Identification of differentially expressed genes in human lung adenocarcinoma: ERGIC3 as a novel lung cancer-related gene

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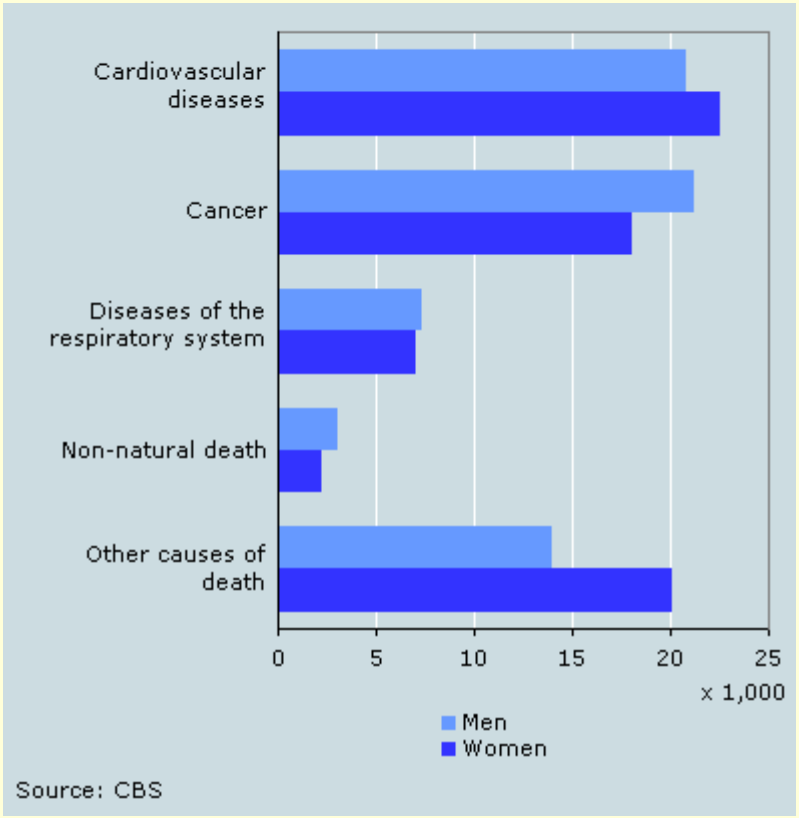
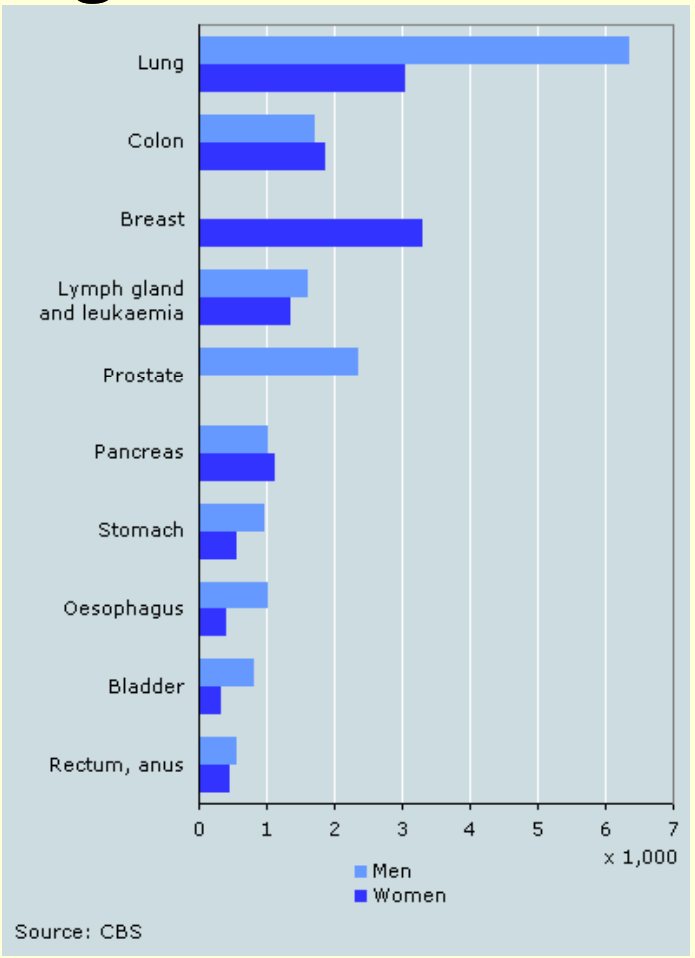
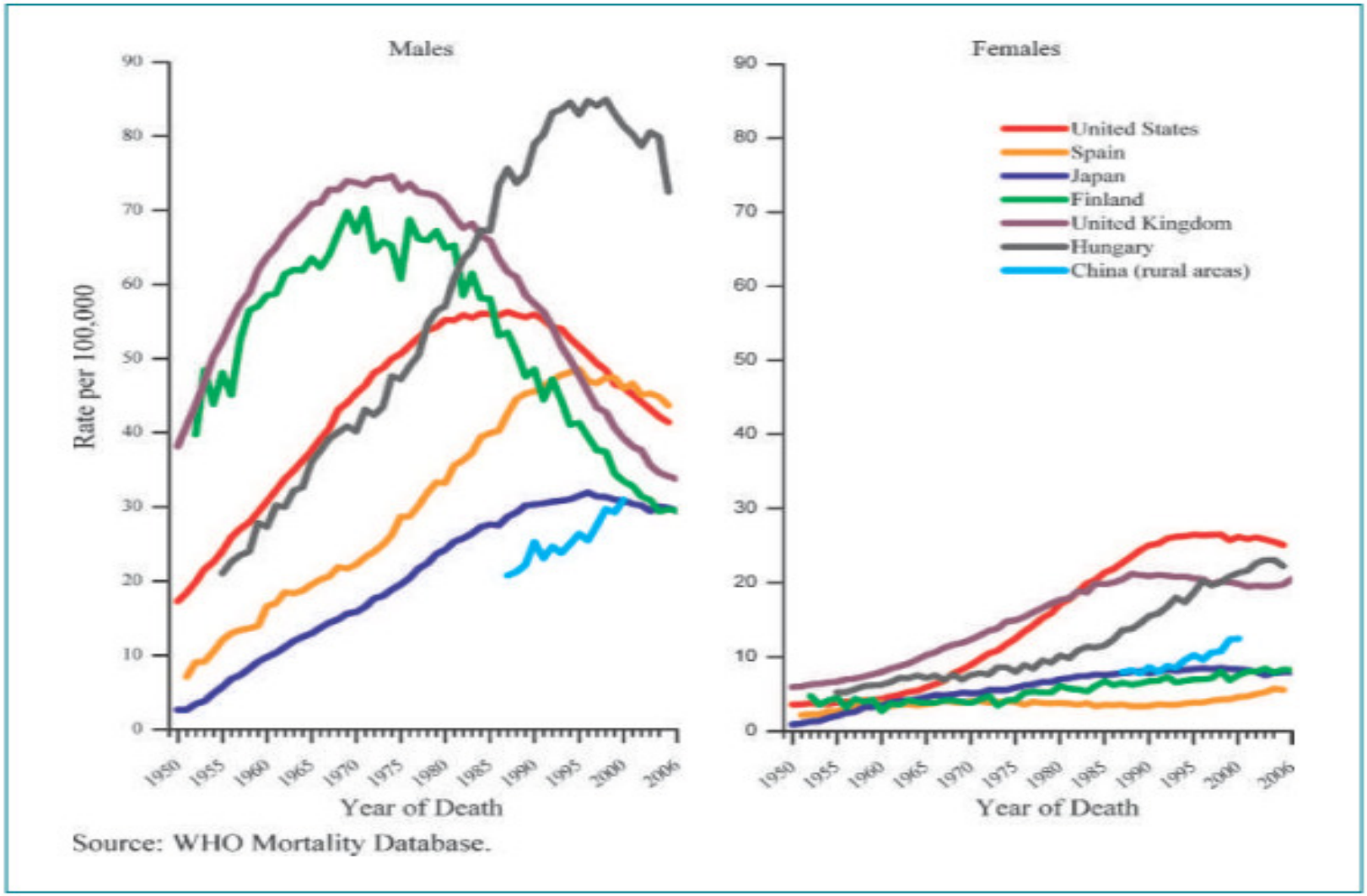
Part I 1、Background

