

Targeting The Stress-Induced Cytoprotective Chaperone, Clusterin, to Overcome Treatment Resistance in Advanced Prostate Cancer



THE PROSTATE CENTRE
AT VANCOUVER GENERAL HOSPITAL

Martin Gleave MD, FRCSC, FACS

***Professor, Department of Urologic Sciences
University of British Columbia
B.C. Leadership Chair in Prostate Research
Director, The Prostate Centre at VGH***

Disclosure of Conflicts of Interest

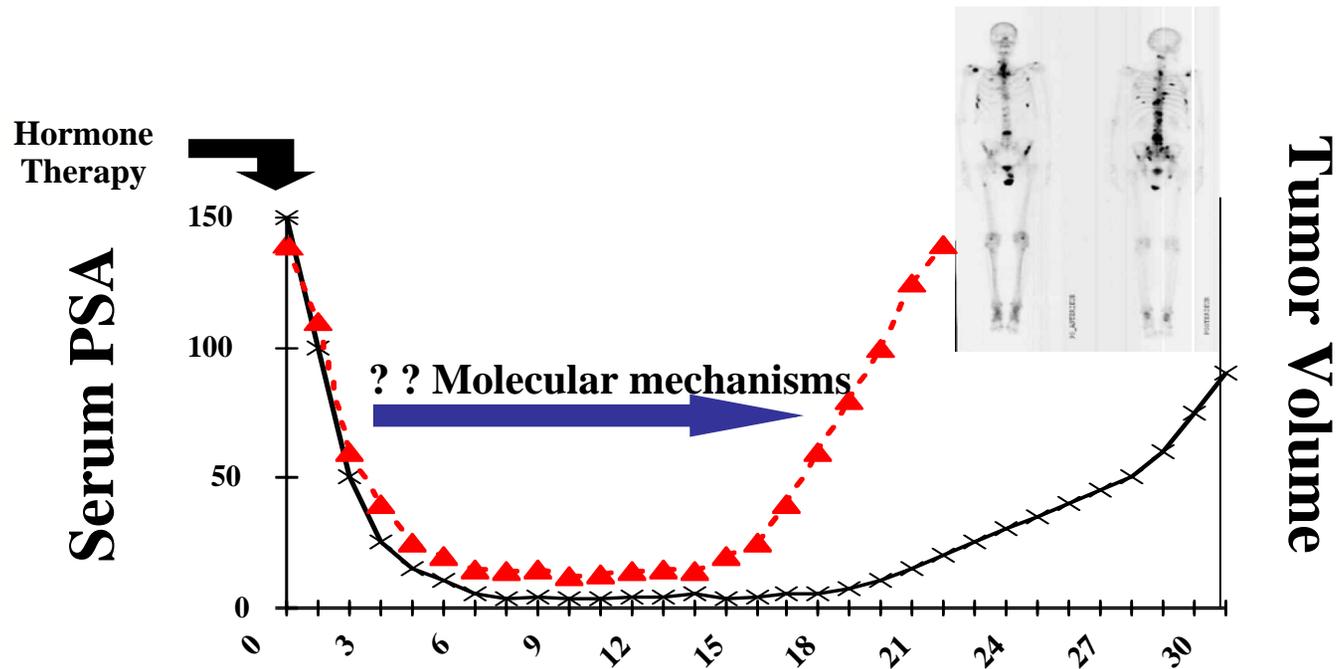
Patent - OGX-011

Founder - OncoGenex Technologies

Consultant - CSO, OncoGenex Technologies

Castration Resistance and Prostate Cancer

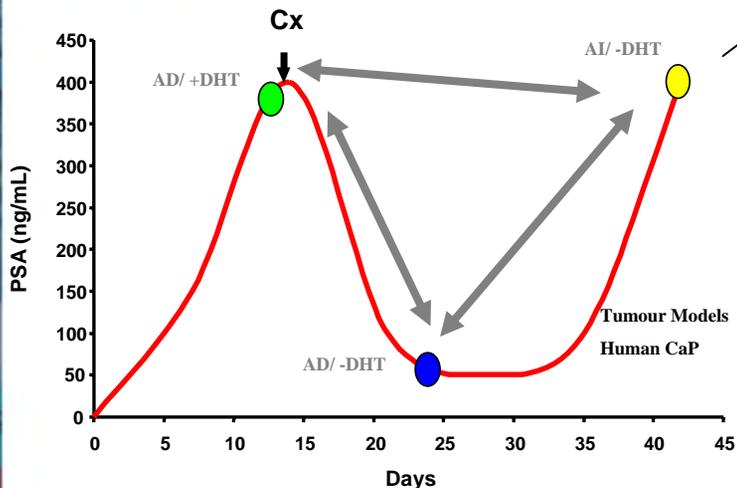
1. Androgen receptor (AR) related
 - Overamplification (hypersensitive)
 - Mutations (promiscuous)
 - cross-talk - TK, PKA, AKT, STAT3 (phosphorylation, co-regulators)
2. Adaptation
 - Up-regulation of survival genes (Bcl-2, clusterin, Hsp27, YB-1)
 - Increased alternative GF pathways (her2/neu; IGF-1/IGFBP2&5; IL-6/STAT3)



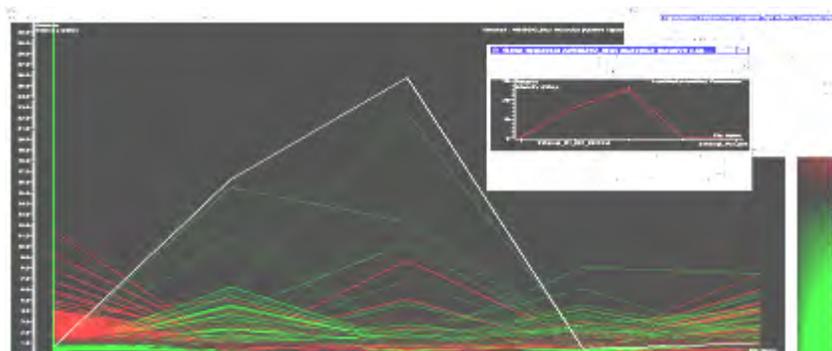
High-Throughput Bioprofiling of Hormone-Treated Prostate Cancers to Identify Stress-Induced Targets

Hypothesis:

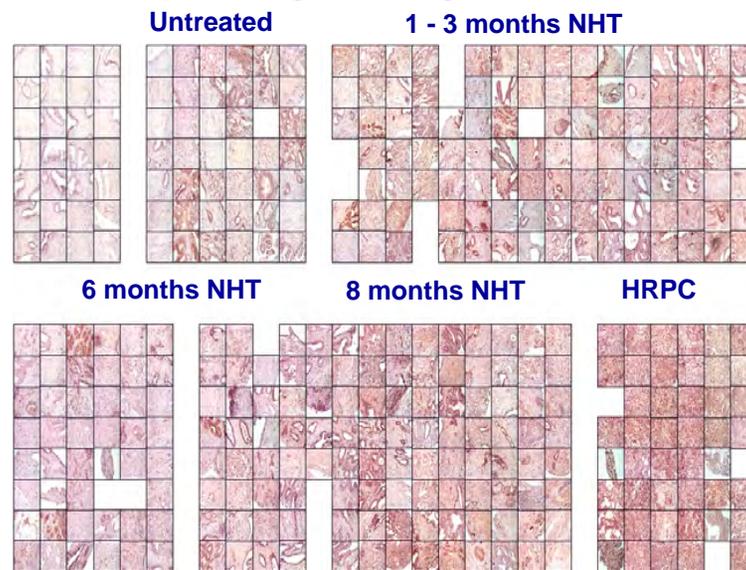
➤ Treatment-induced changes in gene expression after hormone therapy render cells resistant to castration- and chemotherapy.



Gene expression profiling for Target I.D.



TMA profiling for Target Validation

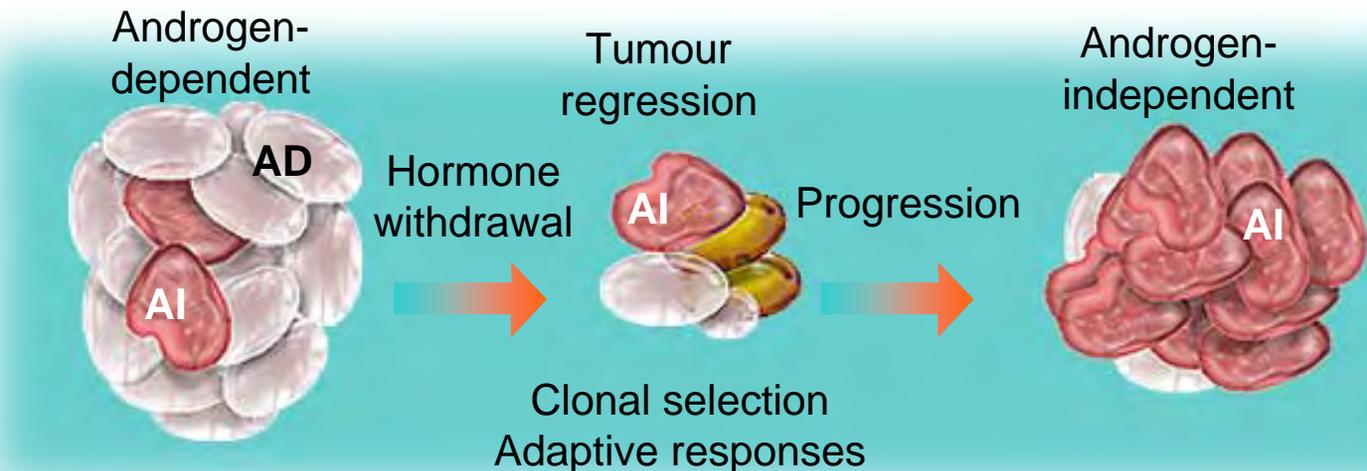


Post-Hormone-Treated TMA

Tissue Bank:

- Xenografts
- Post-NHT-treated human CaP
- pre-surgery NHT + Taxotere RP specimens
- rapid autopsy HRPC specimens

Changes in Gene Expression After Castration and During AI Progression



- ++ PSA
- Bcl-2
- EGFR
- clusterin
- IGFBP 2&5
- TGFβ
- ++IGFBP 3 & 4
- YB-1
- ++survivin
- Hsp27
- + PKC-α

- ↓
- PSA
 - ++ Bcl-2
 - ++Bclx-L
 - EGFR
 - +++ clusterin
 - ++++IGFBP 5
 - IGFBP 3 & 4
 - +c-myc
 - +YB-1
 - survivin
 - +Hsp27
 - + PKC-α

- ++ PSA
- ++ **Bcl-2**
- ++Bclx-L
- + EGFR
- +++ **clusterin**
- ++ **IGFBP 2**
- ++ **IGFBP 5**
- ++**YB-1**
- ++ survivin
- +++**Hsp27**

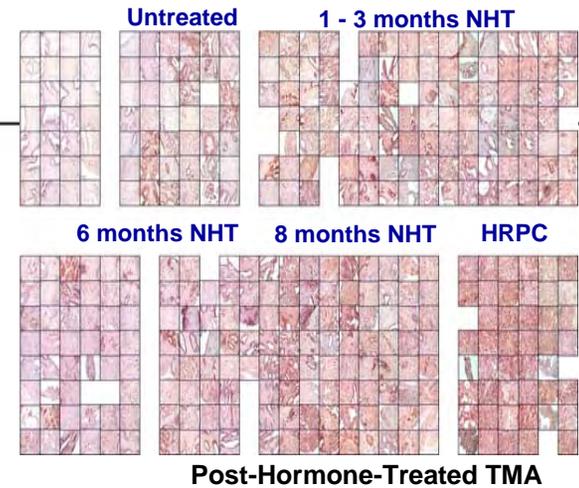


Therapeutic Stress Increases Clusterin Levels in CaP Tumor Models and Human Tissues

