Develop Better Assays for Every Human Protein

OriGene overview

Application of transfected over-expression lysates

Application of purified recombinant human proteins

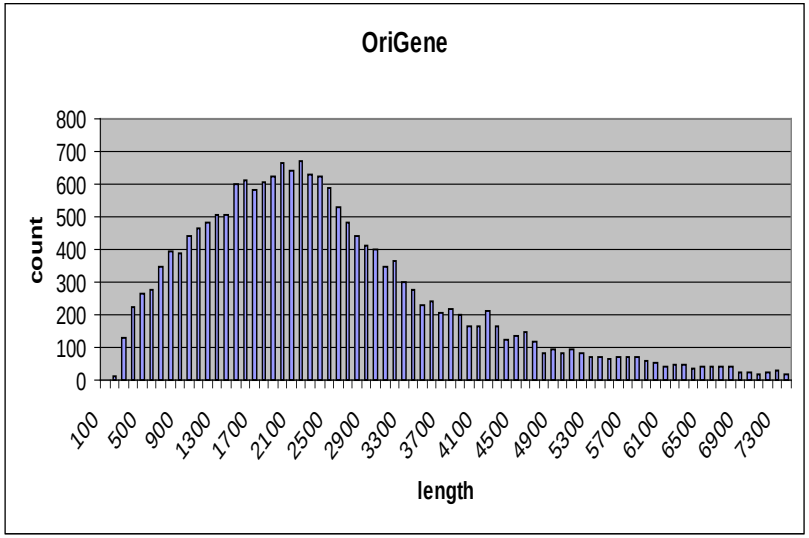
YOUR GENE COMPANY

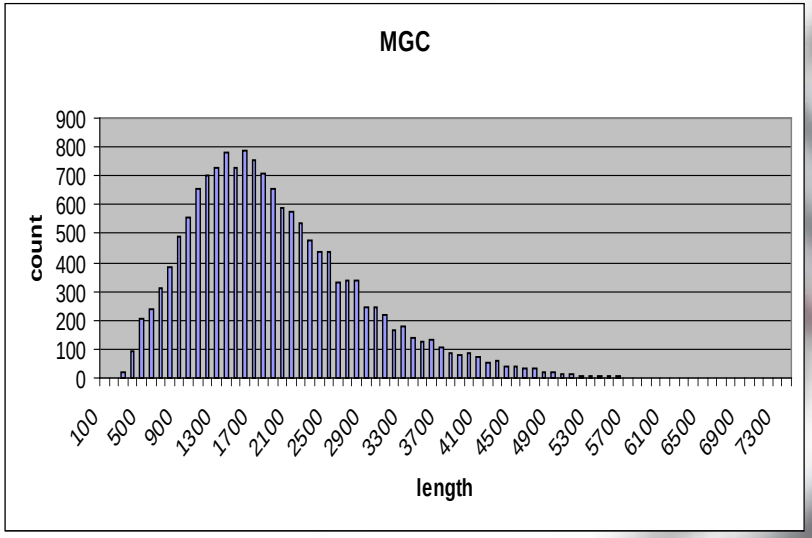
About OriGene

- Started in1996 with the goal to create largest commercial collection of full-length human cDNAs
- Develop technologies focusing on systems biology approaches to study gene function
- Provide comprehensive technology platforms for cancer biomarker discovery and validation

Largest ORF Collections

- 25,000 human and 12,000 mouse ORFs
- Including 5,000 tough genes that are only available from OriGene
- > 13,000 ready for immediate delivery

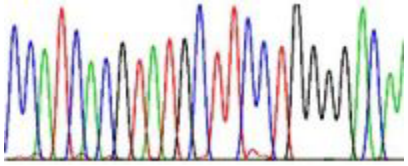




Highest Quality Standard

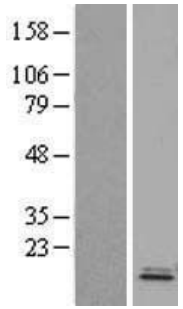
ORIGENE Technologies

Not just sequence-verification

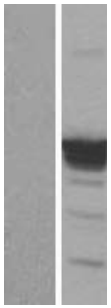


But also

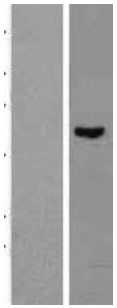
Expression Validation on 11,000 TrueORF clones