1.1 epidemiology of lung cancer



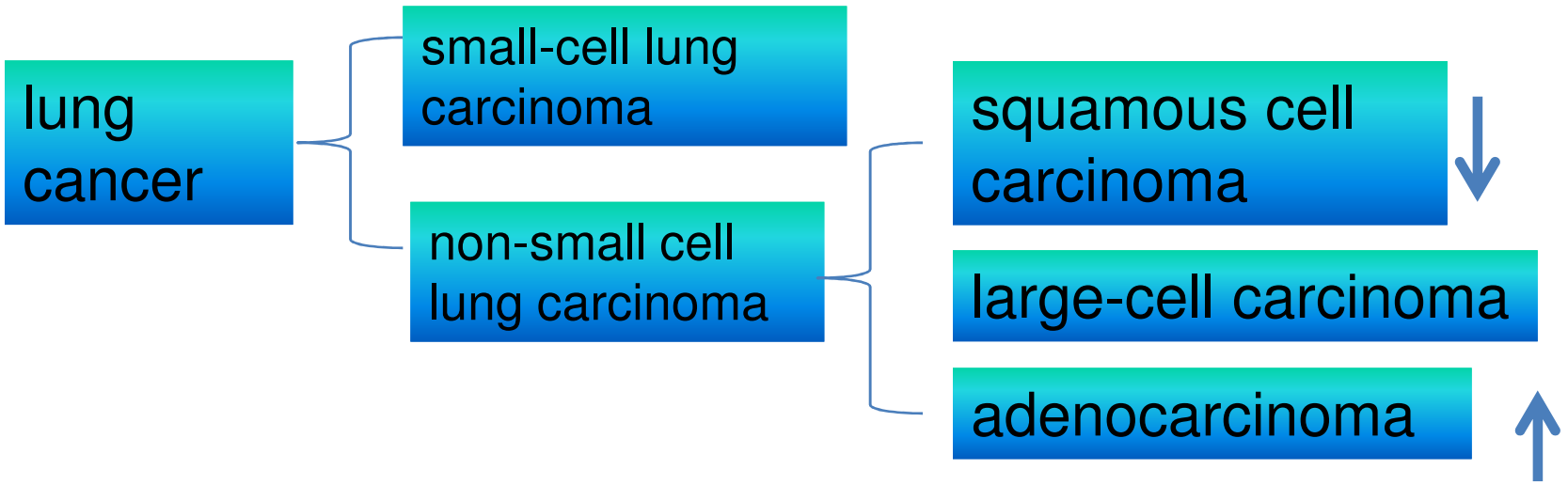


Mortality in lung cancer in selected country 1950—2006



Jemal et al.Cancer Epidemiology, Biomarkers & Prevention. (2010)

Subtype of lung cancer



New tumor marker of lung cancer
for early diagnosis and treatment

To investigate genetic mechanism
and
new target

1.2 Molecular basis of LC

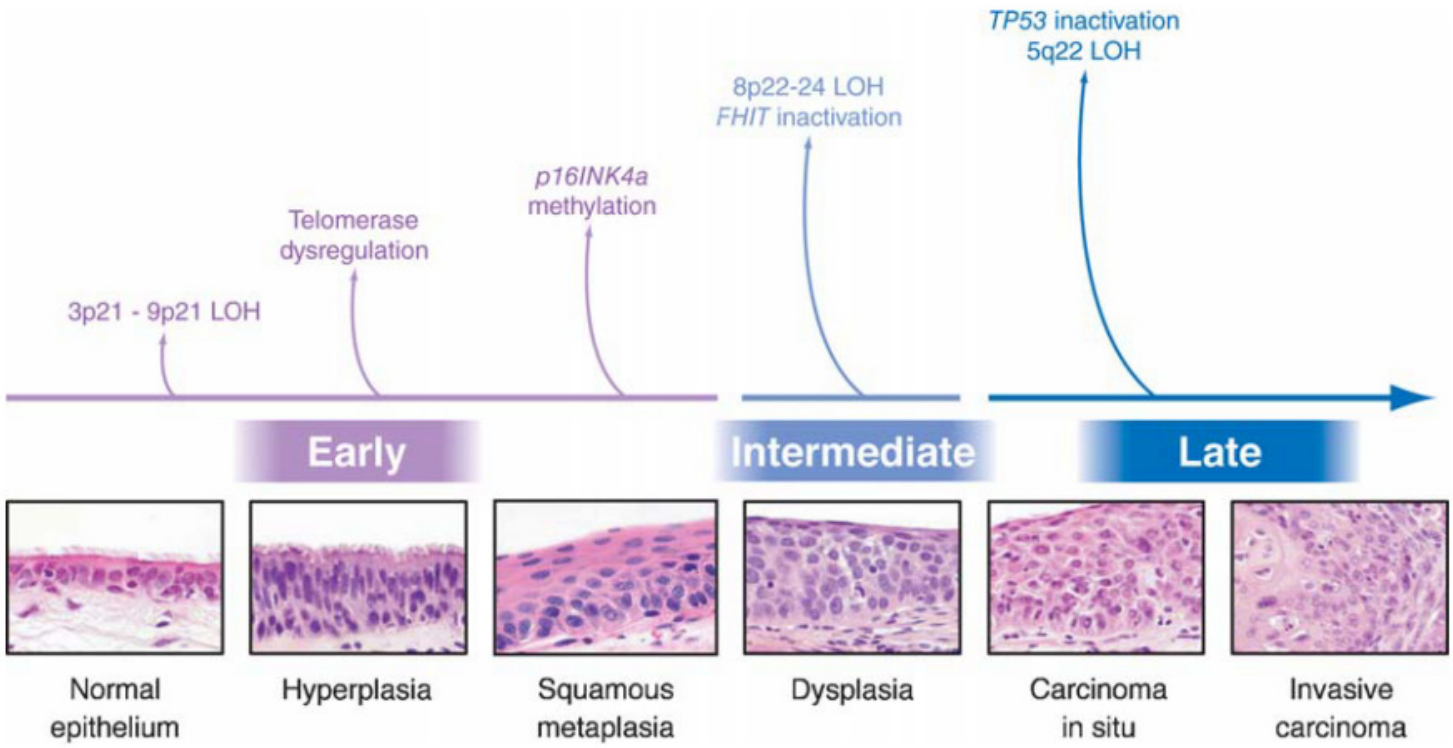


Figure 1

Histopathological and molecular changes during the pathogenesis of squamous cell carcinoma of the lung with molecular changes commencing at early stages, and a histologically normal respiratory epithelium. There is stepwise molecular and histopathological sequence of events leading to dysplastic and invasive carcinoma stages. LOH, loss of heterozygosity.

1.3 Suppression subtractive hybridization(SSH)

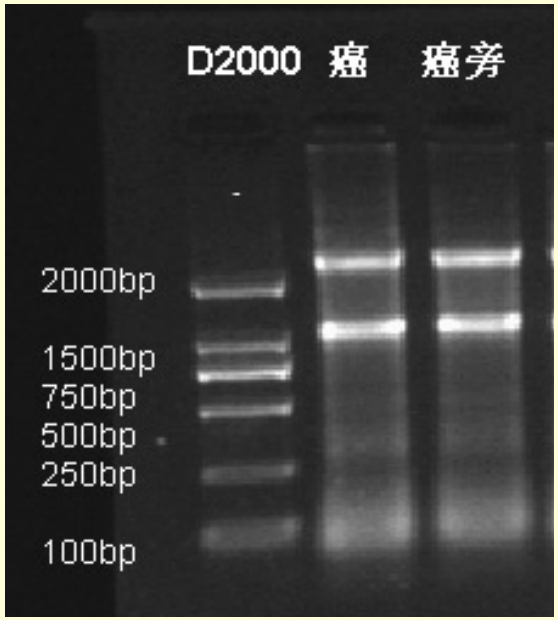
- cDNA Microarray
- mRNA Differential display，DD
- Representational diff-erence analysis，RDA
- Suppression subtractive hybridization, SSH