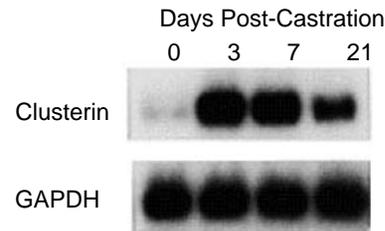
Transcriptionally activated by

- Hormone withdrawal (a.k.a. *TRPM-2*)
- Chemotherapies
- Radiotherapy
- Targeted therapies (Herceptin, Velcade, OGX-225, etc)

TMA profiling for Target Validation



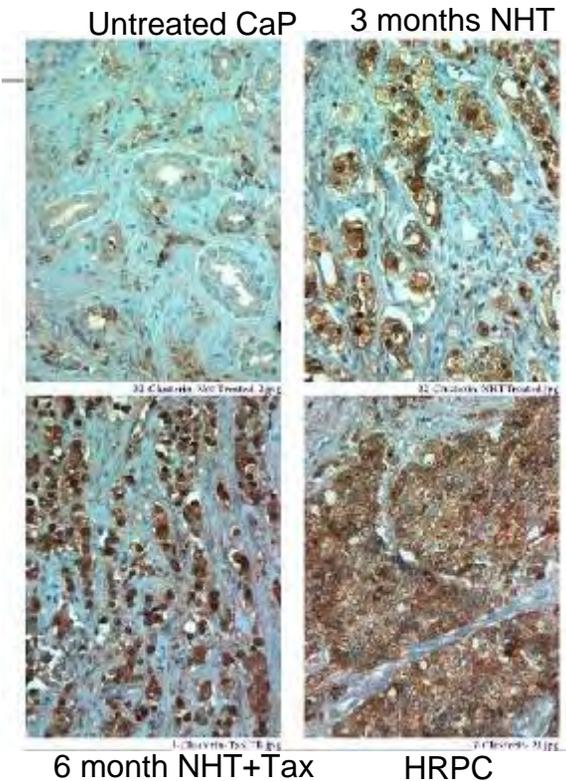
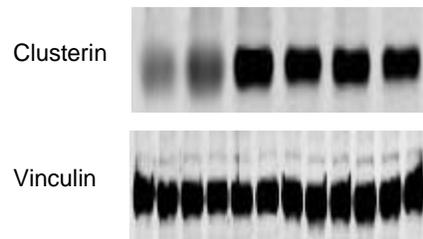
Androgen Ablation in Shionogi Tumors



Cancer Research 60; 170, 2000

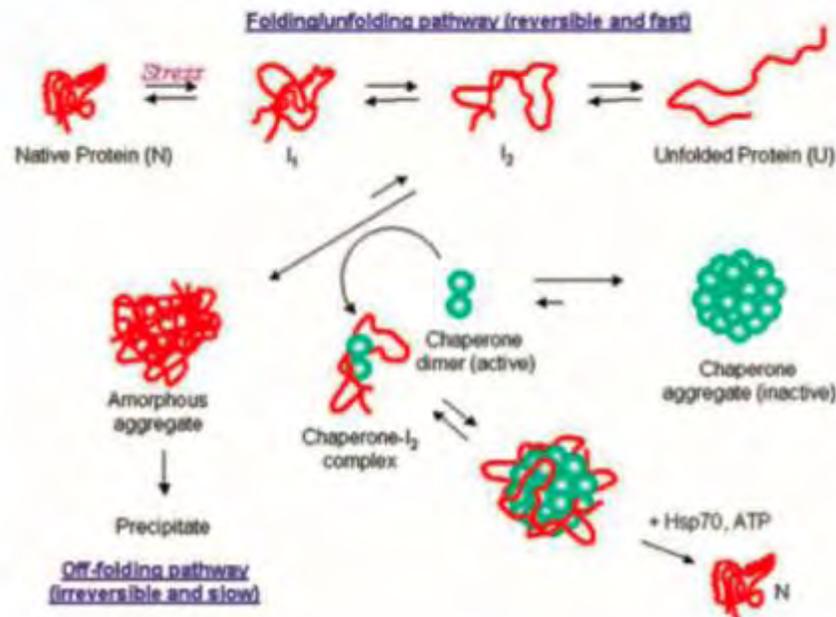
Docetaxel Rx in PC-3

Docetaxel Rx (nM) 0 10 25 50 100 500



SCLU-2: Stress-induced Cytoprotective Chaperone

1. Transcriptionally activated by HSF-1, repressed by p53
 - a. Increased by diverse array of therapeutic triggers (HT, CT, RT, velcade, herceptin)
 - b. Increased by cell survival factors like androgen, IGF-1
2. Intrinsically disordered and flexible protein
3. Potent inhibitors of aggregation of client proteins under stress conditions
4. Often associated with neurodegenerative diseases and cancer
5. Interact with and inhibit activated Bax; enhances NF- κ B transcriptional activity



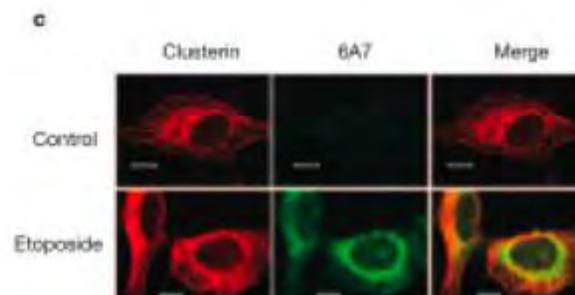
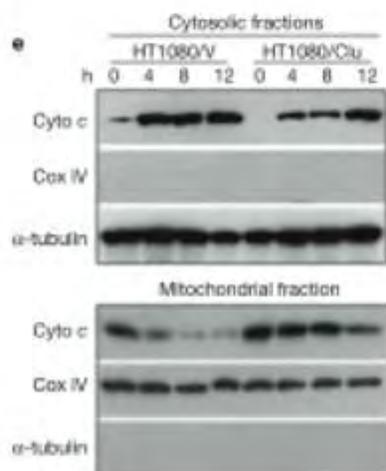
Clusterin: Cytoprotective Mechanisms

LETTERS

nature
cell biology

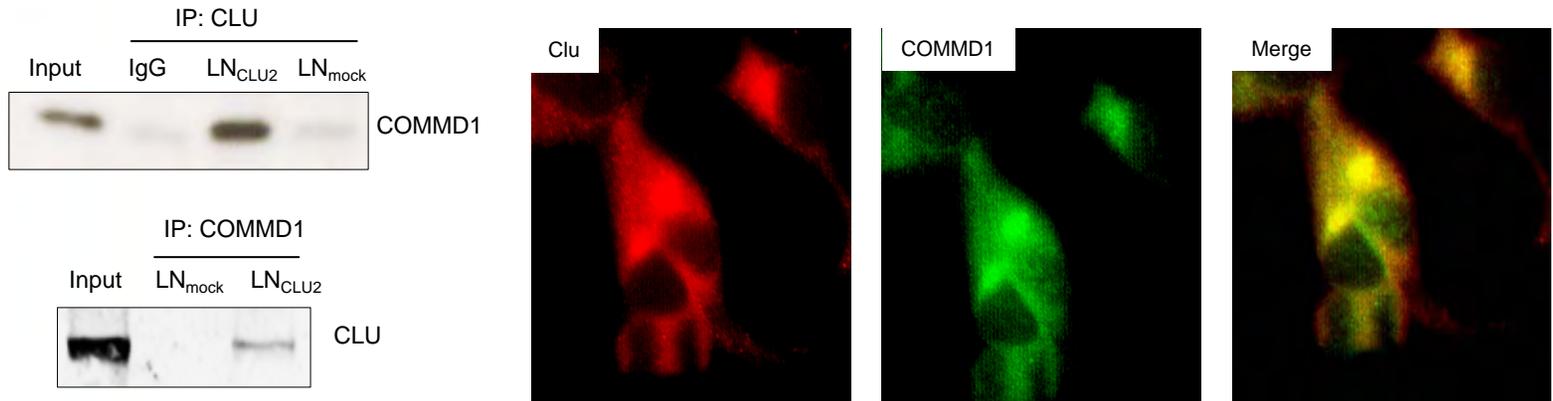
Clusterin inhibits apoptosis by interacting with activated Bax

Honglai Zhang¹, Jin Koo Kim¹, Chris A. Edwards², Zhaohui Xu³, Russell Taichman⁴ and Cun-Yu Wang^{1,5}



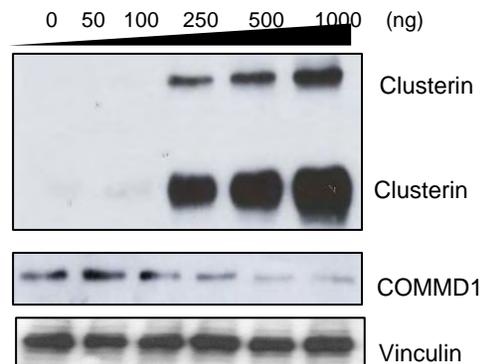
sCLU-2 is a COMMD1 and ubiquitin binding partner in cancer cells

1. sClu-2 and COMMD1 co-localize in cytoplasm with a juxtannuclear aggregation

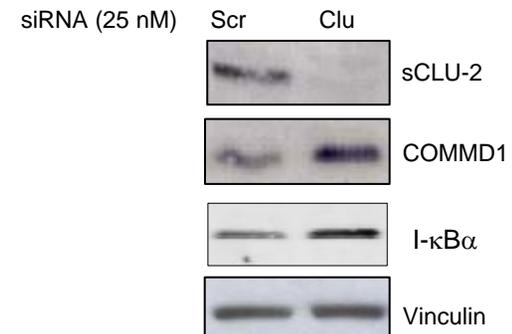


2. sCLU-2 Levels Negatively Correlate with COMMD1 & I κ -B Levels

Clu transient transfection

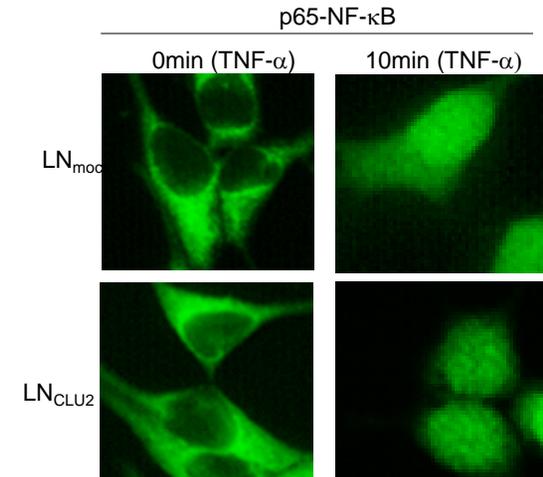
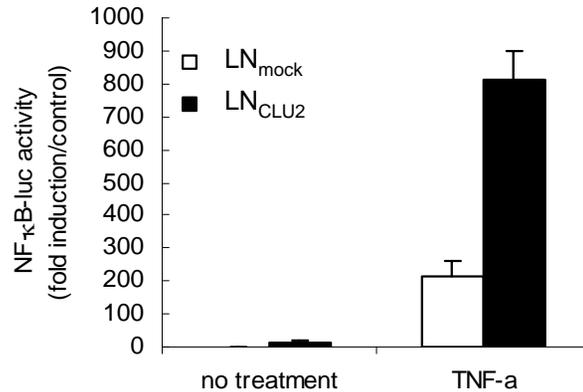


