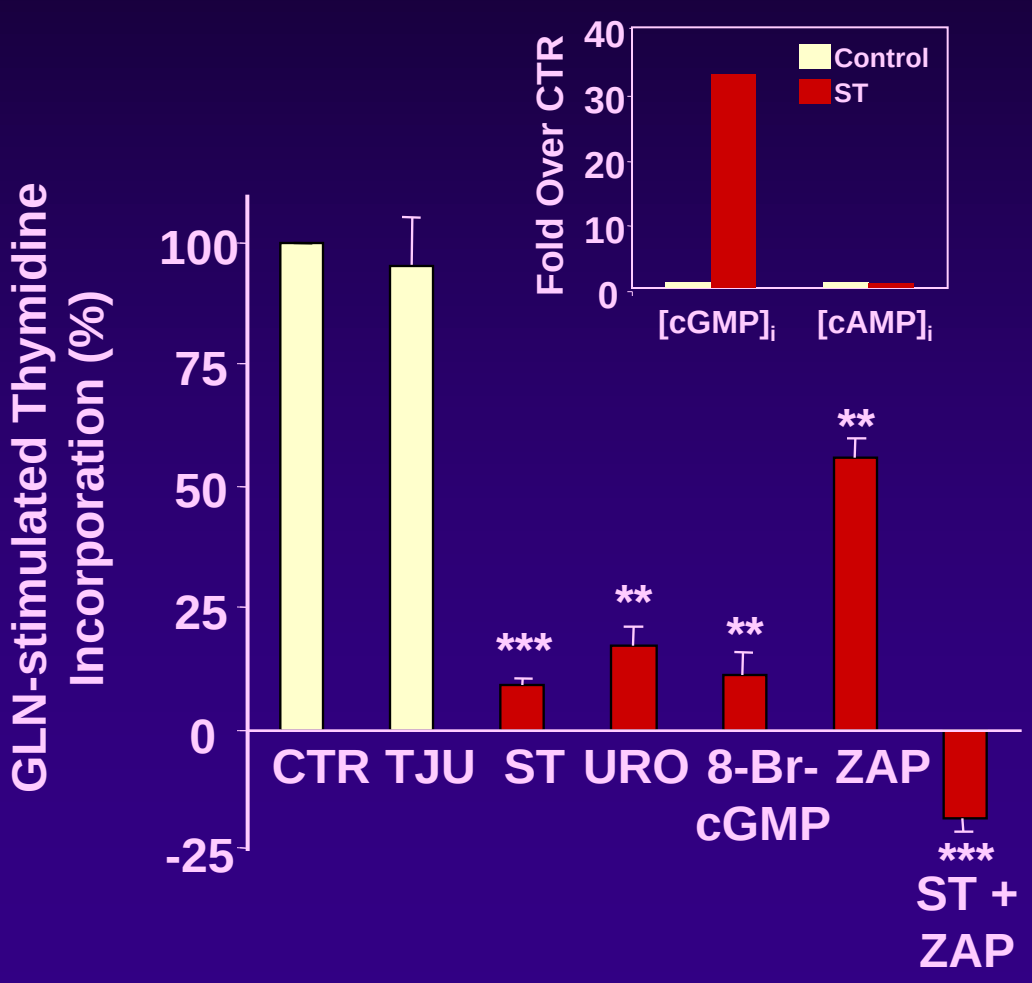


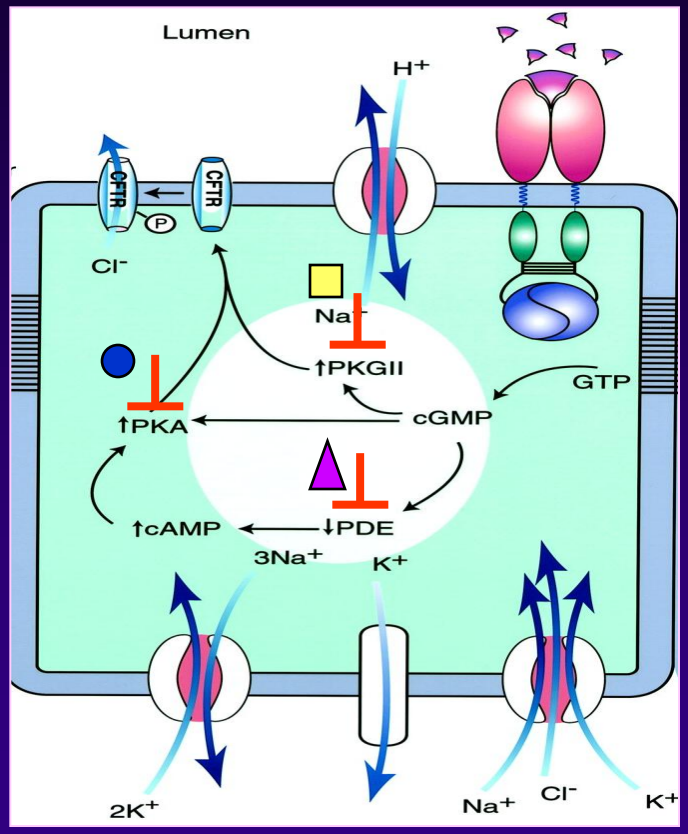


	Control	ST (1 μM)	Uro (1 μM)	TACS
% Apoptosis	7.4 ± 0.5	9.1 ± 1.2	6.9 ± 0.9	75.3 ± 2.1**