lower false positive,higher specific & sensitive

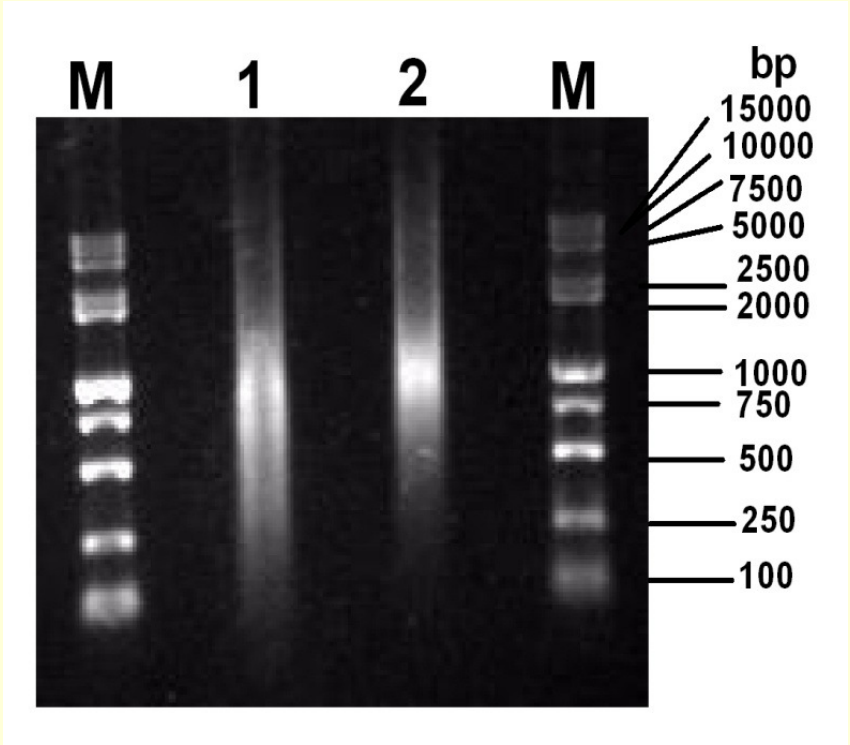
2、Results

2.1 Construction of AC-associated l

2.1.1 Total RNA & mRNA

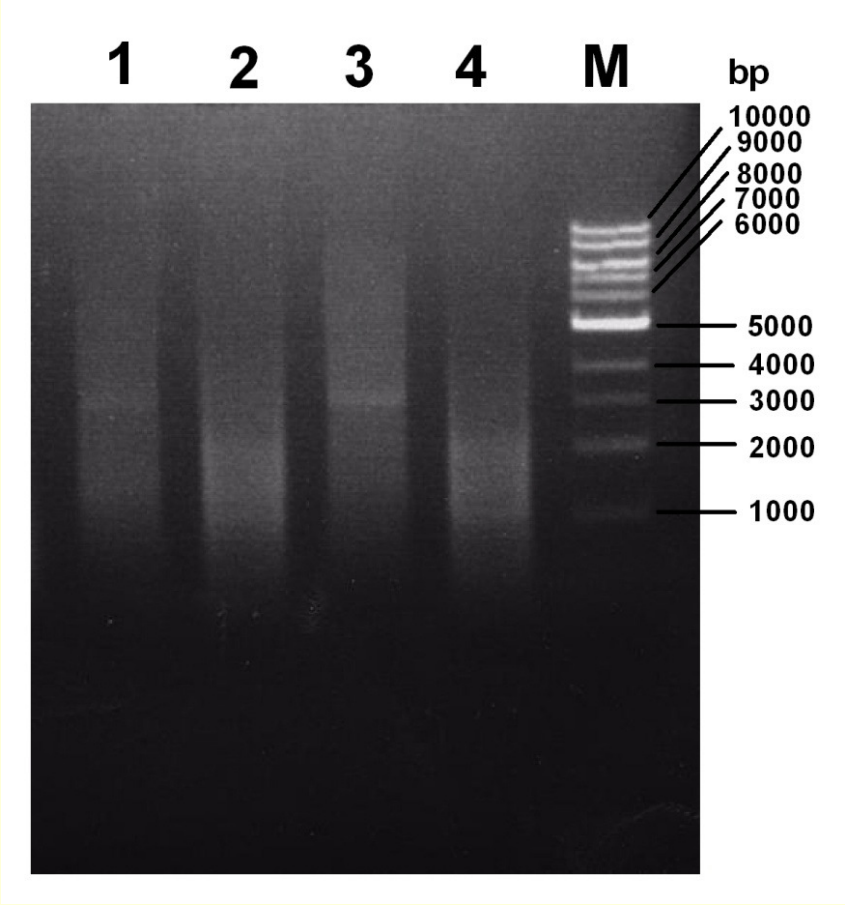


total RNA



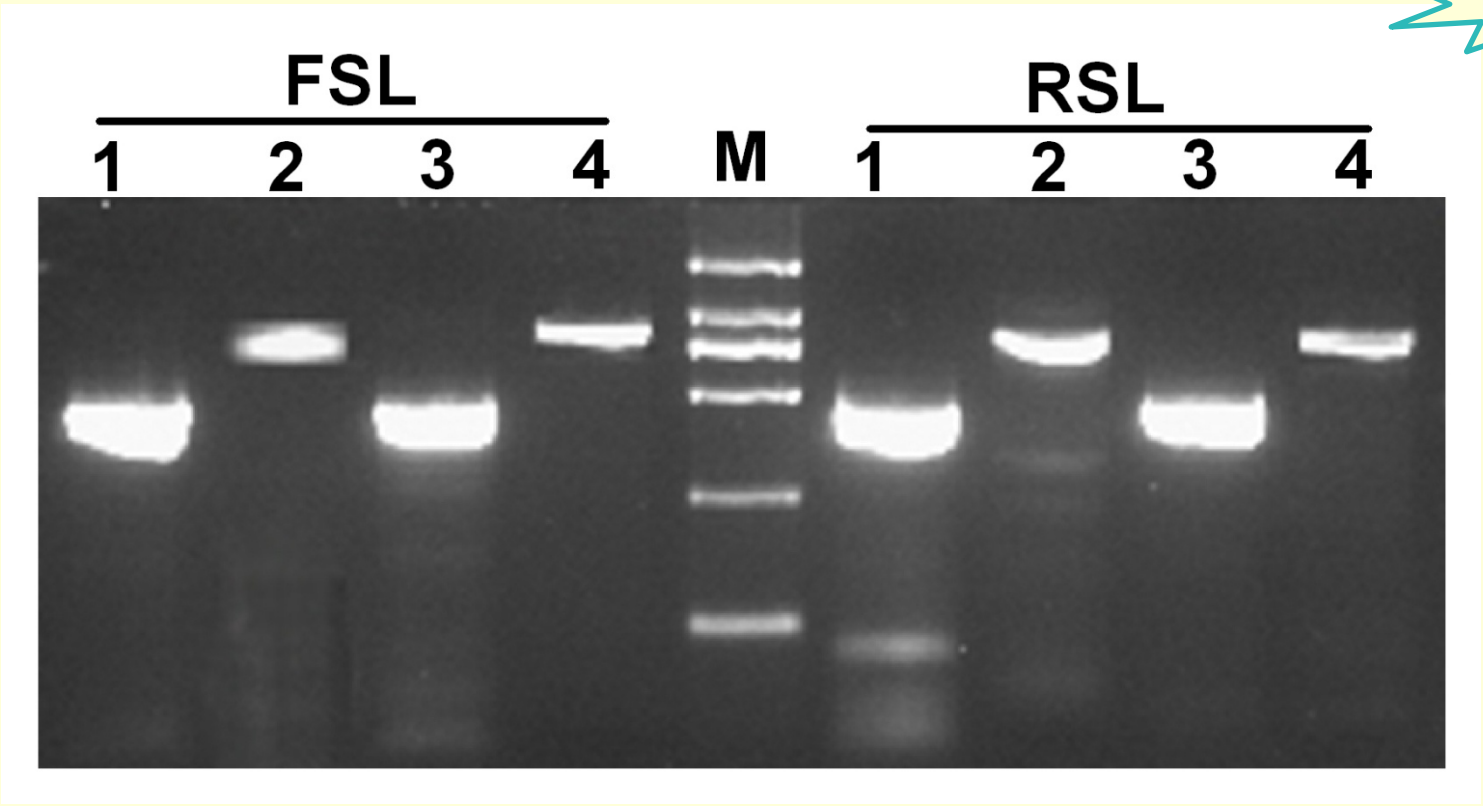
mRNA

2. 1. 2 **Analysis of Rsa I digestion**

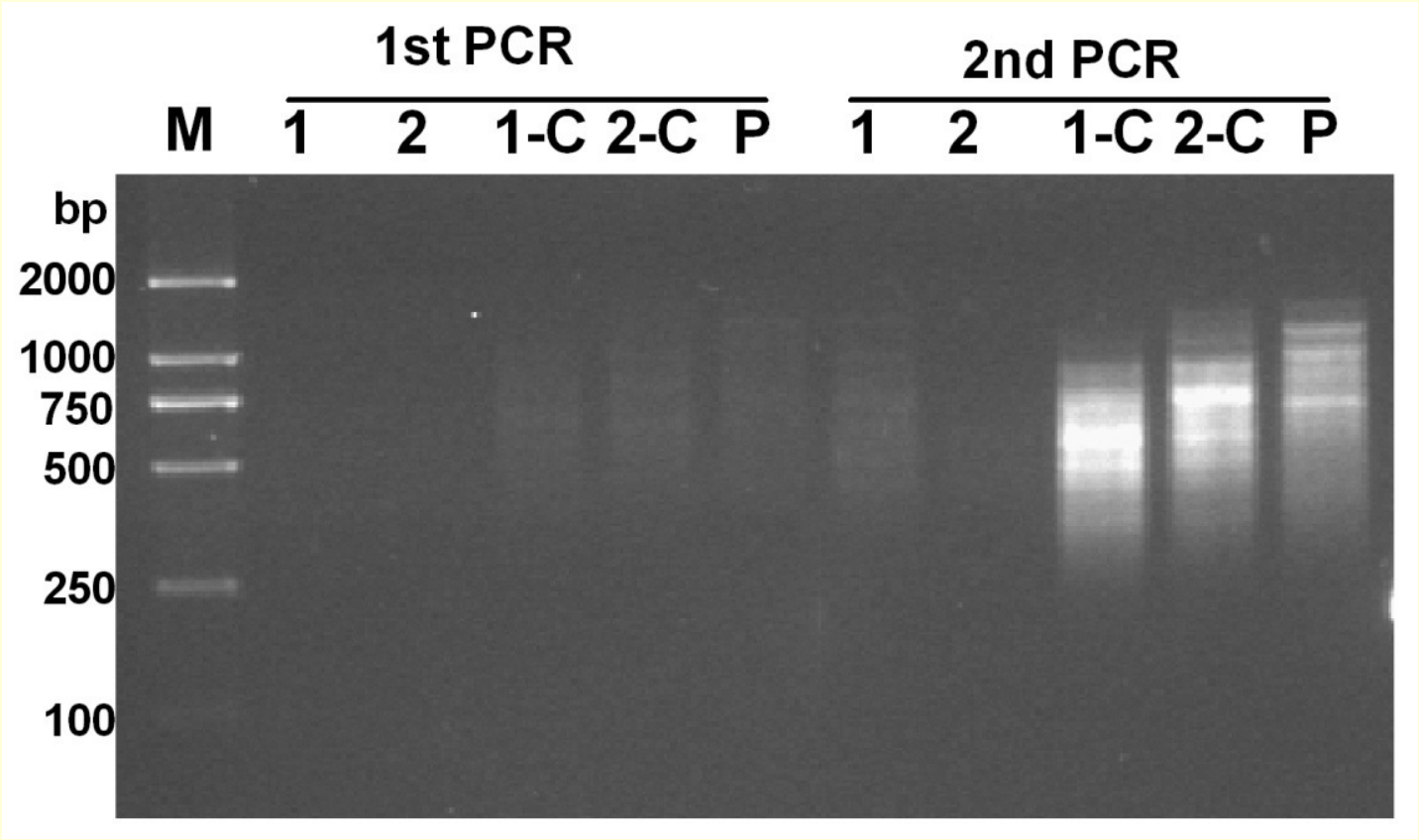


digestion of double cDNA

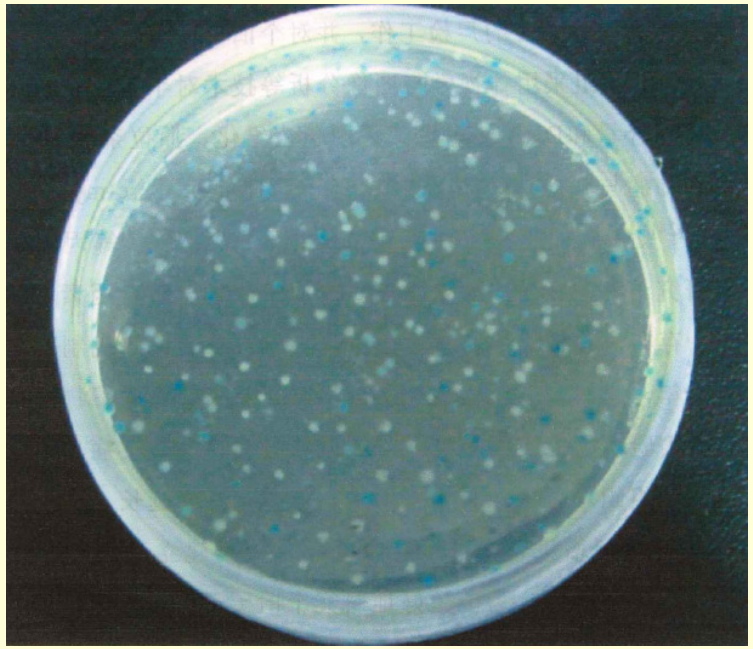