APO3C
TrueORF:RC206566
ORF size: 300 bp
Predicted MW: 11 kd

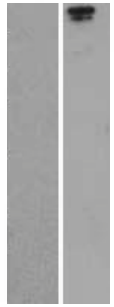


TP53
TrueORF:RC200003
ORF size: 1182 bp
Predicted MW: 53 kd



ENC1
TrueORF:RC201842
ORF size: 1770 bp
Predicted MW: 65 kd

CDH3
TrueORF:RC207346
ORF size: 2490 bp
Predicted MW: 91 kd



LRP1
TrueORF:RC218369
ORF size: 13635 bp
Predicted MW: 504 kd

Application Ready

Vectors

All ORF clones are in an expression ready vector with Myc-DDK tag or GFP tag

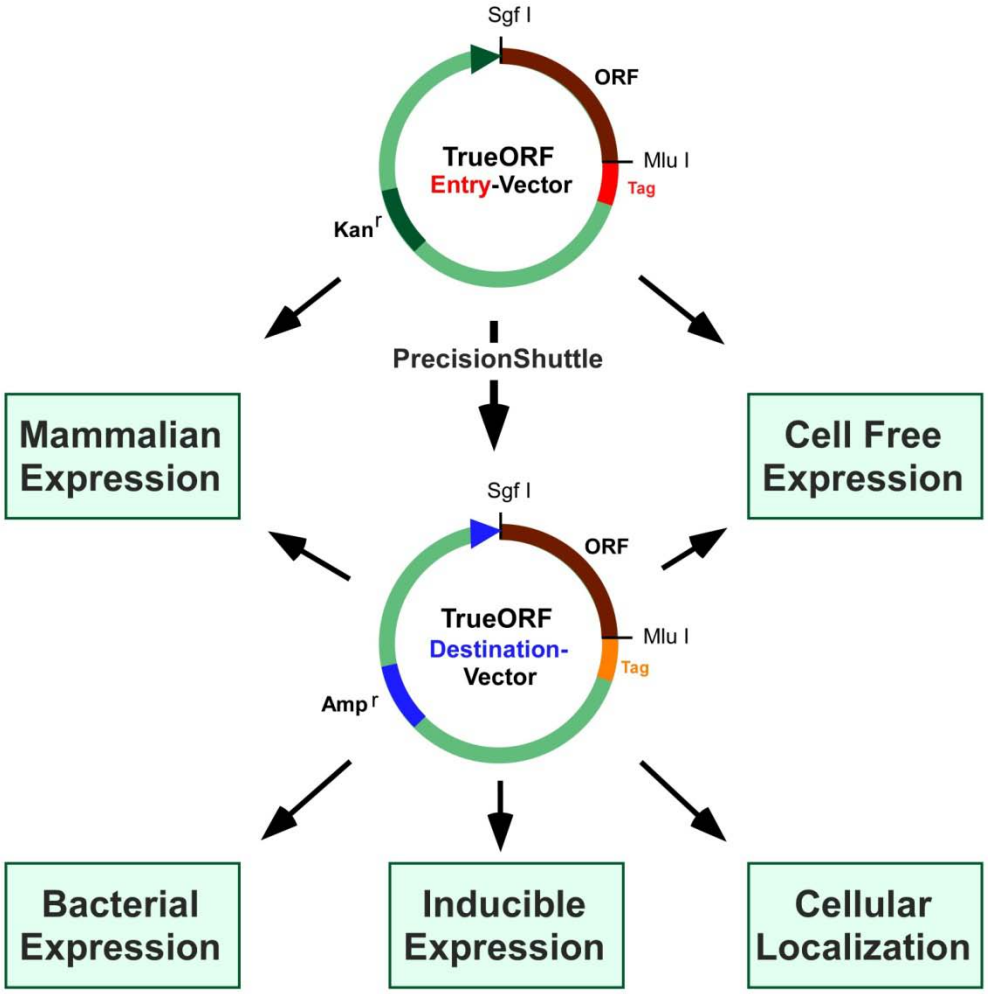
All gene inserts can be easily shuttled to any of > 60 destination vectors through simple “cut-and-paste” protocols (PrecisionShuttle)

Minimal Post-purchase manipulation

10 ug transfection grade plasmid DNA is supplied for each ORF

Large quantity plasmid for follow-up screen can be re-ordered at affordable price