** $p<0.01$

ST Cell Signaling Pathway for the Inhibition of Proliferation



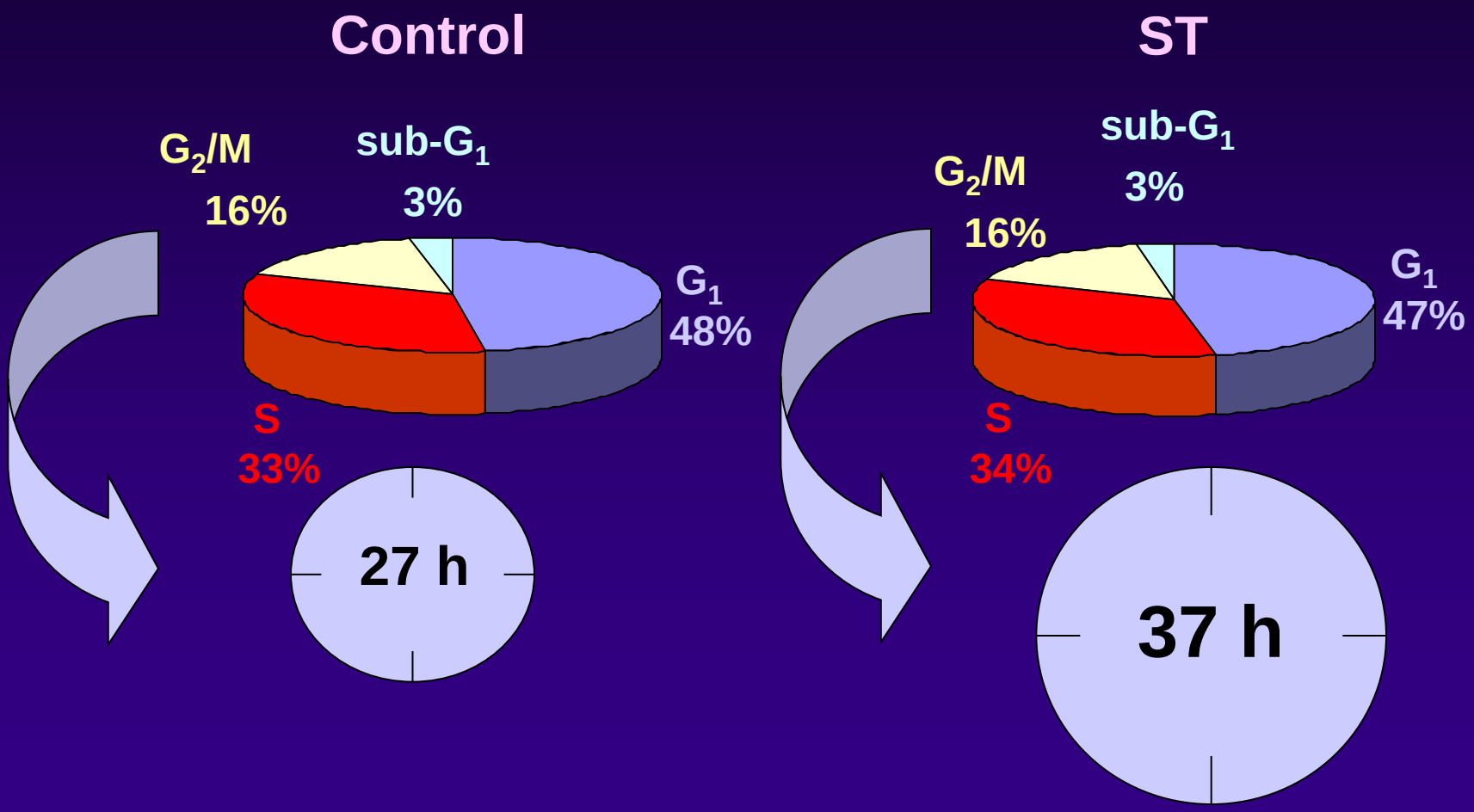


Milrinone
KT5823, RP8pCPT-cGMP
KT5720, Rp-cAMPs

Summary

- *GC-C activation inhibits colon carcinoma cell proliferation* in vitro
- *Inhibition of proliferation results from a prolongation of the cell cycle, not cell death*
- *The cytostatic effect of ST is mediated by an increase in* $[cGMP]_i$

ST-Dependent Cytostasis Does Not Reflect Arrest, but Retardation, of the Cell Cycle



Implications of GC-C Regulation of Proliferation

- *Endogenous GC-C ligands (guanylin and uroguanylin) may represent cell cycle regulators*
- *Along the crypt-to-villus axis, GC-C may regulate the transition of intestinal epithelial cells from proliferative to differentiated states*
- *GC-C agonists may be utilized as novel cytostatic agents for the prevention and treatment of colorectal cancer*

Acknowledgements

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