2.1.3 Ligation efficiency analysis

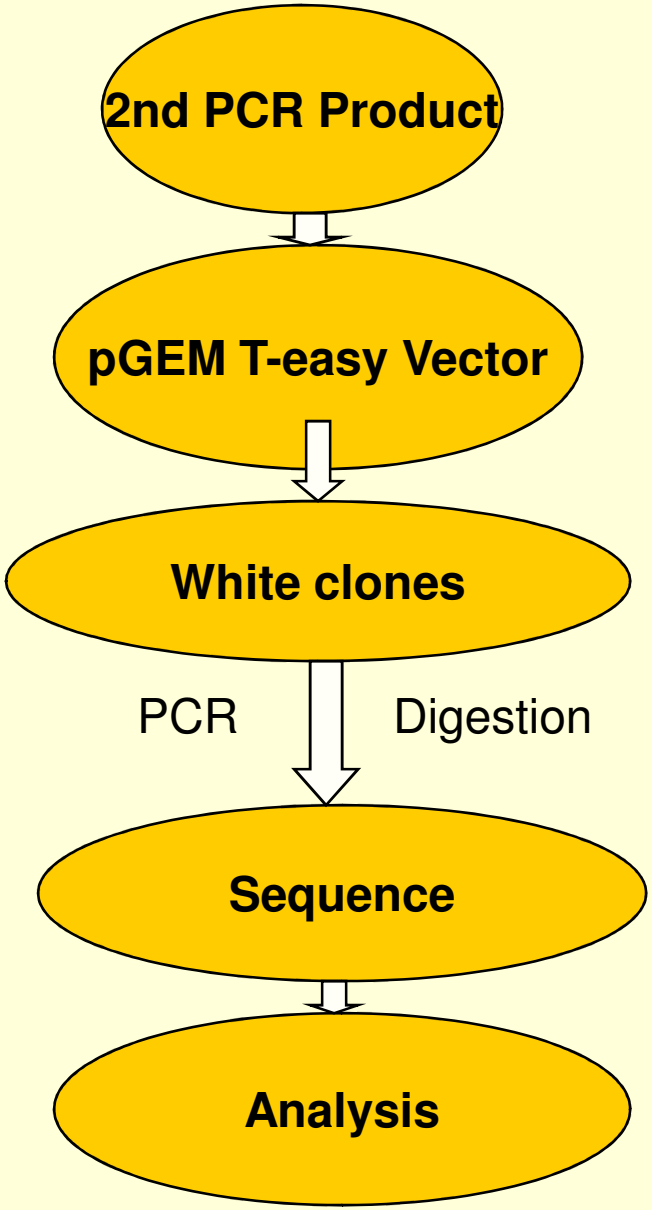


2.1.4 Supression PCR



2.2 Clone of EST





FSL: 485 up-regulated

RSL: 172 down-regulated

2. 3 **Bioinformatic analysis**

2.3.1 BLAST

表 1. 正向杂交文库中差异表达基因的定义、染色体座位和重复的克隆数

Accession	Definition	Cytoband	Recovery
XM_002345305	Homo sapiens similar to DC48 (LOC100293563)		
NM_002790	proteasome (prosome, macropain) subunit, alpha type, 5	1p13	7