PrecisionShuttle



Complete Gene/clone Resource

ORIGENE Technologies

ORIGENE Your Gene Company

TrueORF/Gold
Human/mouse
Expression vectors

5,000 human
Custom purification

TrueMAB

TruePlex

Clones/genes → **Proteins** → **Antibodies** → **Assays**

Any species
Any sequences
Any variants

Optimized expression

Antibody engineering

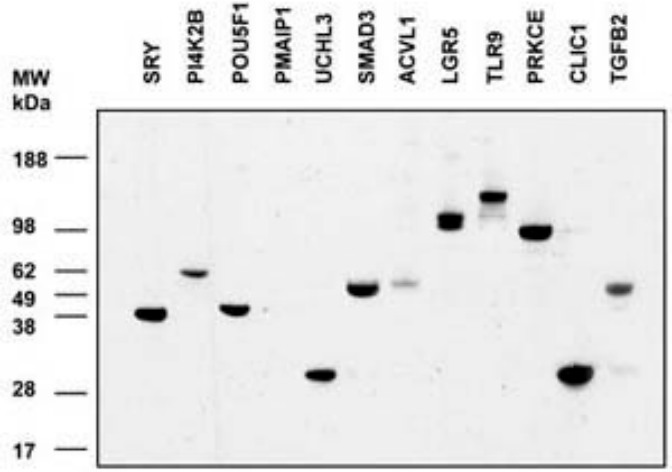
BlueHeron® The Gene Synthesis Company

Application of Over-expression Protein Lysates

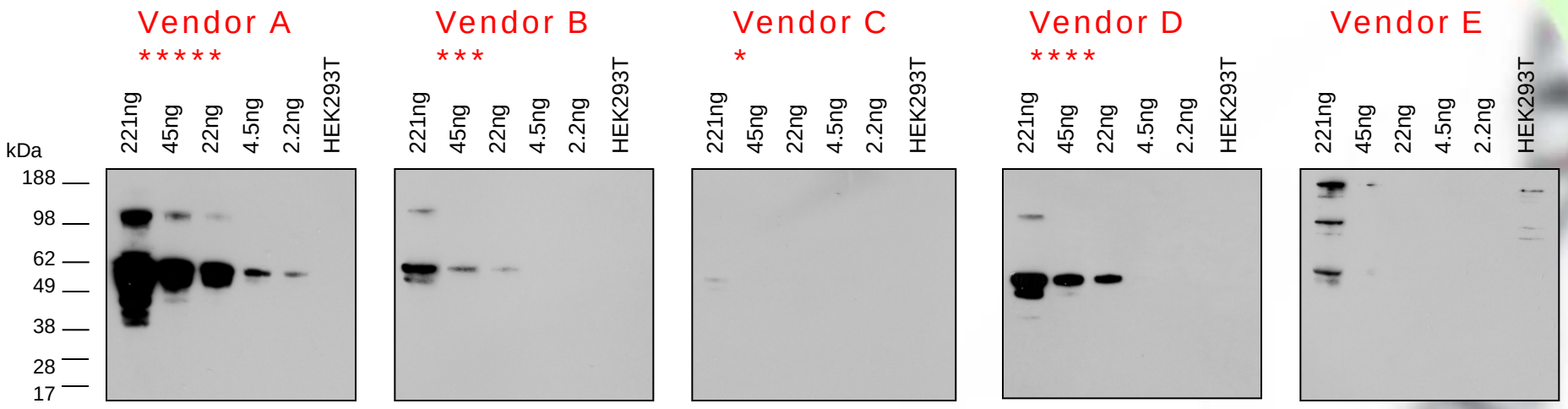
Transfected Over-Expression Lysates

ORIGENE Technologies

- 11,000 TrueORF clone transfected HEK293T cell lysates
- Mammalian cell expressed full length, WB validated
- Delivered in modified RIPA buffer to preserve protein function
- Ideal tools for antibody validation, protein function studies, and as assay standards



Antibody Validation

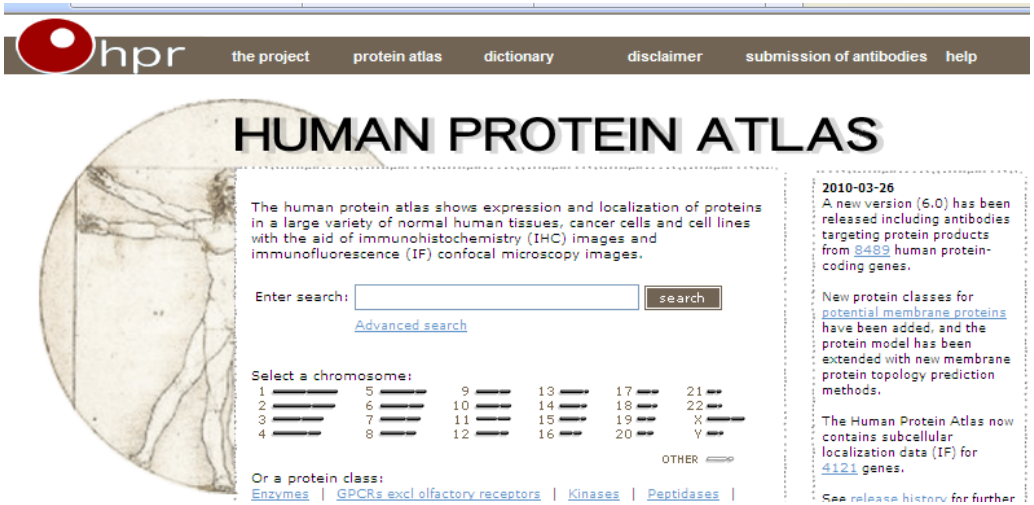


Five commercial antibodies against human p53 were evaluated in Western blot experiments with p53 over-expression lysate. (According to Ab vendors' suggested protocols)

The p53 protein level in a cell lysate was pre-determined using a purified GST-Myc-DDK standard. Lysate was serially diluted before immunoblotting.