Clu Knockdown increases Levels of COMMD1 & total I κ -B α

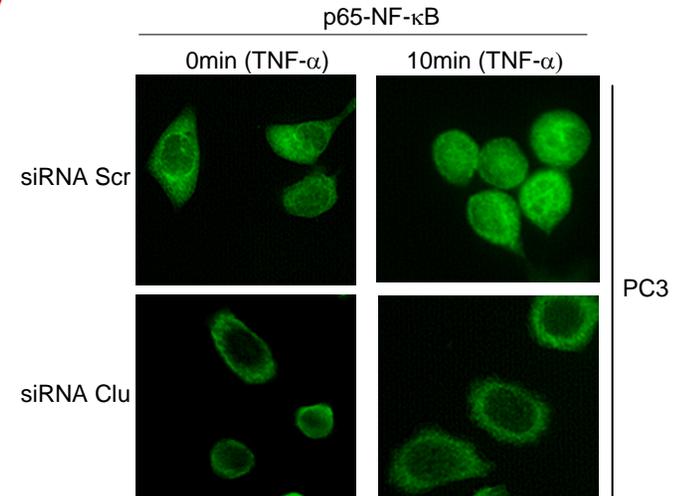
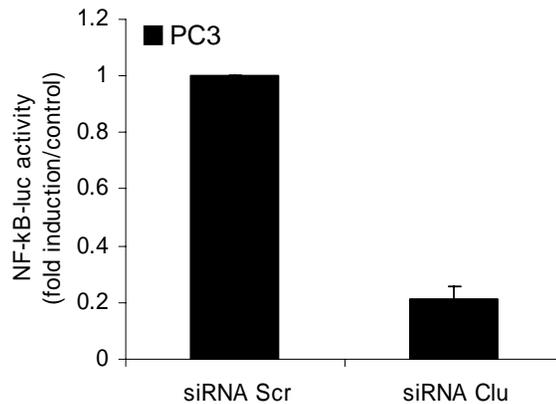


sCLU-2 Enhances TNF- α induced NF- κ B Nuclear Translocation and Transcriptional Activity

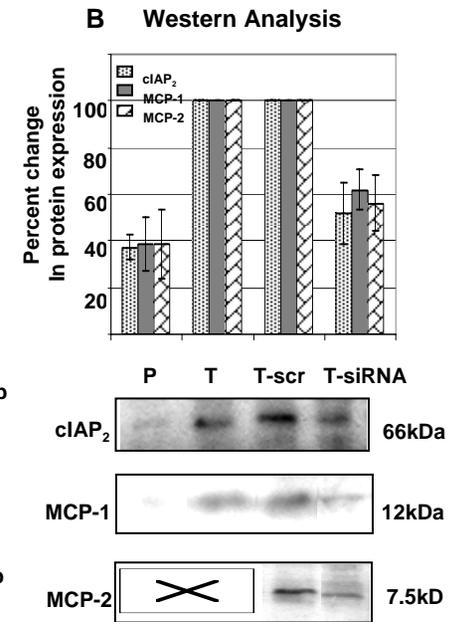
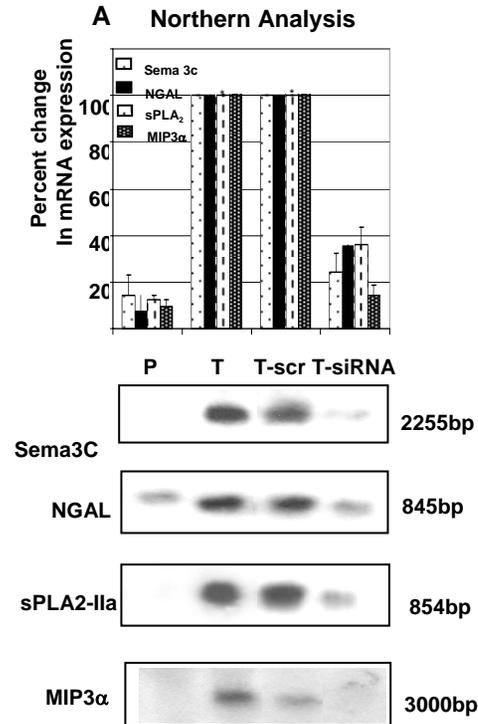
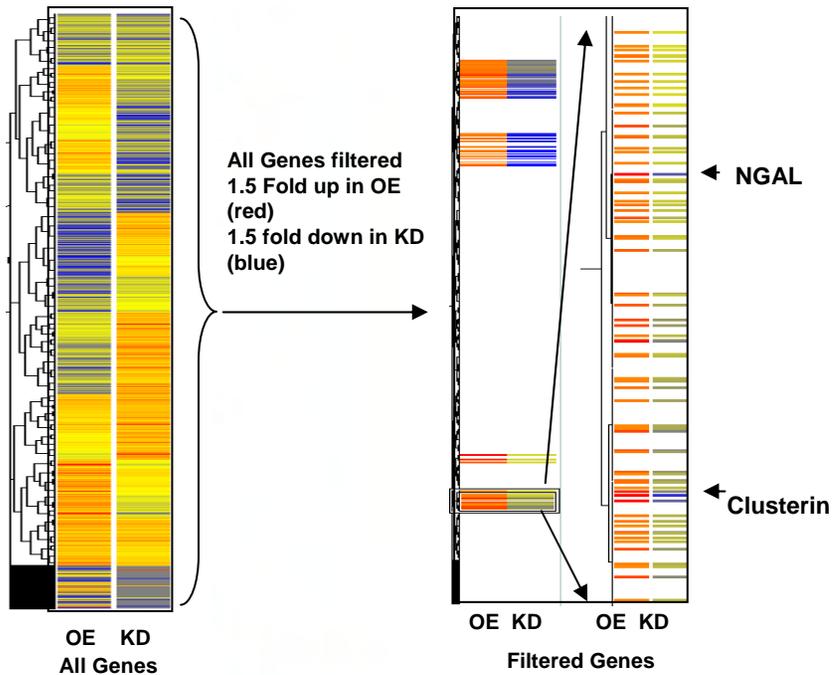
1. sClu-2 Overexpression $\uparrow\uparrow$ NF- κ B Activity



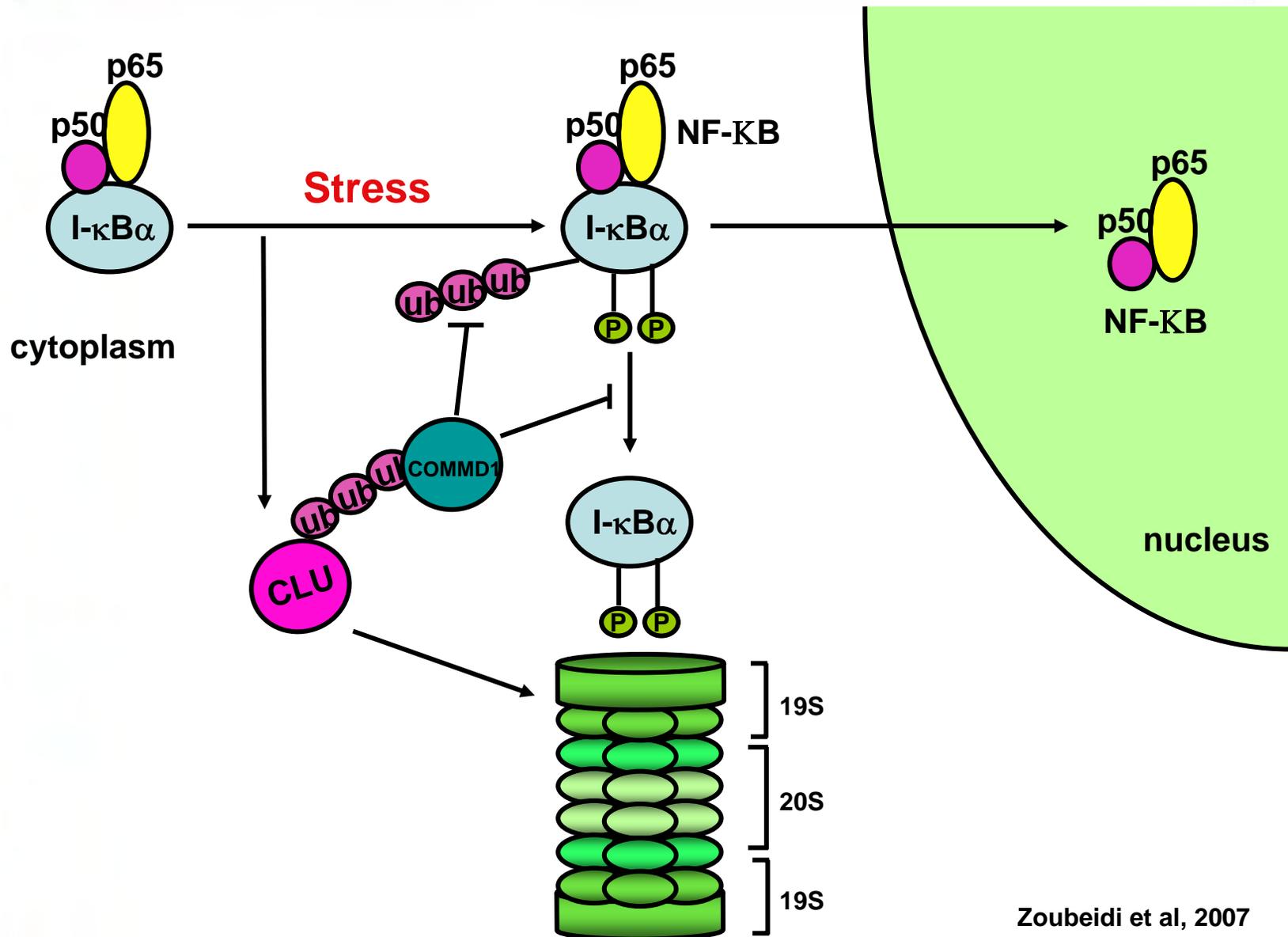
2. sClu-2 Knockdown $\downarrow\downarrow$ NF- κ B Activity