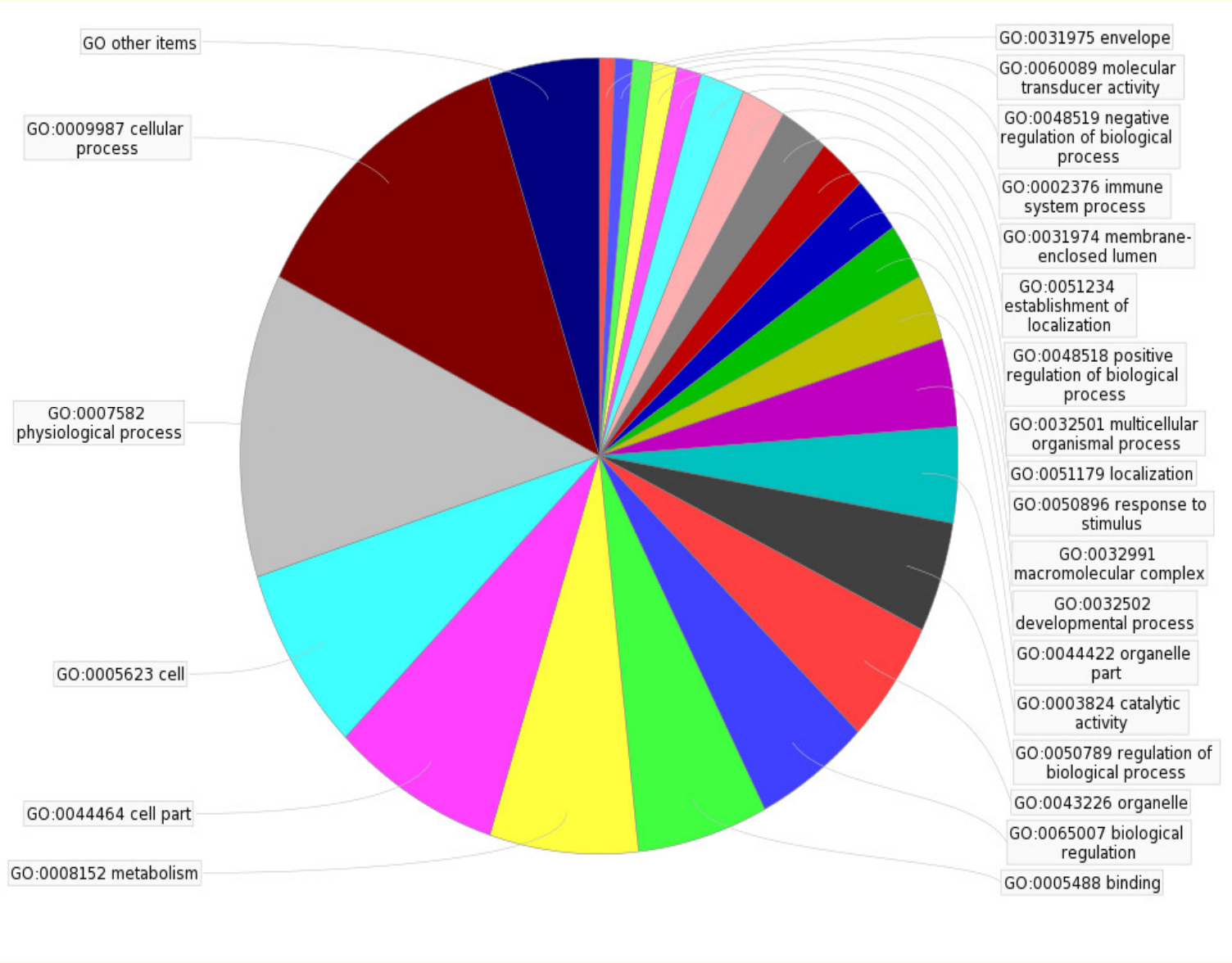
Gene：177（new：152）
Unknown EST: 44

表 2. 反向杂交文库中差异表达基因的定义、染色体座位和重复的克隆数

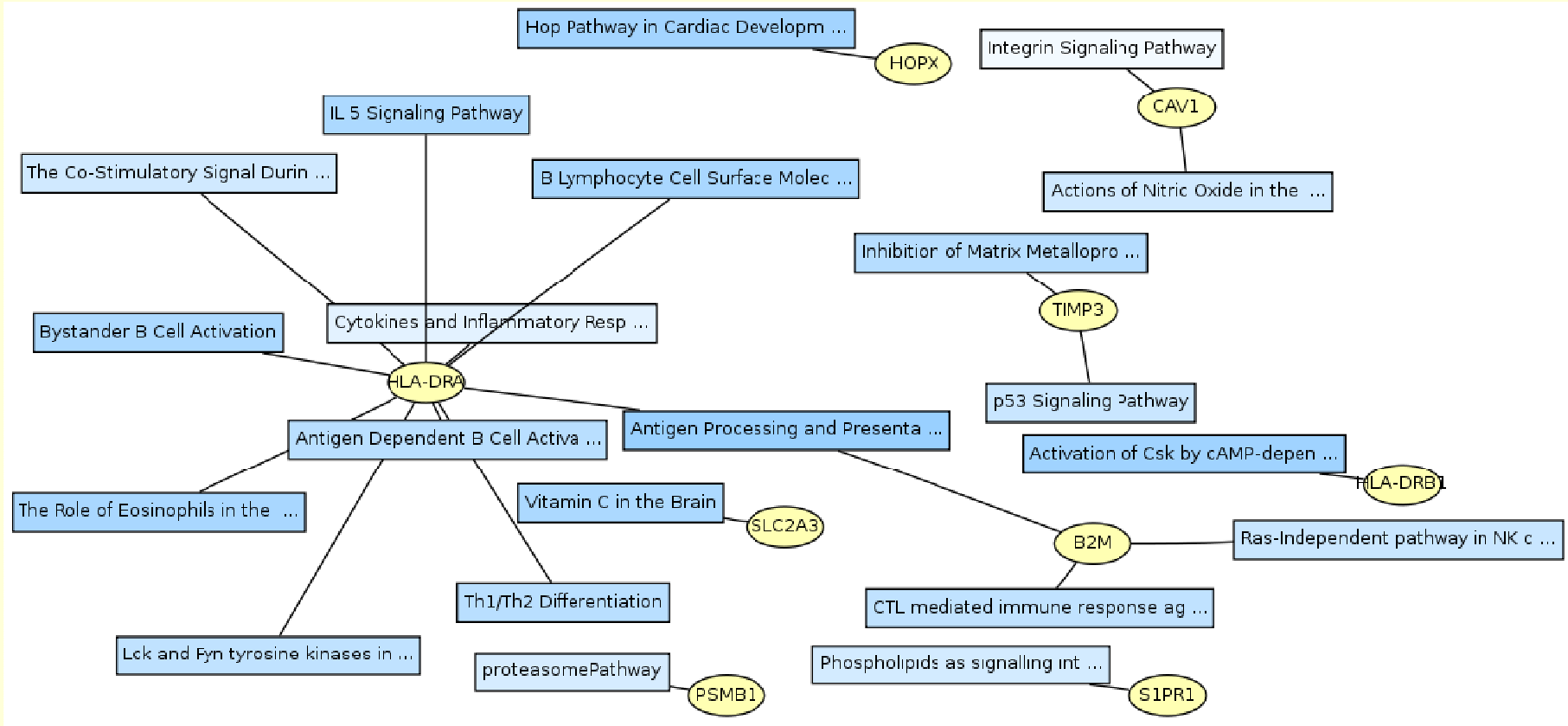
Accession	Definition	Cytoband	Recovery
NM_001400	sphingosine-1-phosphate receptor 1 (S1PR1)		
NM_002038	interferon, alpha-inducible protein 6 (IFI6)		
NM_173452	ficolin (collagen/fibrinogen domain containing) 3 (Hakata antigen) (FCN3)		
NM_014624	S100 calcium binding protein A6 (S100A6)	1q21	1
NM_001043353	tropomyosin 3 (TPM3)	1q21.2	9
NM_139119	YY1 associated protein 1 (YY1AP1)	1q22	1
NM_148964	cathepsin E (CTSE)	1q31	2
NM_020216	arginyl aminopeptidase (aminopeptidase B) (RNPEP)	1q32	1
NM_014713	lysosomal protein transmembrane 4 alpha (LAPTM4A)	2p24.1	1
NM_207328	hypothetical protein LOC150763	2q11.1	1
NM_004657	serum deprivation response (phosphatidylserine binding protein) (SDPR)	2q32-q33	1
NM_032495	HOP homeobox (HOPX), transcript variant 1	4q11-q12	2
NM_001042734	SEC24 family, member B (S. cerevisiae) (SEC24B)	4q25	1