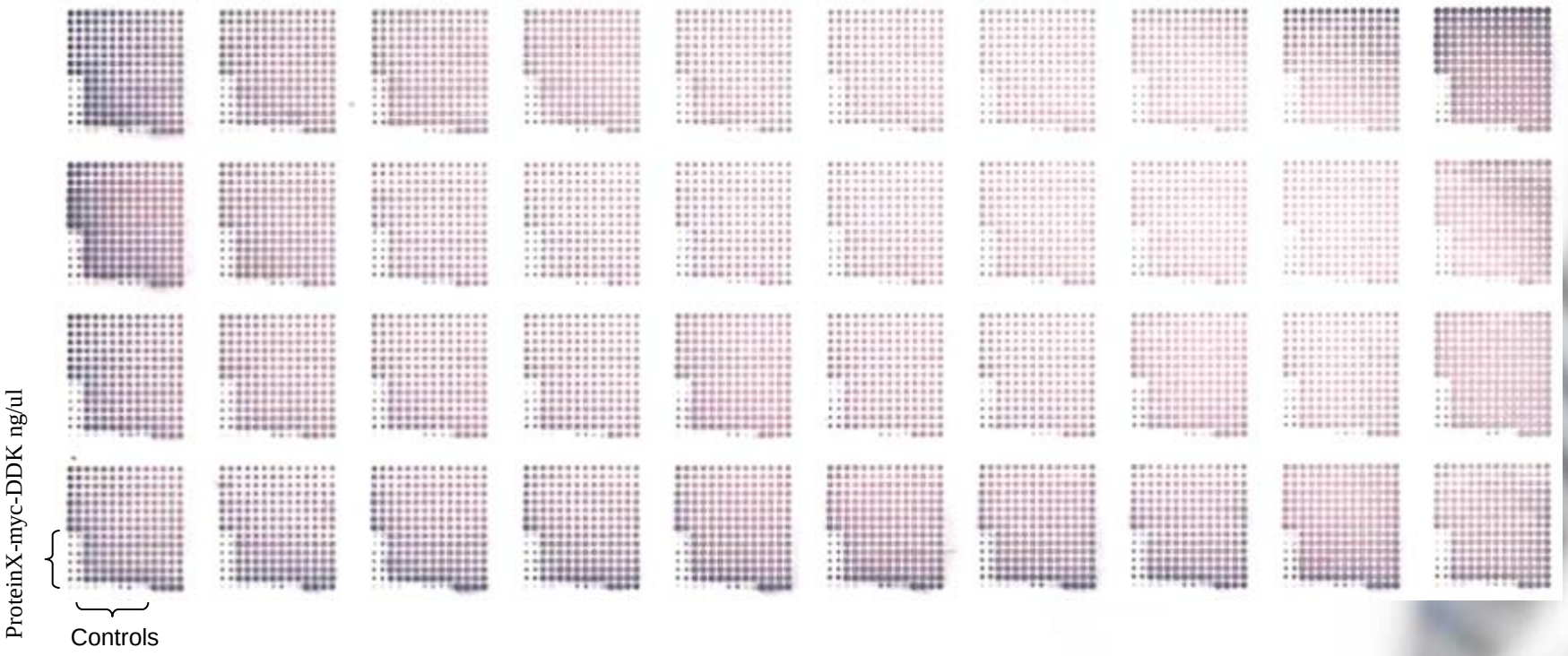
The antibody quality and star rating is based on specific p53 protein detection level.

Over-expression Lysates



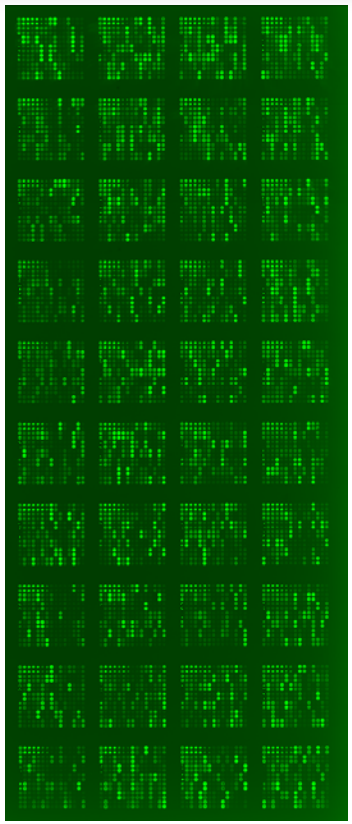
OriGene over-expression lysates adopted in Antibody Atlas Program to validate polyclonal antibodies. Success rate is improved from 40% with regular cell lysates to 80% with over-expression lysates. Data from Prof Mathias Uhlén, Program Director.

10,000 Lysate Protein Arrays

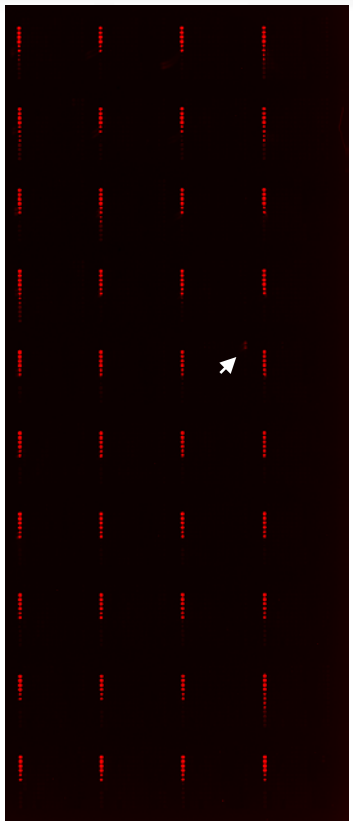


More than 20,000 features (10,000 unique overexpression lysates) and purified control proteins were spotted on this single nitrocellulose slide. To examine the morphology for each feature, the slide was stained with colloidal gold for total protein.