Clusterin Expression Levels Positively Correlate with NF- κ B - regulated Genes

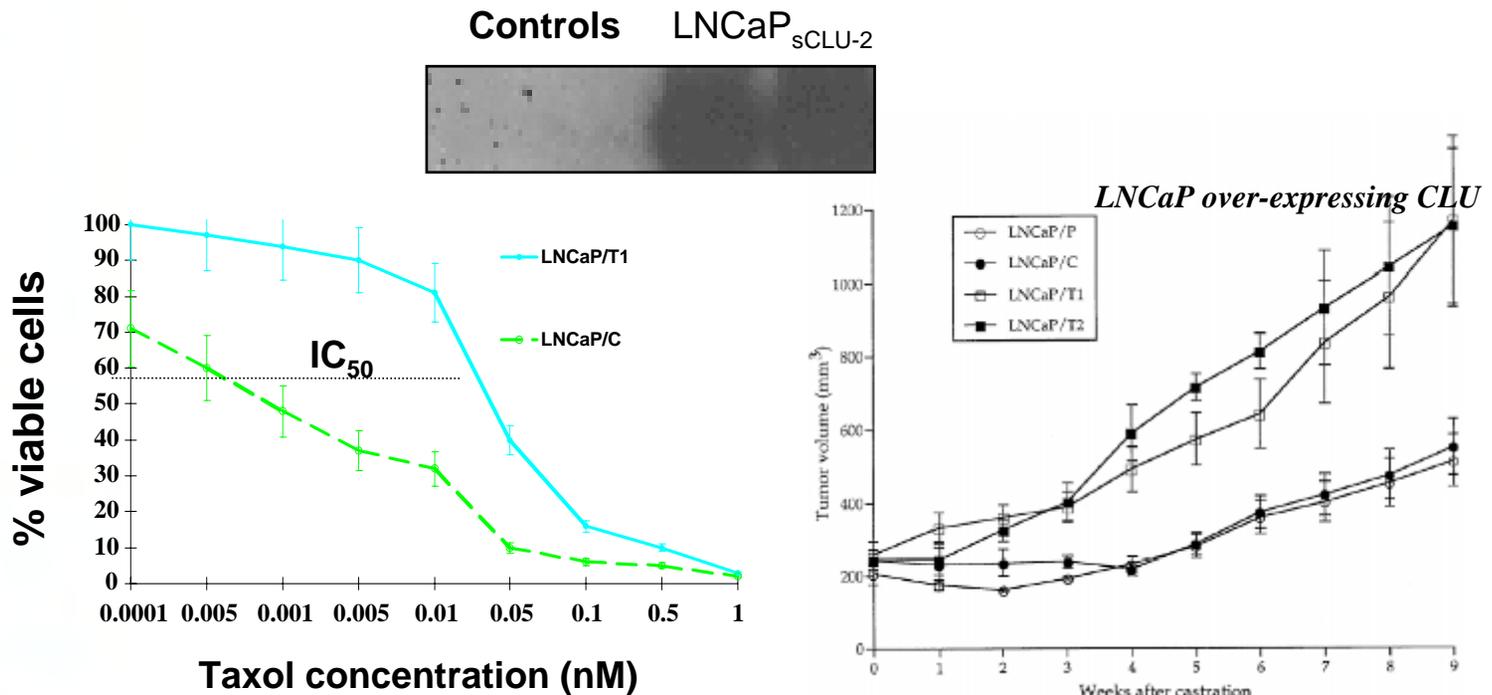


sClu-2 Enhances COMMD1 and I- κ B α Degradation by the Proteasome



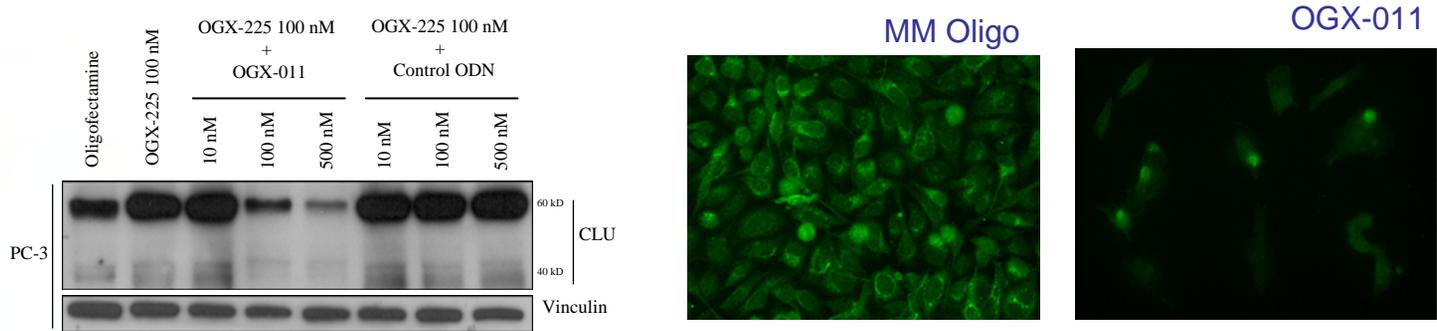
Functional Significance of sCLU-2 Over-expression in Prostate Cancer

- sCLU overexpression is antiapoptotic: confers broad spectrum treatment resistance including hormone, radiation, and chemo-therapy

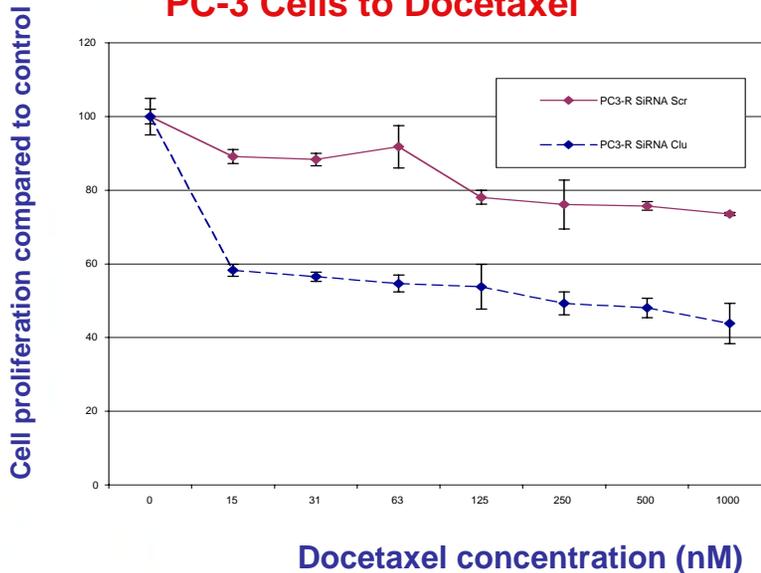


Inhibition of Clusterin Expression Enhances Activity of Chemotherapy in Prostate Cancer Cells

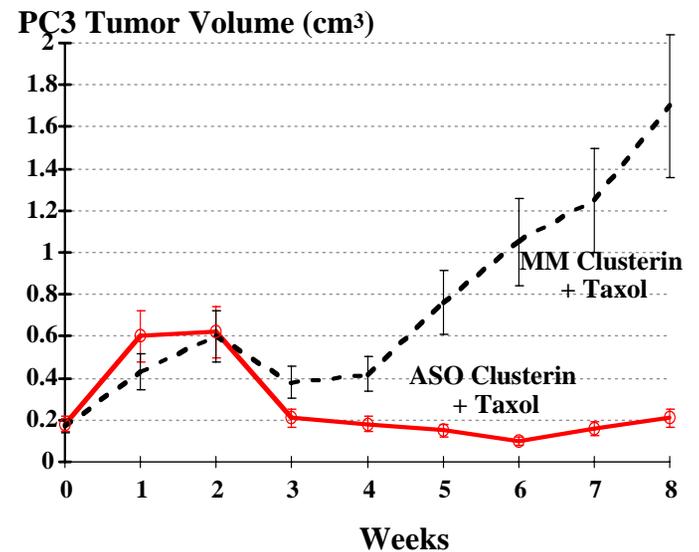
CLU ASO (OGX-011) Suppress Clu Levels in PC-3 Cells



OGX-011 Chemosensitizes PC-3 Cells to Docetaxel

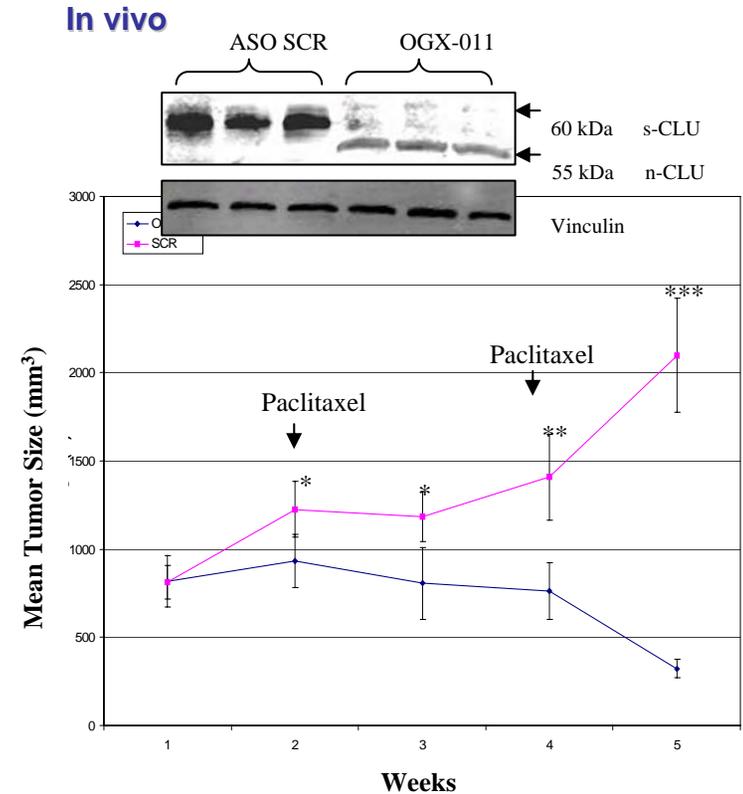
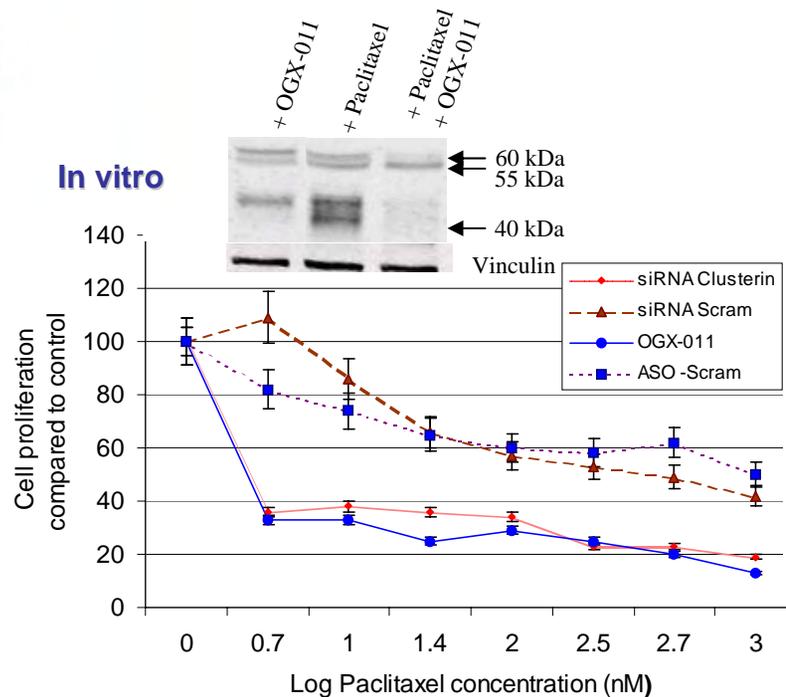
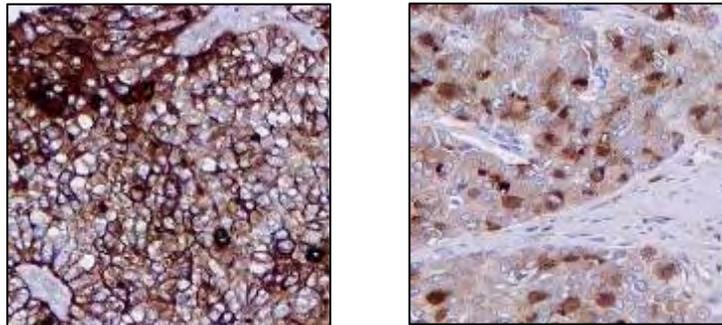


OGX-011 Enhances Taxol Activity in PC3 Tumours in vivo



CLU ASO (OGX-011) Suppresses sCLU Levels and Chemosensitizes MCF-7 Xenografts to Paclitaxel in vivo