Gene：59（new：54）
Unknown EST: 10

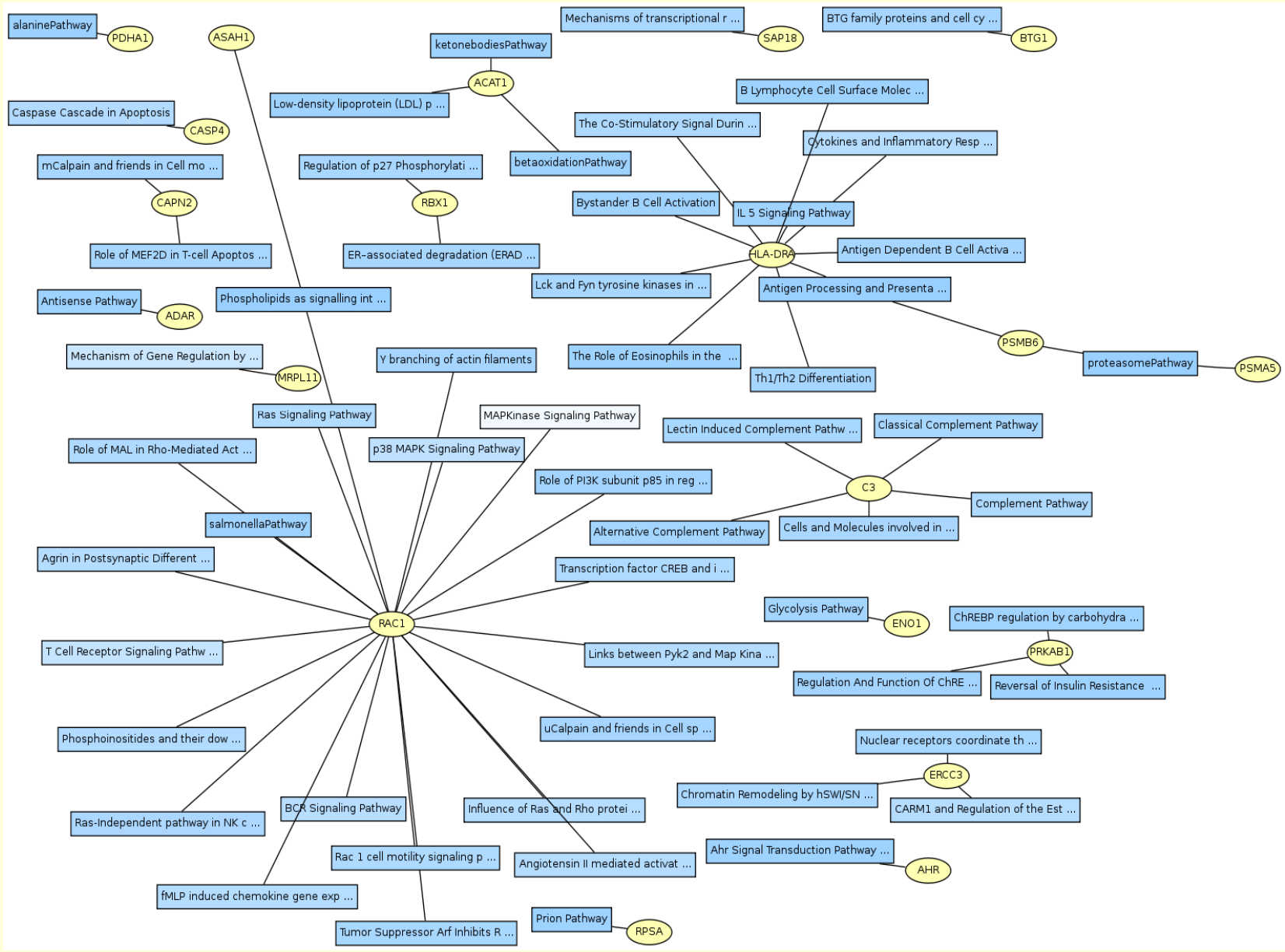
2. 3. 2 GO Ontology



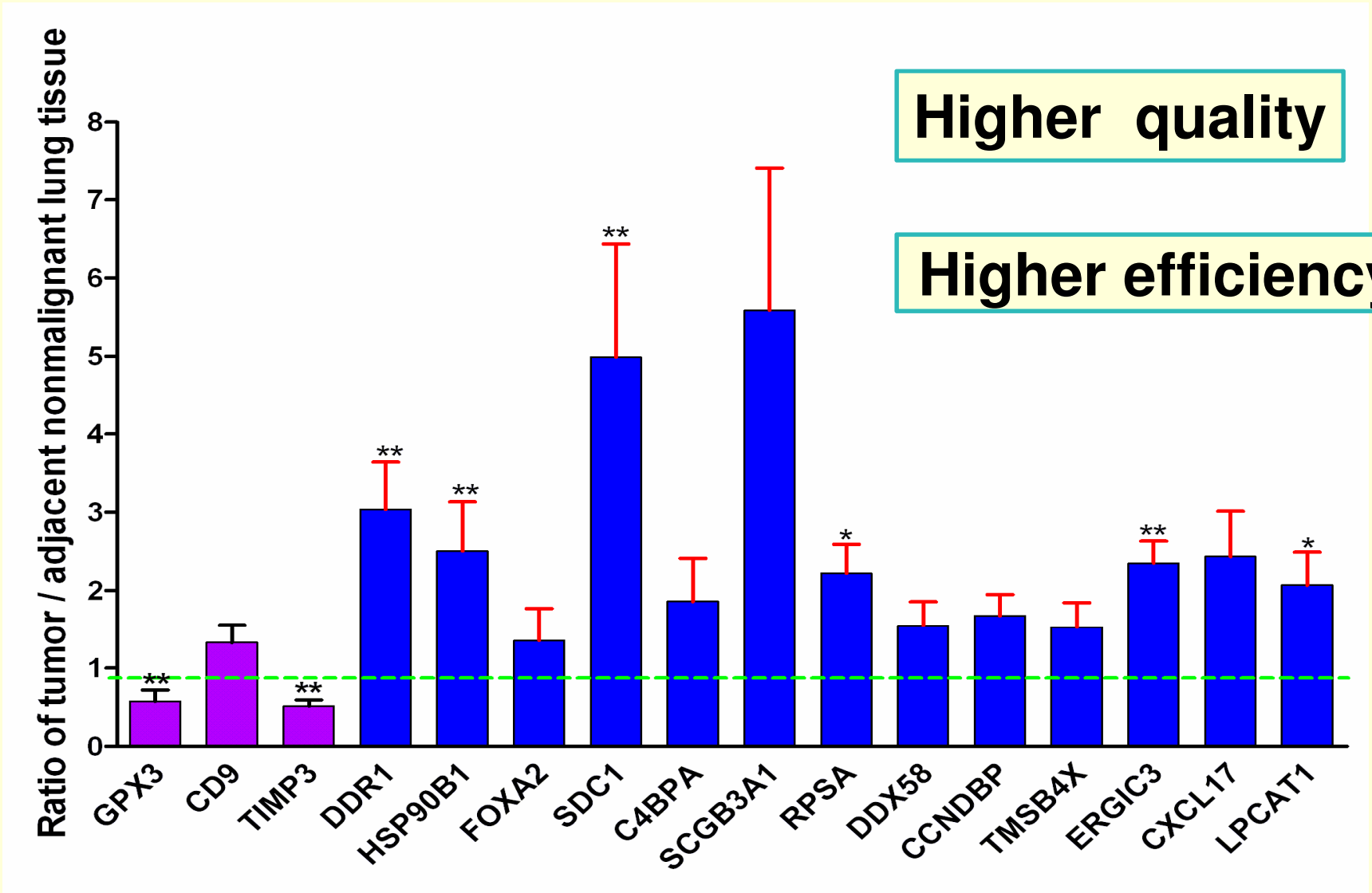
2. 3. 3 **Biocatar pathway** RSL



2. 3. 4 Biocatar pathway FSL

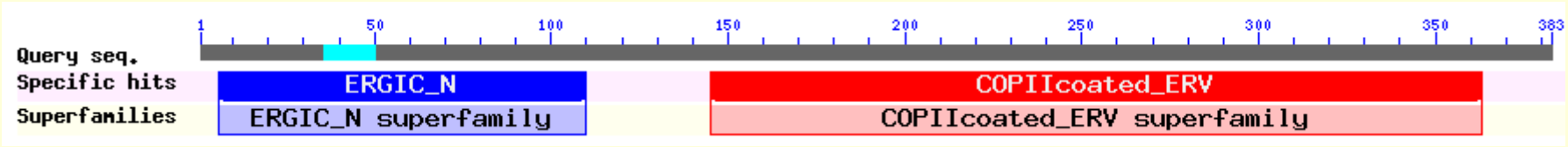


2.4 Sreen of SSH-libraries

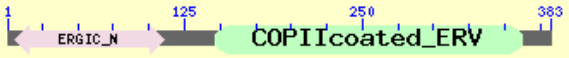


Part II 1、Background

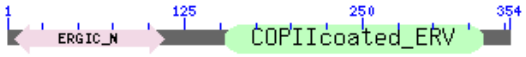
- (Endoplasmic reticulum-Golgi intermediate compartment 3，ERGIC3)
- Cycles between the endoplasmic reticulum and Golgi
- II type transmembrane protein, 383 aa, 43.2 kDa



[Query] NP_057050.1
endoplasmic reticulum-Golgi intermediate compartment protein 3
Total architectures: 8

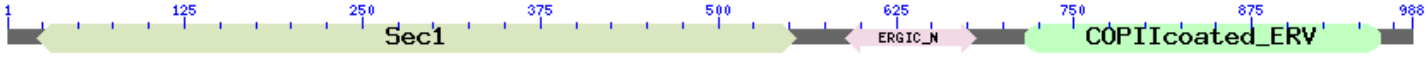


[+] Endoplasmic reticulum vesicle
taxonomy span: Eukaryota
Similarity score: 2
Total nr sequences: 858



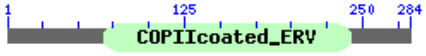
[+] protein PDI-like 5-4
taxonomy span: Eukaryota
Similarity score: 2
Total nr sequences: 63

[+] hypothetical protein
taxonomy span: Agaricomycetidae
Similarity score: 2
Total nr sequences: 3



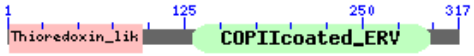
[+] pyruvate dehydrogenase (acetyl-
taxonomy span: Trichosporon asahii var. asahii
Similarity score: 2
Total nr sequences: 2

[+] unnamed protein product
taxonomy span: cellular organisms
Similarity score: 1
Total nr sequences: 83