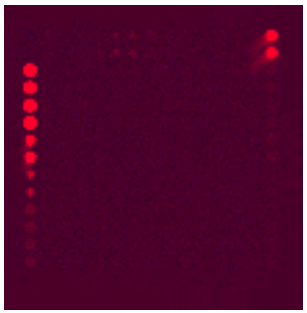
10,000 Lysate Protein Arrays



Anti-DDK (Rabbit)



Anti-E-Cadherin1 (mAb)



Enlarged image

Using OriGene antigen microarray chip technology
to decode antibodies that were generated by whole cell immunization

ORIGENE Technologies

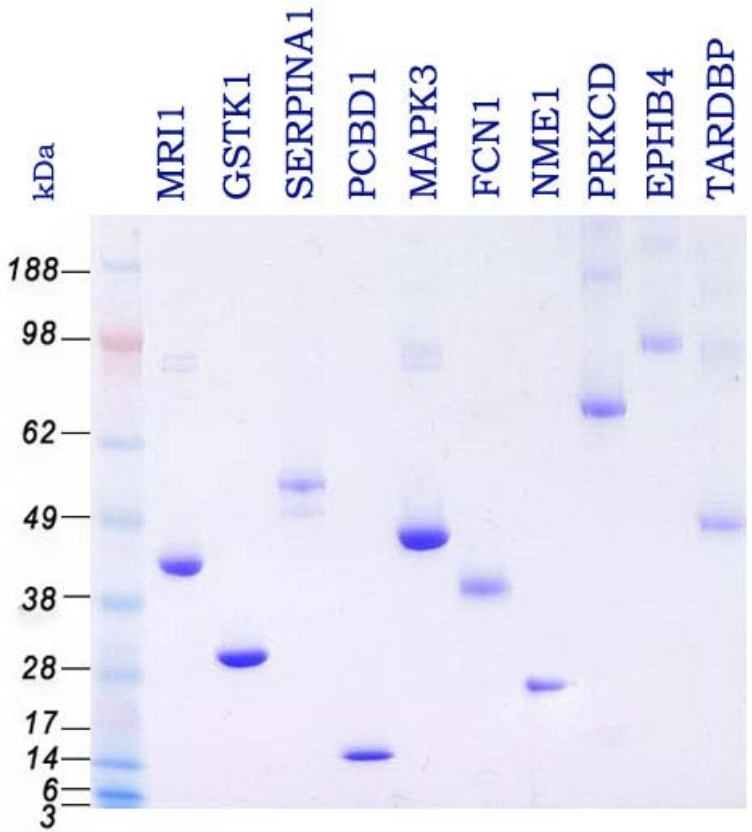
Application of Purified Proteins

YOUR GENE COMPANY

Full Length Human Proteins

ORIGENE Technologies

- Produced from TrueORF cDNA clones
- 5,000 full length human proteins
- Expressed in HEK293 cells
- Optimal preservation of protein structure, post-translational modifications and functions