Clusterin is expressed in 65% of Primary Breast Cancers



From Bench to Bedside: Translational Research in Action

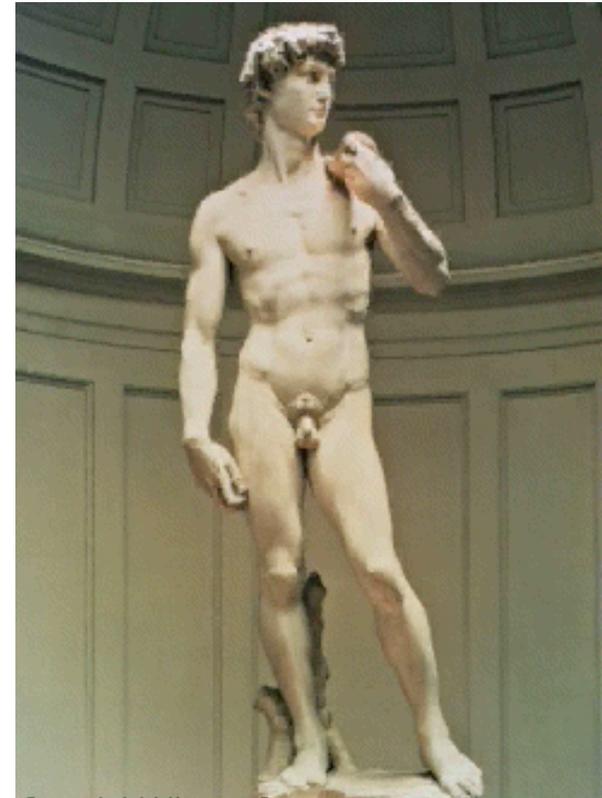
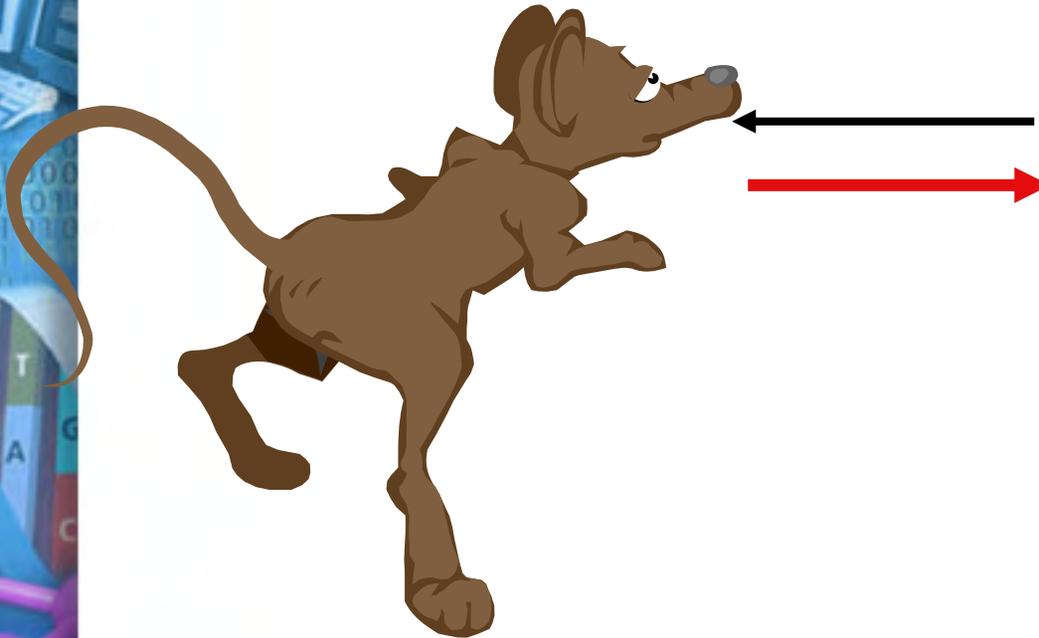


sCLU as a Therapeutic Target: Preclinical Studies For Proof of Principle

Of Mice and Men

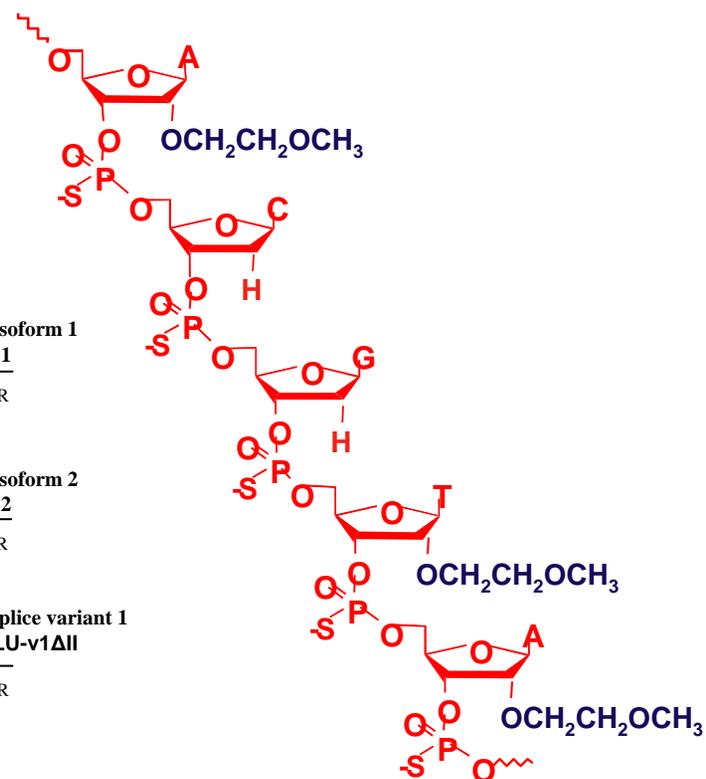
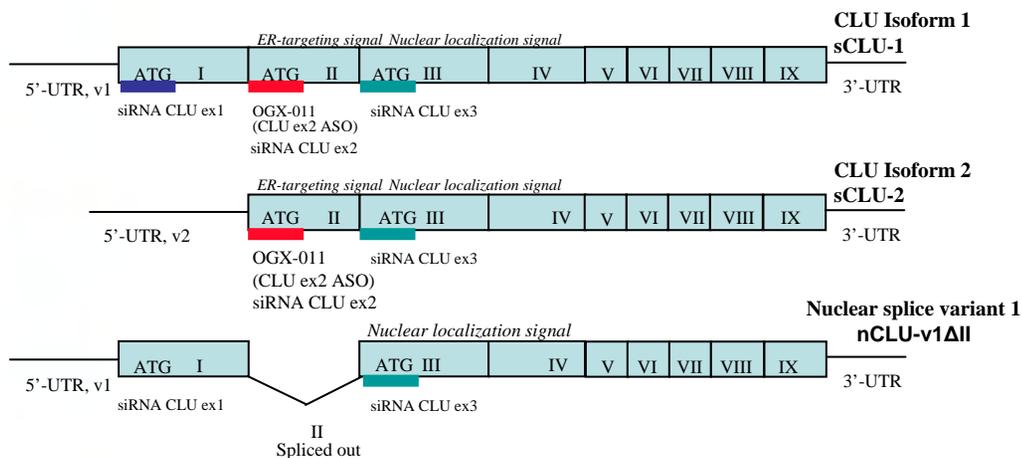
Clusterin:

- Stress-induced survival response
- confers resistance
- knockdown enhances chemo & HT in many tumor models



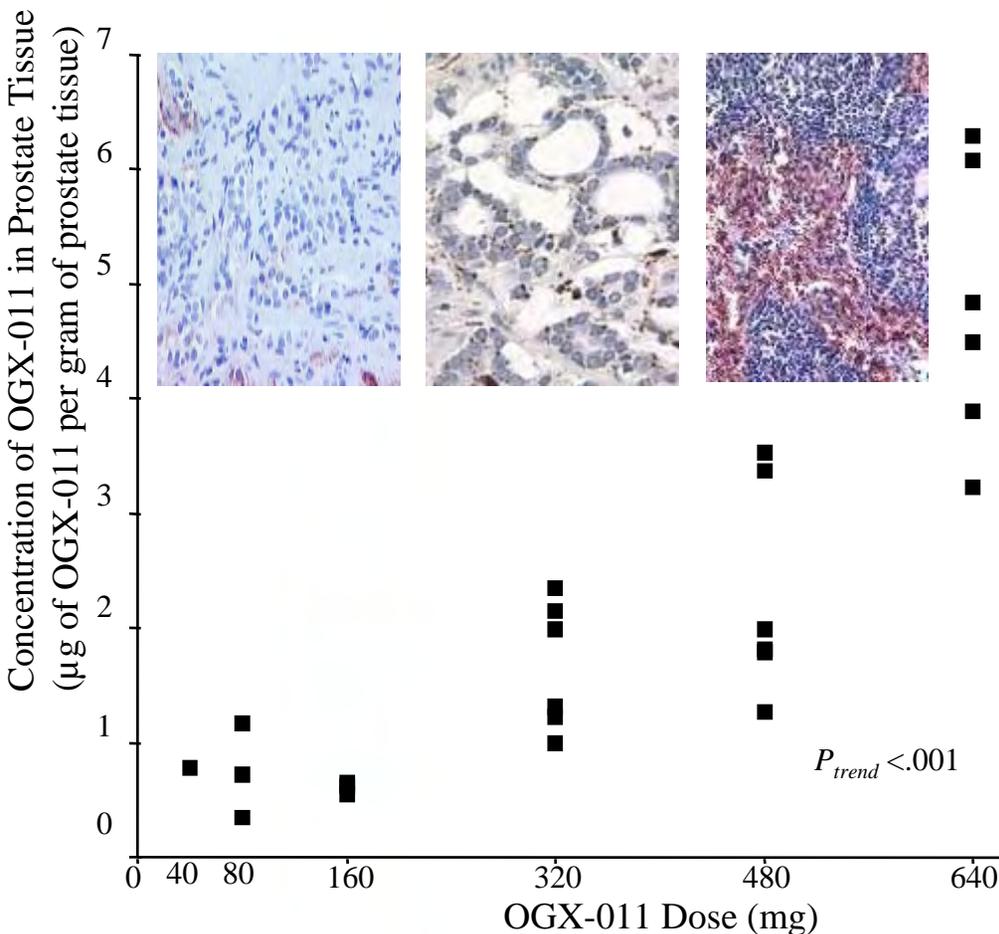
Antisense Clusterin: OGX-011 Product Description

- Licensed from UBC for development by OncoGenex in collaboration with Isis**
 - 2nd generation antisense molecule
 - 4-13-4 21-mer MOE gapmer oligonucleotide
- Advantages of 2'MOE analogues**
 - Increased potency and resistance to degradation
 - Facilitates more convenient dosing regimen
 - once-weekly infusion
 - J Pharmacol Exp Ther. 298(3):934-40, 2001