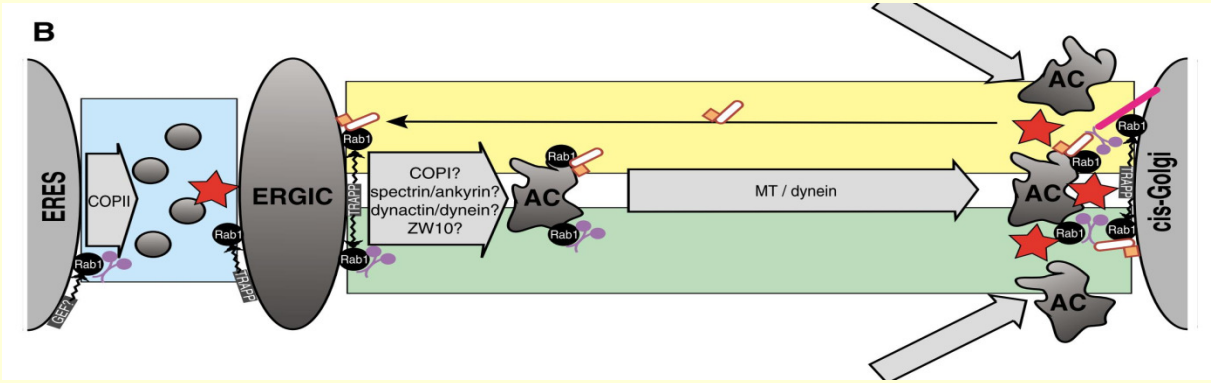


[+] hypothetical protein
taxonomy span: Eukaryota
Similarity score: 1
Total nr sequences: 26

[+] unknown
taxonomy span: Eukaryota
Similarity score: 1
Total nr sequences: 4



[+] hypothetical protein
taxonomy span: Chlamydomonas reinhardtii
Similarity score: 1
Total nr sequences: 1