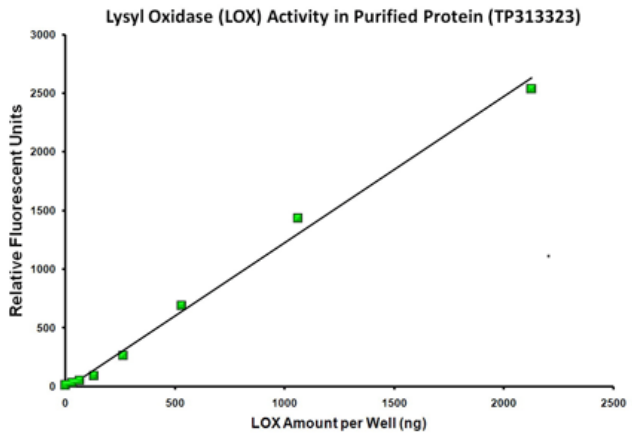


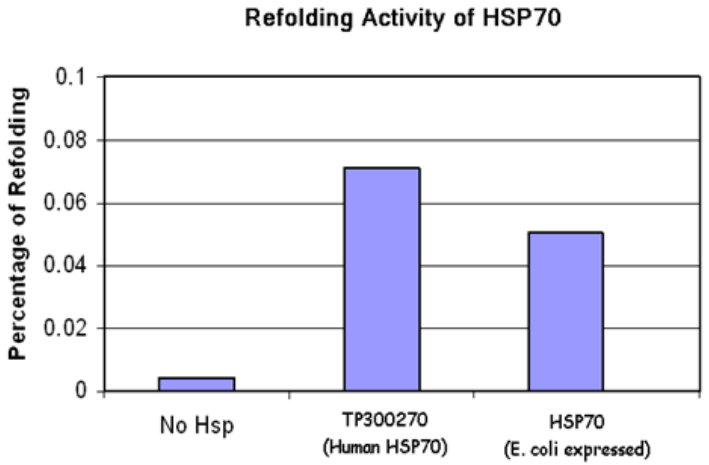
Benefits of Mammalian Expression

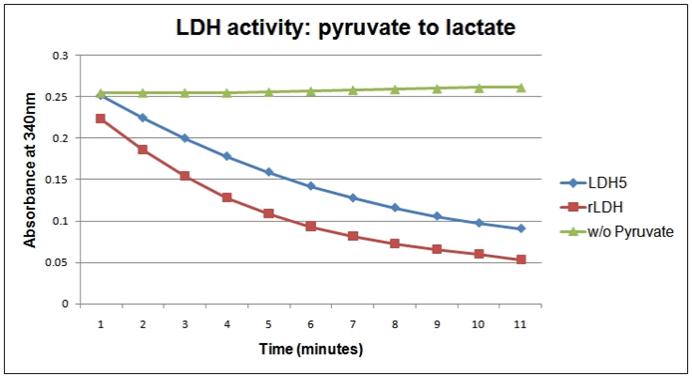
ORIGENE Technologies

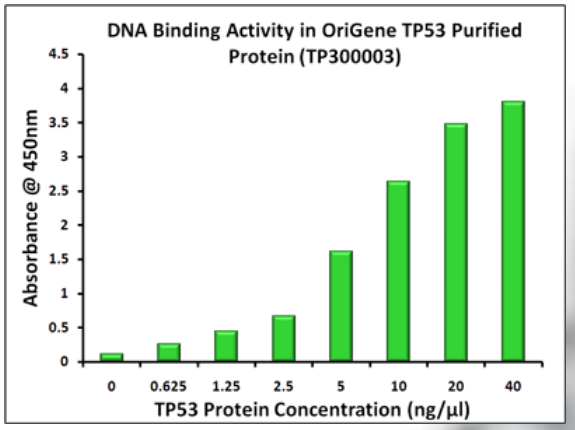
	Mammalian	Yeast	Insect cells	E. coli
Protein folding and purification	Optimal	Poor	Low	Poor
Post-translational processing	Yes	Low	Low	No
Authenticity & Bioactivity	Native and active	Poor	Poor	Very poor

Protein Functional Data









MS Collaboration with ISB

ORIGENE Technologies

- Generate signature peptide sequences for all 5,000 human proteins
  - Sequences will be in PeptideAtlas database
- Heavy-isotope labeled purified proteins
  - 5,000 available now for SRM/MRM application
- Isotope labeled peptides
  - Coming in 2011 for all 5,000 human protein

INSTITUTE FOR Systems Biology™ *Revolutionizing science. Enhancing life.*

Rapid Assay Development

ORIGENE Technologies

- c-Myc

  FELLPTPPLSPSR, LASYQAAR, DSGSPNPAR, SESGSPSAGGHSKPPHSPLVLK, DYPAAK, QISNNR, SSDTEENVK, THNVLER, SFFALR, DQIPELENNEK, ATAYILSVQAEEQK, LISEEDLLR

- Notch 1

  QGVDVAR, ASLLPGGSEGGR, GSIVYLEIDNR, QHGQLWFPEGFK, EPLGEDSVGLKPLK, DIAQER, LLDEYNLVR, LAFETGPPR

- Bcl-XL

  EAGDEFLR, ELVVDFLSYK, GYSWSQFSDVEENR

- Mcl-1

  QSLEIISR, LLFAPTR

- NFκB1

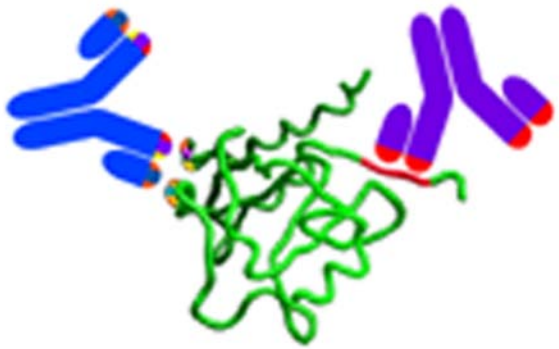
  VLSILLK, QFAIVFK, NTVNLFGK, QQIDELR, VFETLEAR, AGADLSLLDR, VIVQLVTNGK, LGNSVLHLAAK, LGLGILNNAFR, QEDVVEDLLR, LEPVVSDAIYDSK, EESAGQVDNLFLEK, HANALFDYAVTGDVK, TTSQAHSLPLSPASTR, NDLYQTPLHLAVITK