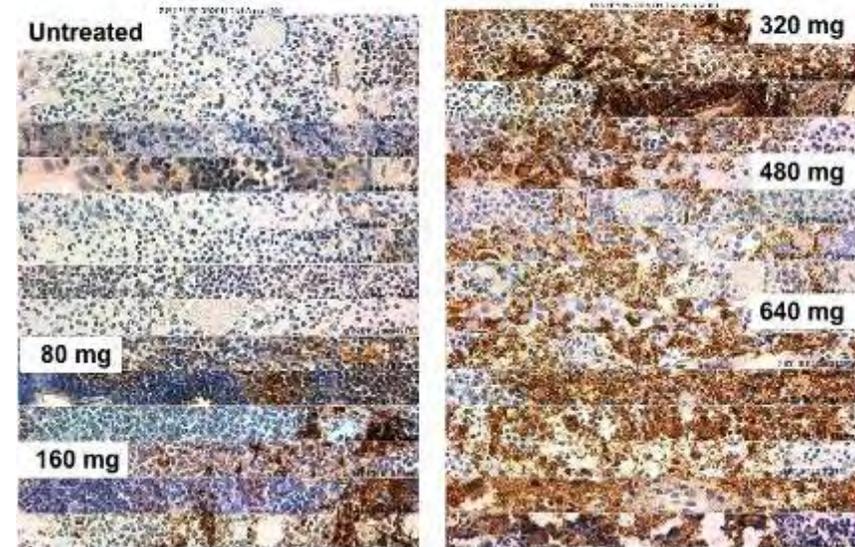


NCIC IND.153: Phase I Pre-Surgery pk/pd Trial of OGX-011 - Tissue Pk data

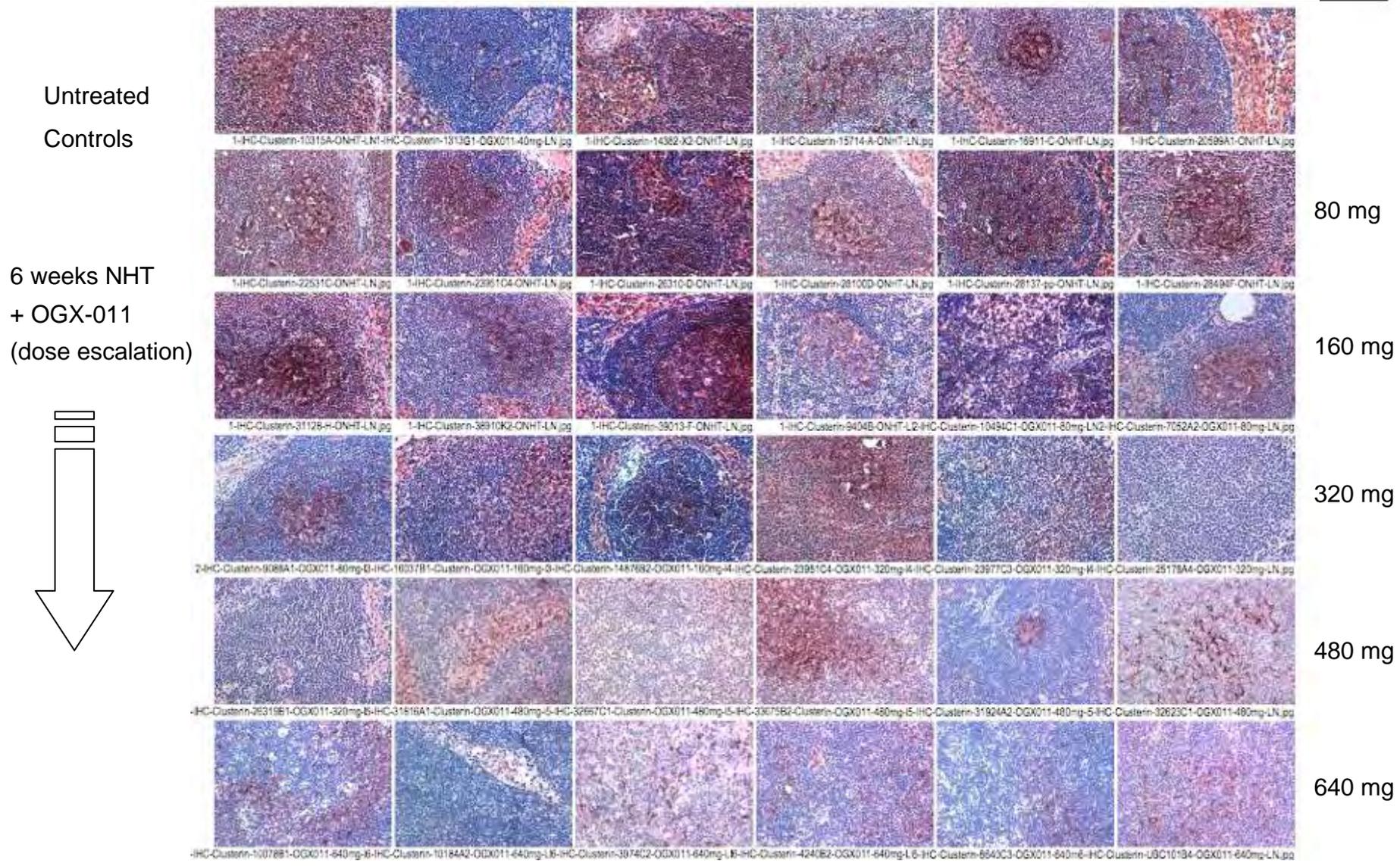
- 25 men with localized CaP treated with 5 weeks of NHT + escalating doses of OGX-011



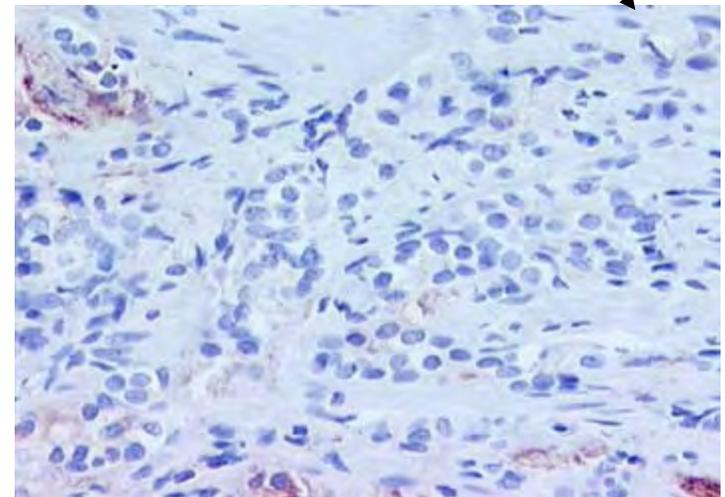
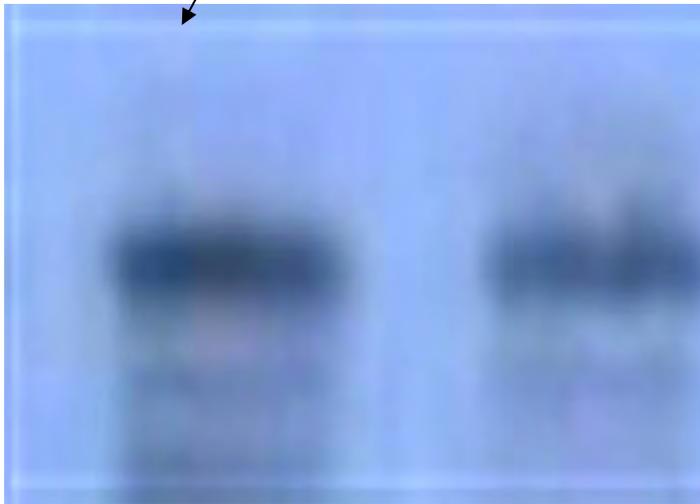
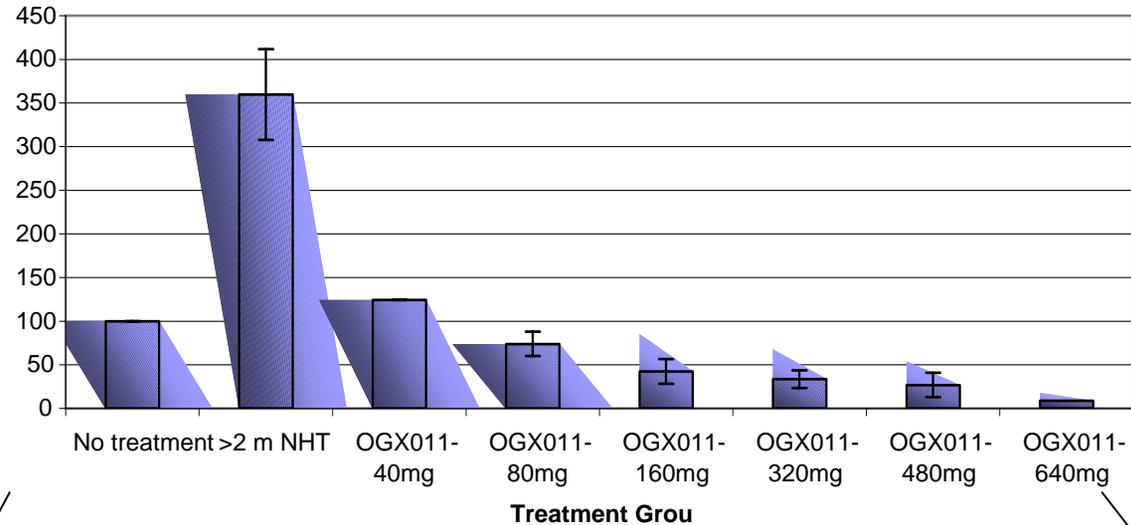
OGX-011 IHC in Lymph Nodes In NCIC IND.153



IND.153: Target Regulation Data: Dose-dependent suppression of clusterin in Regional Lymph Nodes



Clinical Proof-of-Concept: Dose-dependent Decreases in Clusterin Levels in RP Specimens using LCM and Real-Time PCR



Clinical Trial Development with Clusterin ASO (OGX-011)

Phase I
NHT
Pre - surgery

**Established 640 mg once weekly
as phase II dose (OBD) based on
tissue pk and pd criteria**

Phase I
Solid Tumors +
docetaxel n = 35

- Completed:**
- 2 regimens found well tolerated -
 - OGX-011 + weekly Taxotere
 - OGX-011 + q 3 weekly Taxotere
 - 30-60% decrease in serum CLU levels

**NCIC IND.165:
First Line mHRPC
Phase II RCT -
Taxotere +/- OGX-011**

- 82/80 pts enrolled
- ASCO May 2007

**CUOG P-06a:
2nd line mHRPC
Phase RCT Taxotere or
mitoxantrone + OGX-
011**

- 42/40 pts
enrolled

**Phase II stage III/IV
NSCLC
Gem/cis + OGX-011**

- 10 pts Phase I
- 70 pts Phase II
- ASCO May 2007

**NCIC IND.164:
Phase II Metastatic
Breast- Taxotere +
OGX-011**

- 16 pts enrolled

Phase 2 Study in 1st Line NSCLC: Treatment Schema

•81 pts with stage IIIB/IV NSCLC treated with gem/cis plus OGX-011

Results as of May 24, 2007	
Median Follow-up	12.7 months
Number of Deaths	37/81 (46%)
Median Progression-Free Survival (range)	4.6 months (0.06-15.6+)
Estimated Median Survival	14.1 months
Number of Patients Surviving \geq 1 year	25/46 = 54% *
Number of Patients Surviving \geq 18 months	8/22 = 36%

	Historical Controls*	Phase 1 and 2 (n=81)
Median Survival	8.0 – 10.8 months	14.1 Months (estimated)

•Data from five randomized clinical trials using gemcitabine plus platinum-based chemo in 1st line NSCLC (1260 patients)

NCIC IND.165: Taxotere +/- OGX-011 in First-Line mHRPC

- Randomized Open label, multicentre trial comparing docetaxel +/- OGX-011 in men with mHRPC (PI - K. Chi).

PSA Response Rates

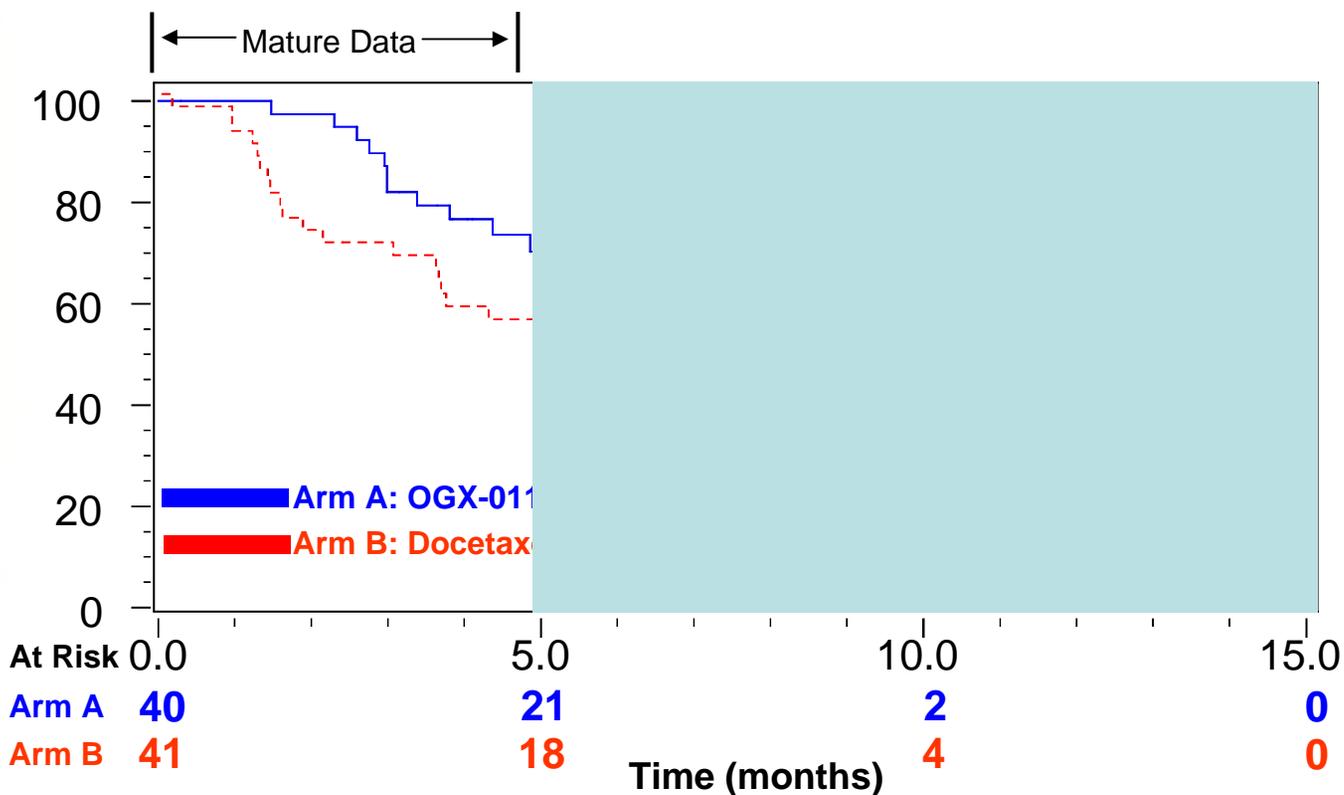
BEST RESPONSE CRITERIA	Arm A (OGX-011 + Docetaxel) N=40	Arm B (Docetaxel) N=41	% Change in favor of OGX- 011
≥ 50% PSA Decline at 12 weeks	45%	34%	32%
PSA Response (50% decline - confirmed)	50%	51%	NA
≥ 80% PSA Decline	38%	22%	73%
PSA Progression (PSAWG Criteria)	0%	10%	100%
PSA Non-Progression/Non- Response	45%	32%	41%
Inevaluable	3%	2%	NA