Data provided by Dr. John Koomen, Moffitt Cancer Center

mAb for Every Human Protein

ORIGENE Technologies

TrueMAB™
Antibodies against authentic antigens

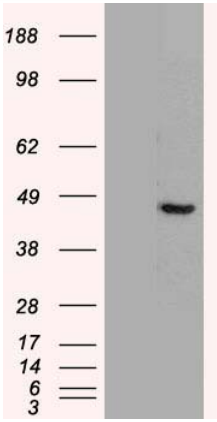


- **Dedicated mAb center**
  - Capacity: 200 immunizations per month
- **Authentic proteins as antigens**
  - Full length proteins made in human cells
- **Extensive validation**
  - WB: 9 different cell lines
  - IHC: 24 different tissues
  - IF
  - Flow
  - Luminex

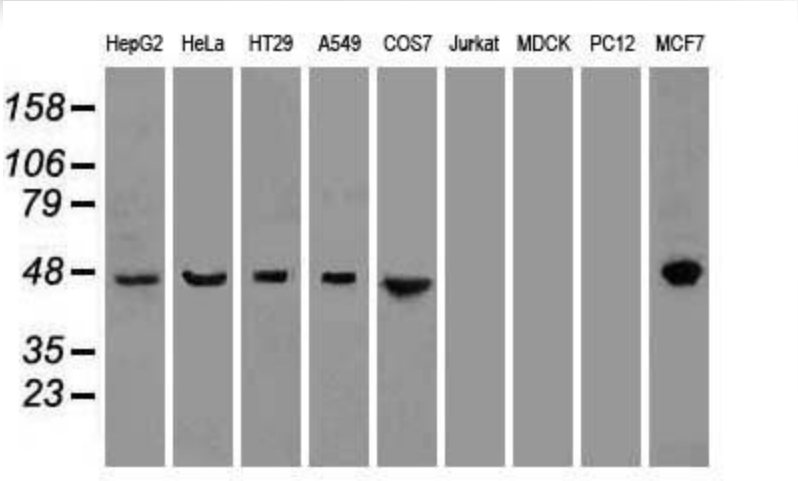
Goal: produce high quality mAb to every human proteins in 5-10 years

YOUR GENE COMPANY

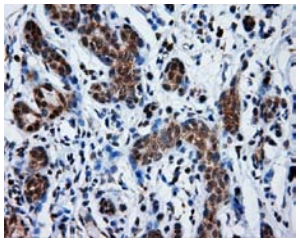
HTP Validation of mAbs



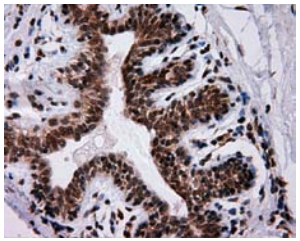
Anti-KRT WB with OriGene over expression lysate



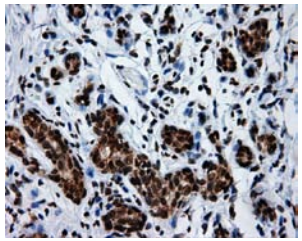
Anti-KRT WB with cell panel of 9 different cell lines (For endogenous protein detection in cell line)



ERCC1 (8F1)



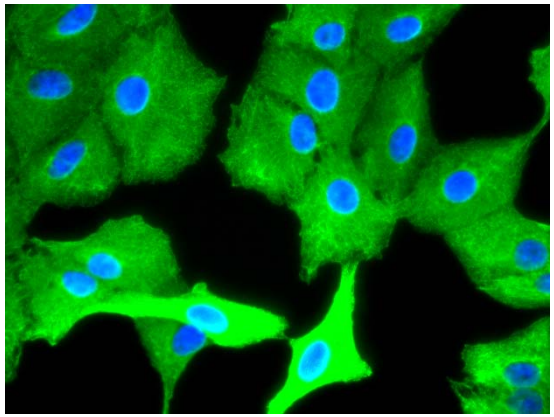
ERCC1 (1A3)



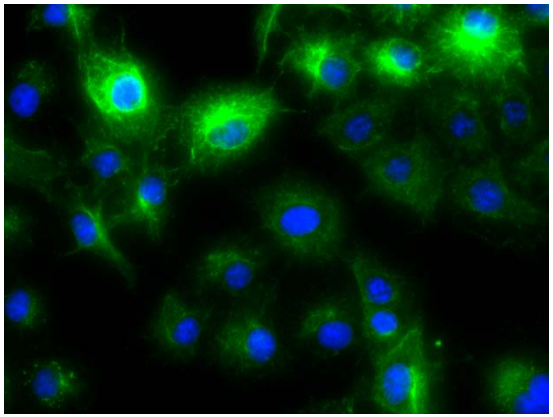
ERCC1 (4F9)

IHC data for human carcinoma liver tissue (12 IHC data from normal human tissue, and 12 carcinoma tissue IHC tests)

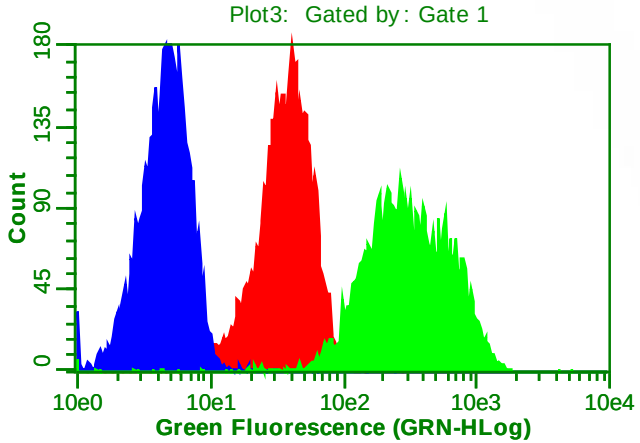
IF and Flow validation of mAbs



IF data with A549 cells
(For endogenous protein)



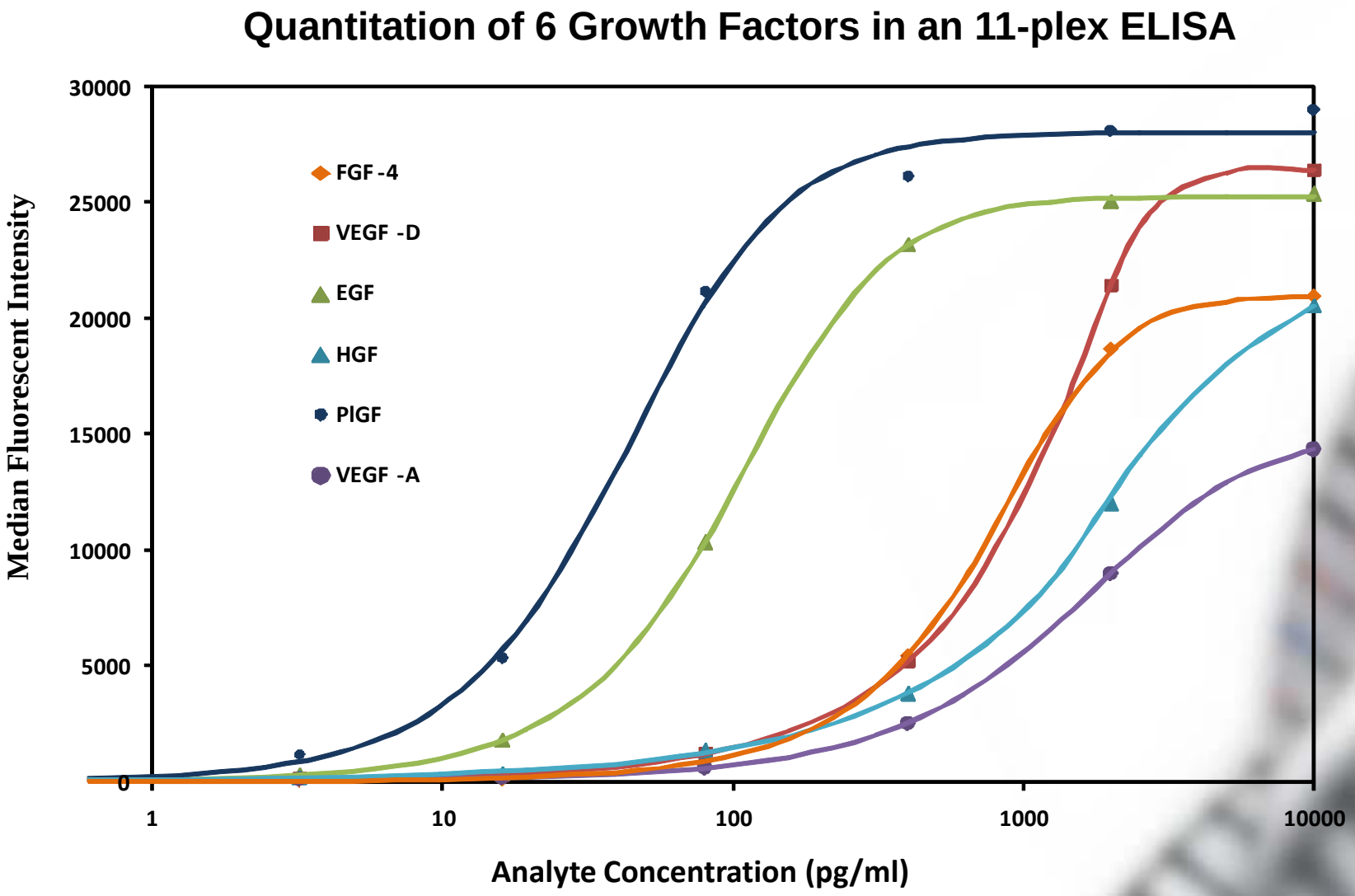
IF data with COS7 cells transiently transfected by RC clone



Flow data analysis for transfected COS7 cells
(Hybridism supernatant analysis)

Immunoassay Development

ORIGENE Technologies



Summary

- OriGene offers the most comprehensive proteins for functional studies and compound screening
  - 11,000 over-expression lysates
  - 5,000 purified human proteins

- Proteome wide assay technologies are coming of age. High density arrays and MRM-MS hold great promise in biomarker discovery and validation

www.origene.com

Walter Tian: wtian@origene.com