NCIC IND.165: Taxotere +/- OGX-011 in First-Line mHRPC

RECIST CRITERIA	Arm A (OGX-011 + Docetaxel) n=26	Arm B (Docetaxel) n=23	% Change in favor of OGX-011
Disease Control (CR+PR+SD)	92%	74%	24%
Complete Response	0%	0%	N/A
Partial Response	19%	22%	-14%
Stable Disease	73%	52%	40%
	9.7 months	7.6 months	28%
Progressive Disease	4%	22%	82%
Inevaluable	4%	4%	N/A
Median PFS	7.3 months	5.9 months	24%

NCIC IND.165: Taxotere +/- OGX-011 in First-Line mHRPC

Progression Free Survival



Median for Arm A (OGX-011 + Docetaxel): 7.26 months (95%CI 5.22-9.33)

Median for Arm B (Docetaxel): 5.85 months (95% CI 3.61-10.74)

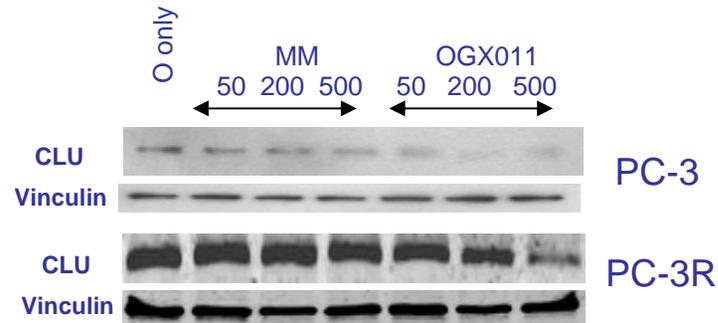
NCIC IND.165: Taxotere +/- OGX-011 in First-Line mHRPC

Indicators of Anti-cancer Activity

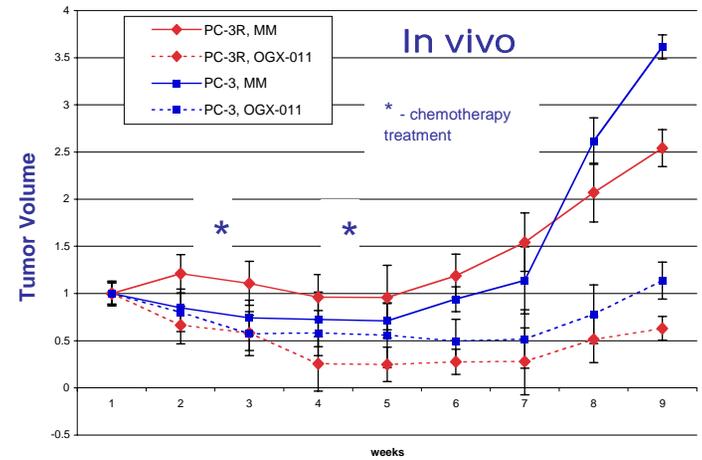
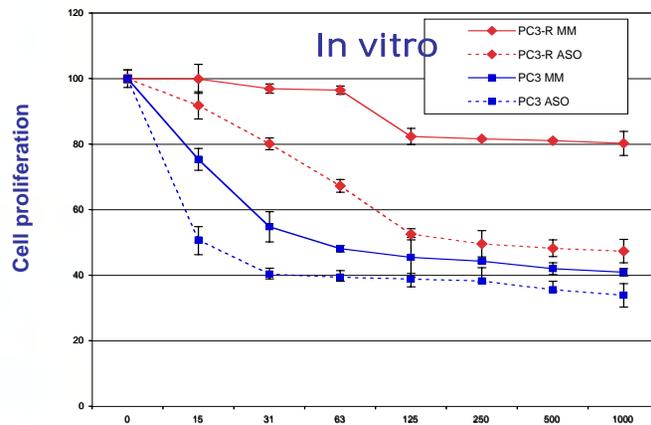
- **Consistent trend in favor of OGX-011/docetaxel arm:**
 - More patients with a 50% decline in PSA within the first 12 weeks
 - More pts (38% vs 22%) with >80% decline in PSA; less pts (0 vs 10%) with primary PSA progression as best response
 - Longer time on treatment and a greater median # of treatment cycles.
 - Higher frequency and longer duration of stable measurable disease.
 - Lower frequency of progressive disease as “best response”.
 - Longer time to progression

OGX-011 in docetaxel-refractory HRPC:

CLU knockdown chemosensitizes taxane-resistant PC3-dR cells to docetaxel

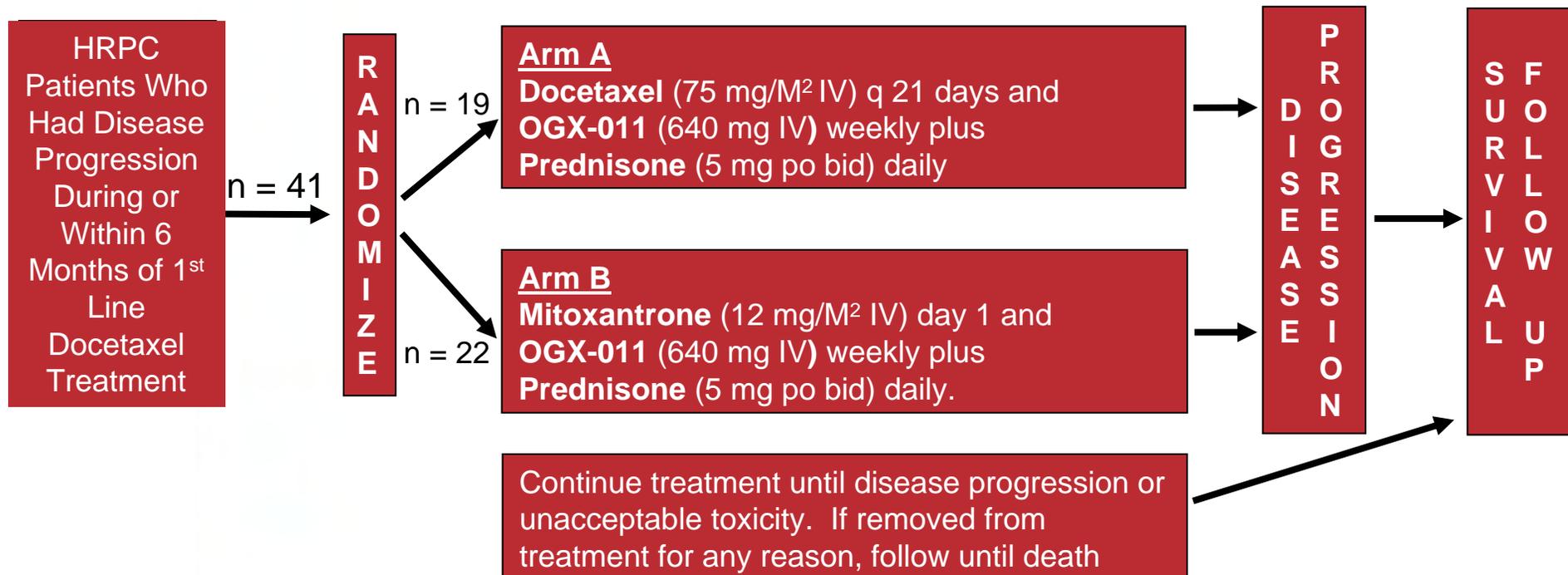


docetaxel +/- OGX-011



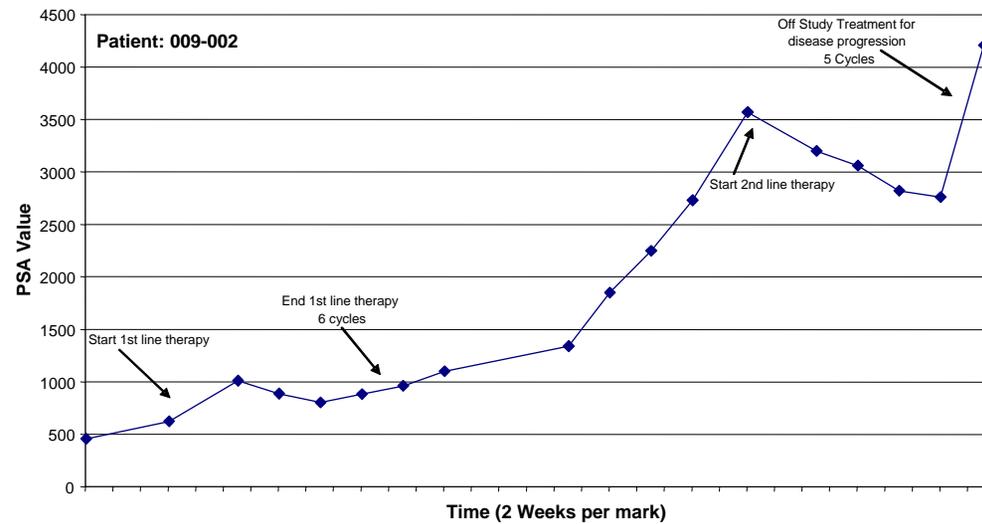
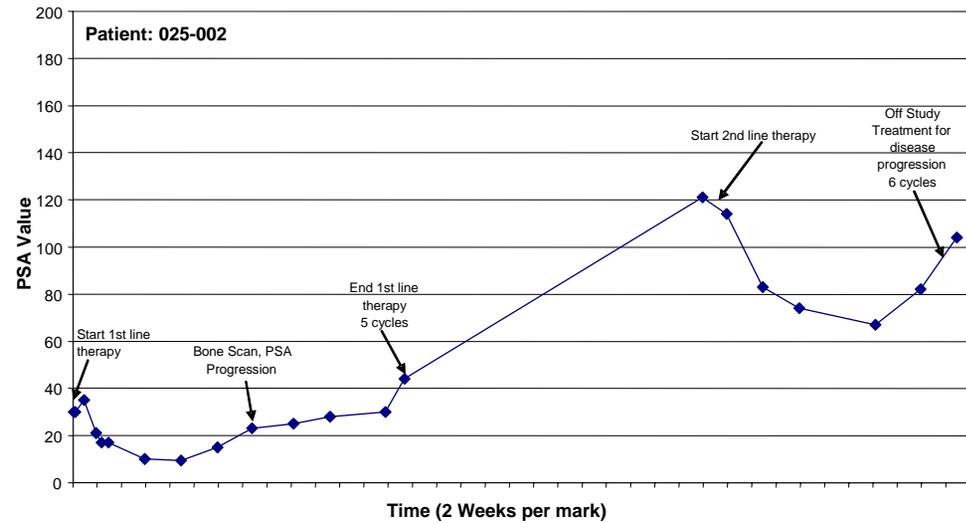
Phase II Feasibility Trial of OGX-011 in 2nd Line Therapy in HRPC:

Study treatment ongoing in 11 (26%) of patients



OGX-011 in 2nd Line Therapy in HRPC:

Chemosensitizes taxane-resistant patients to docetaxel



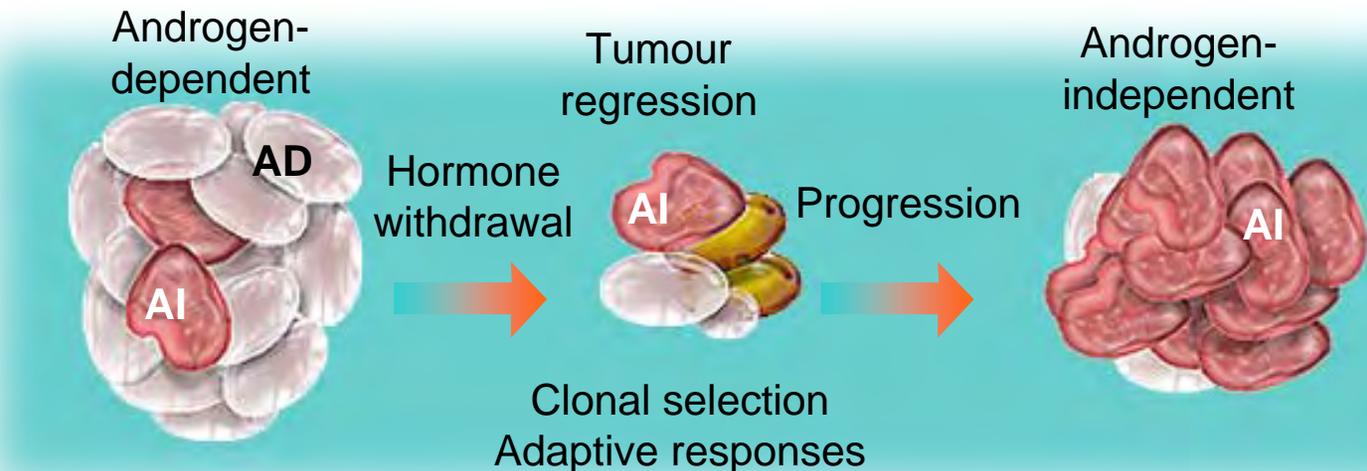


Summary:

Clusterin as a Therapeutic Target in HRPC

- **sCLU is a stress-activated cytoprotective chaperone that is highly expressed in HRPC**
- **Over-expression of sCLU-2 confers broad spectrum treatment resistance**
 - Inhibits protein aggregation, facilitates proteasome degradation of ubiquitinated proteins
 - Interacts and inhibits activated Bax, preventing cytochrome C release
 - Increases NF-κB transcriptional activity
- **CLU knockdown using OGX-011**
 - Enhances treatment-induced apoptosis in vitro and in vivo
 - Pre-clinical proof-of-principle in prostate, breast, lung, urothelial, melanoma, renal cell
 - **OGX-011, a 2nd generation ASO potently suppresses target CLU levels >90% in human CaP tissues**
 - Anti-cancer activity observed in multi-centre Phase II trials in breast, HRPC, lung
 - Phase III registration trial in second-line HRPC set to begin in 2008

Changes in Gene Expression After Castration and During AI Progression



- ++ PSA
- Bcl-2
- EGFR
- clusterin
- IGFBP 2&5
- TGFβ
- ++IGFBP 3 & 4
- YB-1
- ++survivin
- Hsp27
- + PKC-α

- ↓
- PSA
 - ++ Bcl-2
 - ++Bclx-L
 - EGFR
 - +++ clusterin
 - ++++IGFBP 5
 - IGFBP 3 & 4
 - +c-myc
 - +YB-1
 - survivin
 - +Hsp27
 - + PKC-α

- ++ PSA
- ++ Bcl-2
- ++Bclx-L
- + EGFR
- +++ clusterin
- ++ IGFBP 2
- ++ IGFBP 5
- ++YB-1
- ++ survivin
- +++Hsp27



Thanks to...



THE PROSTATE CENTRE
AT VANCOUVER GENERAL HOSPITAL

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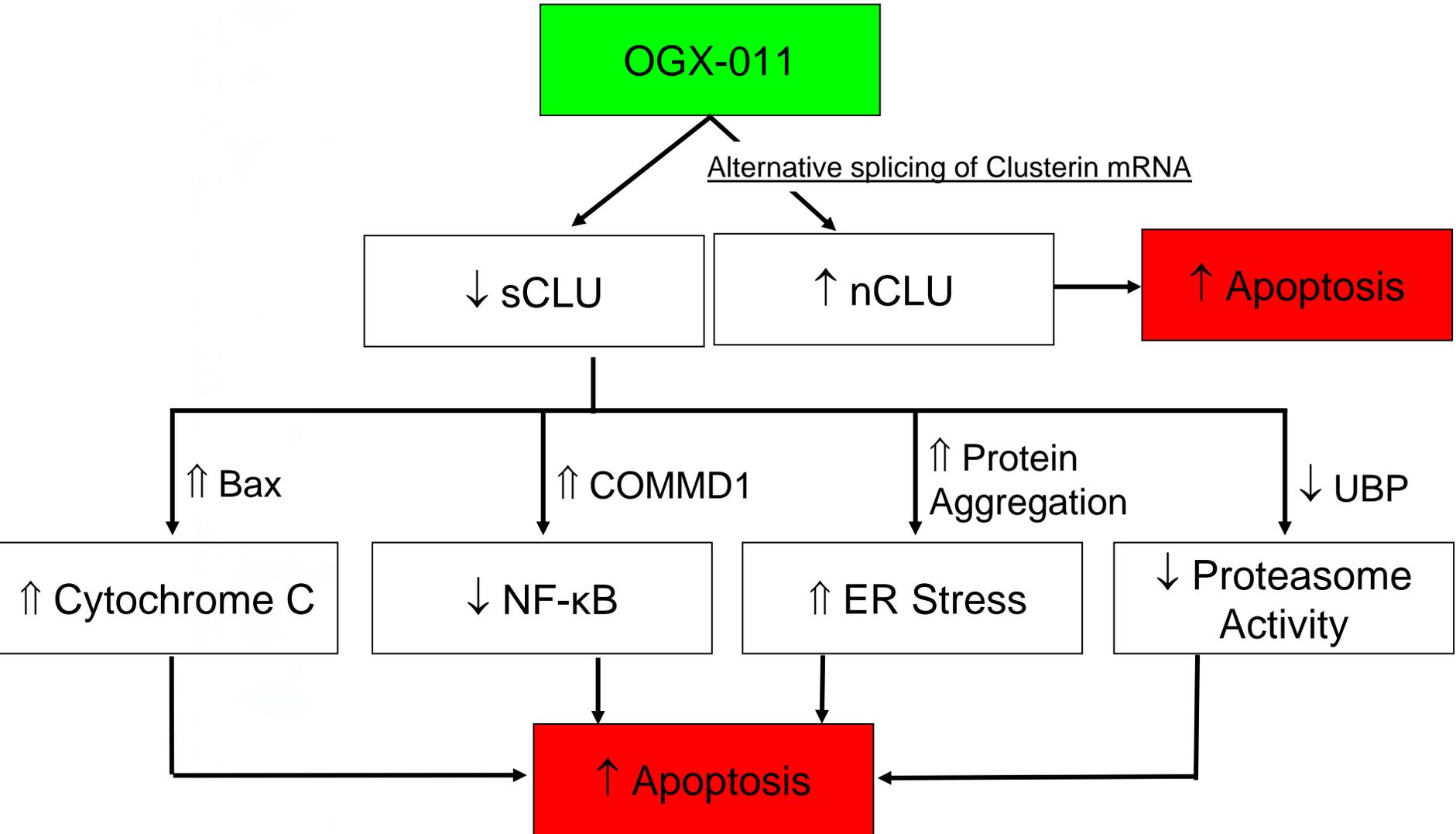
Grant Funding

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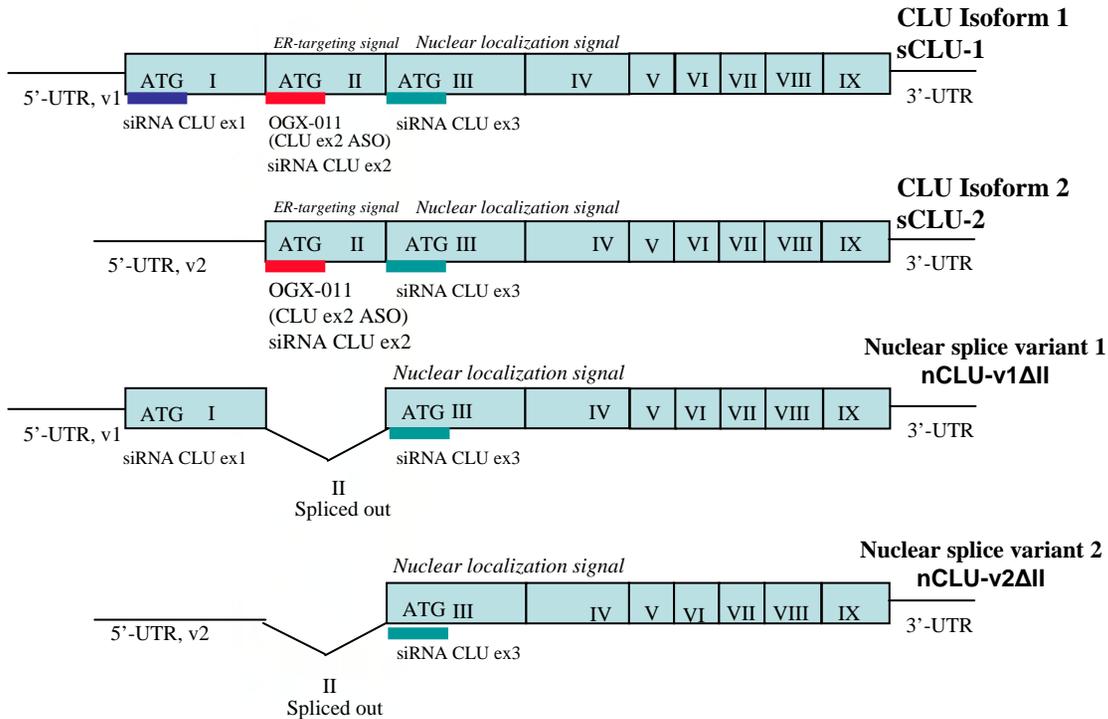
OGX-011: Safety Profile in >270 Patients

- **Well tolerated in all Phase 1 and Phase 2 studies to date**
- **Safety profile of OGX-011 in combination with docetaxel vs docetaxel alone**
 - Increase in Grade 1 or 2 AE's events (fever, rigors/chills and sweating during the loading-dose week and sensory neuropathy during therapy)
 - lymphopenia was more prevalent in the OGX-011 + docetaxel arm (no clinical sequelae)
 - No increase in SAEs in the OGX-011 + docetaxel arm
- **OGX-011 in combination with gemcitabine/platinum-based or mitoxantrone regimens**
 - Safety profile similar to that expected for regimen (no increase in expected rate of Grade 3 or higher AEs)

OGX-011 Mechanism of Action



Clusterin: Isoforms and Splice Variants



CLU is an androgen-regulated gene