Herzog， et al. 2006

- Mutation: COPII ↓ 25%

  Accumulation of protein and lipid in ER

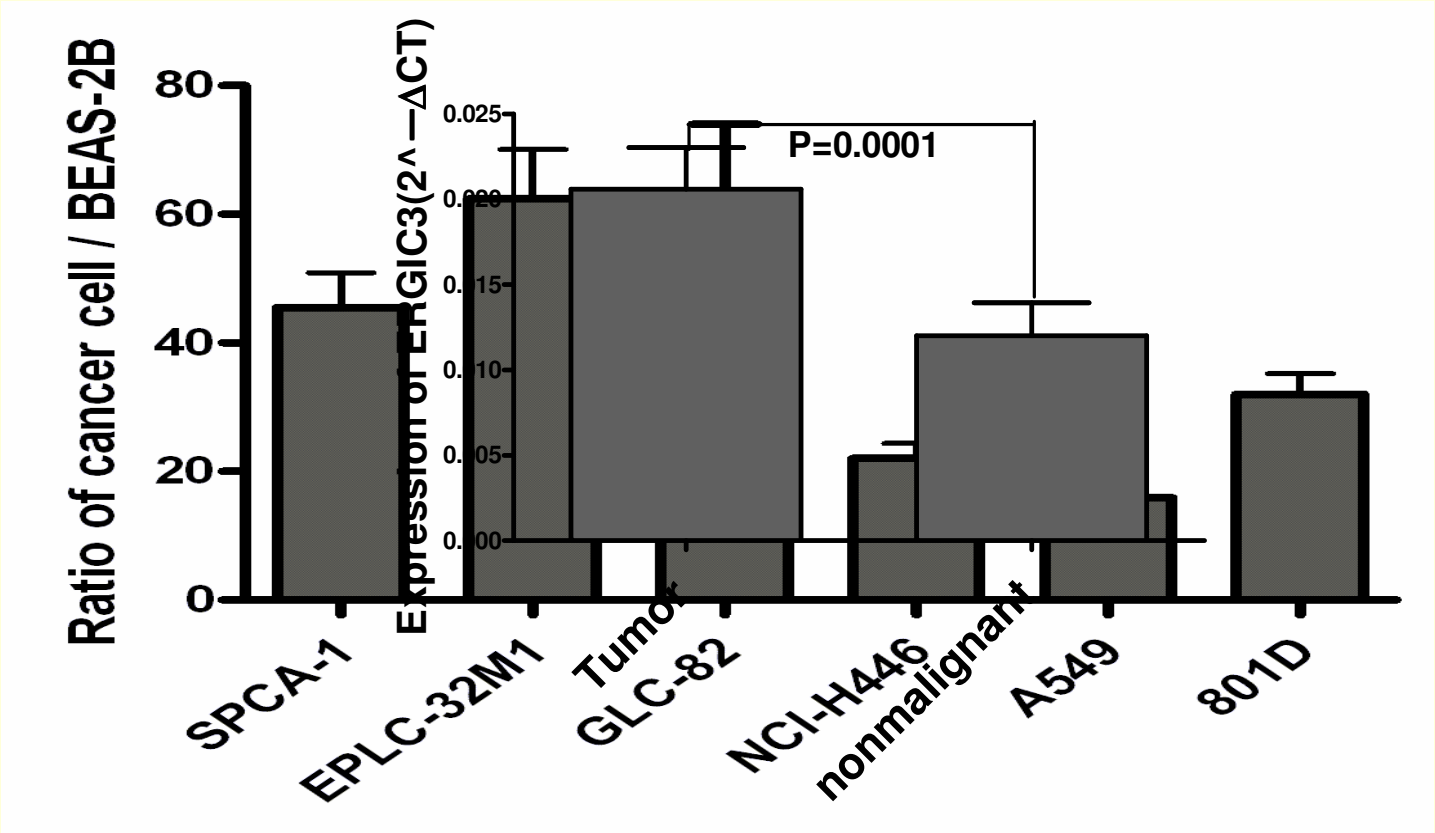
- Inhibits ER stress-induced cell death

  - **FUNCTION?**
  - **TUMOR MARKER**

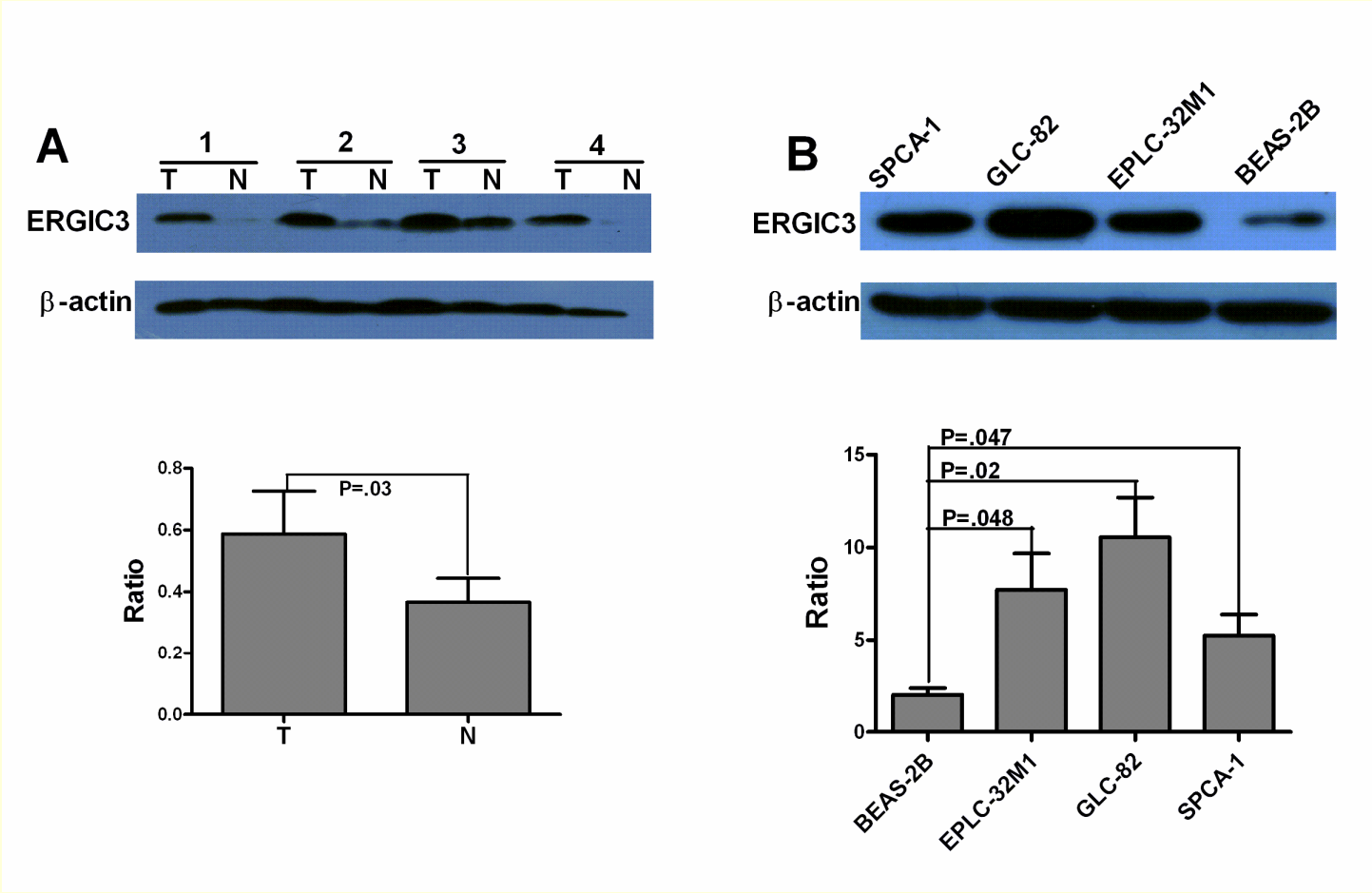
2、RESULTS

2.1 Expression of ERGIC3 in NSCLC

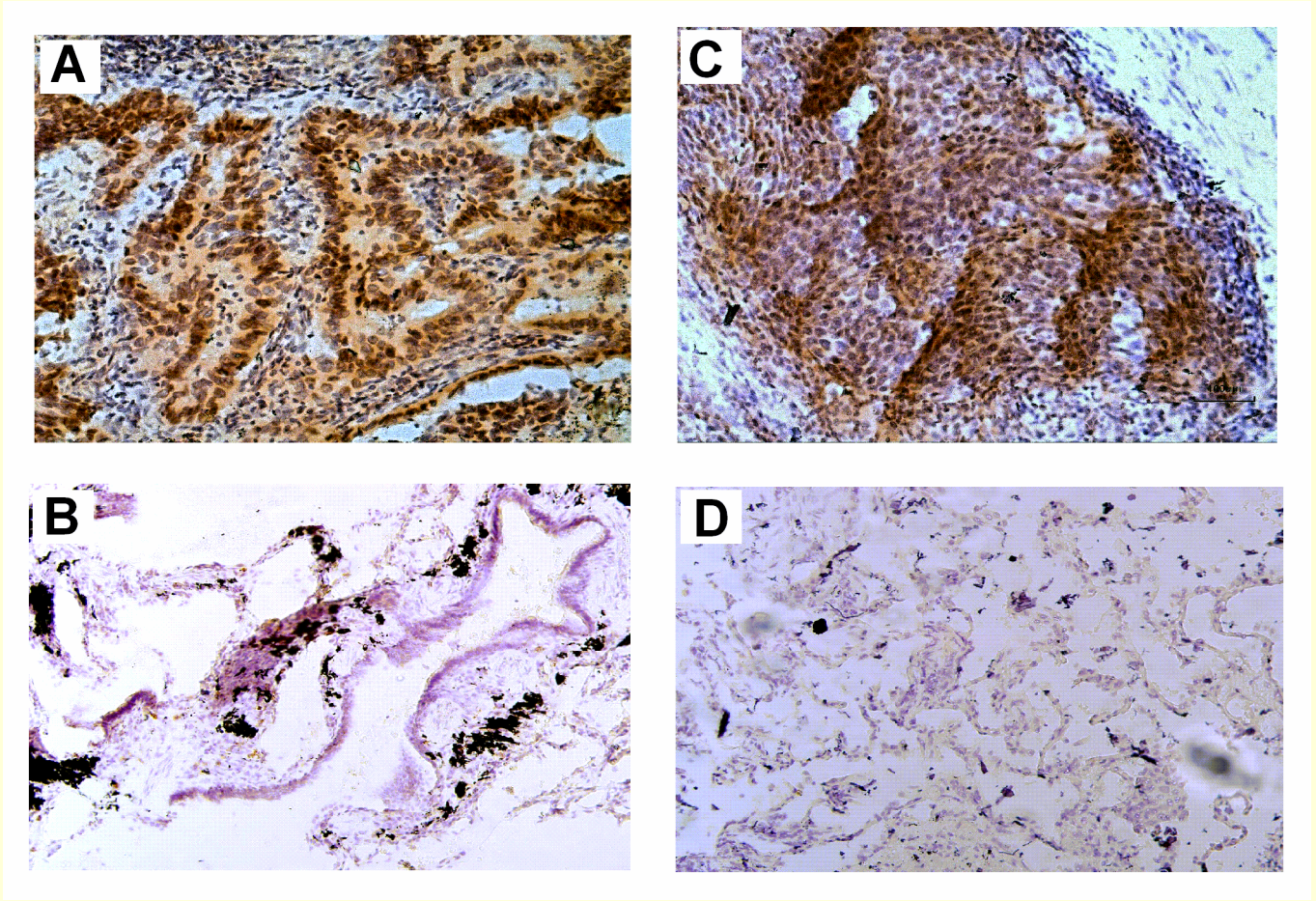
2.1.1 mRNA of ERGIC3



2.1.2 Protein of ERGIC3



2.1.3 Protein of ERGIC3



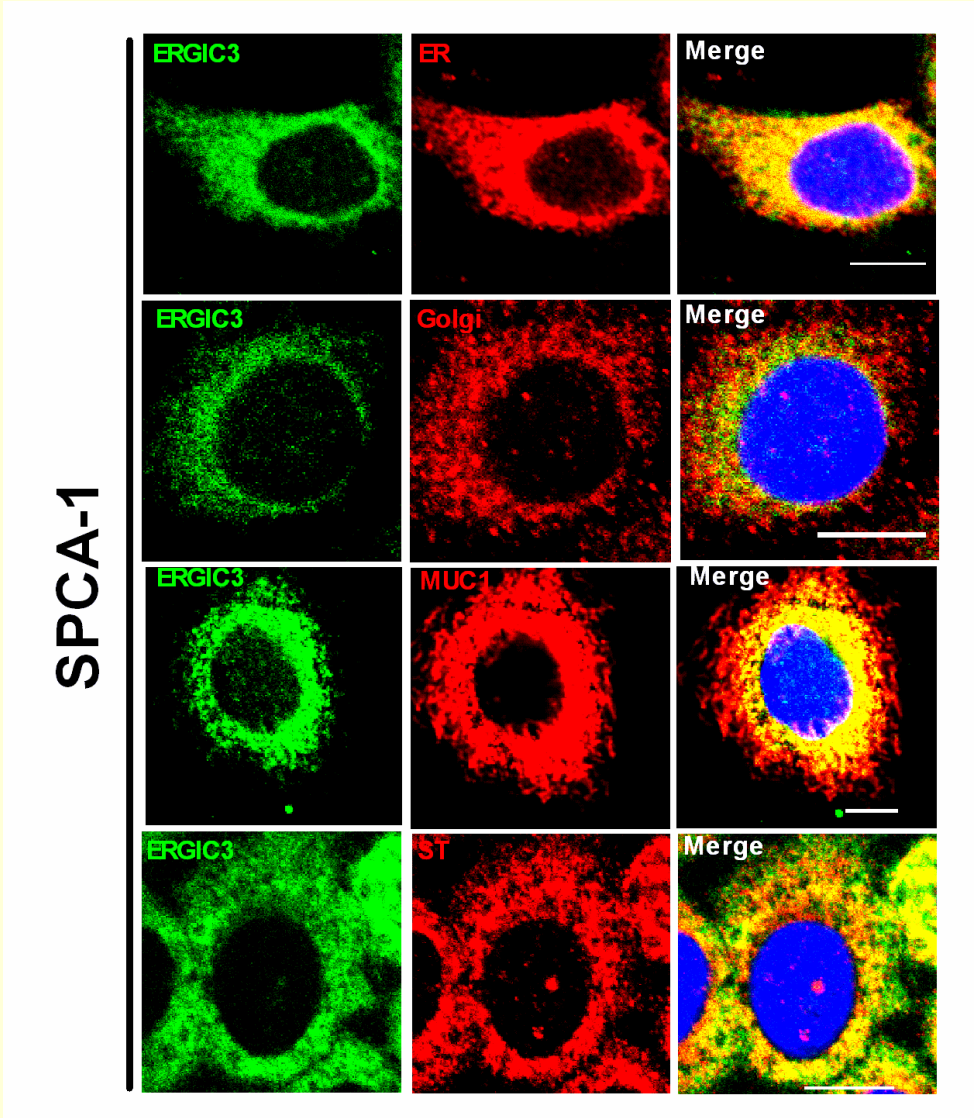
Evaluation of immunohistochemical positivity in lung cancer tissues

Tumour and clinicopathological features	Score			
	–	+	++	+++
Gender				
Male	3/17(17.6)	5/17(29.4)	4/17(33.5)	5/17(29.4)
Female	1/18(5.6)	4/18(22.2)	5/18(27.8)	8/18(44.4)
Histological type*				
AC	0	6/22(27.3)	7/22(31.8)	9/22(40.9)
SCC	4/13(30.8)	3/13(23.1)	2/13(15.4)	4/13(30.8)
TNM stage				
I + II	3/20(15.0)	5/20(25.0)	7/20(35.0)	5/20(25.0)
III + IV	1/15(6.7)	4/15(26.7)	2/15(13.3)	8/15(53.3)
Differentiation*				
Poor	4/11(36.4) *	2/11(18.2) *	1/11(9.1) *	4/11(36.4) *
Well or Moderate	0	7/24(29.2) *	8/24(33.3) *	9/24(37.5) *
Smoking				
No	1/17(5.9)	4/17(23.5)	4/17(23.5)	8/17(47.1)
Yes	3/18(16.7)	5/18(27.8)	5/18(27.8)	5/18(27.8)

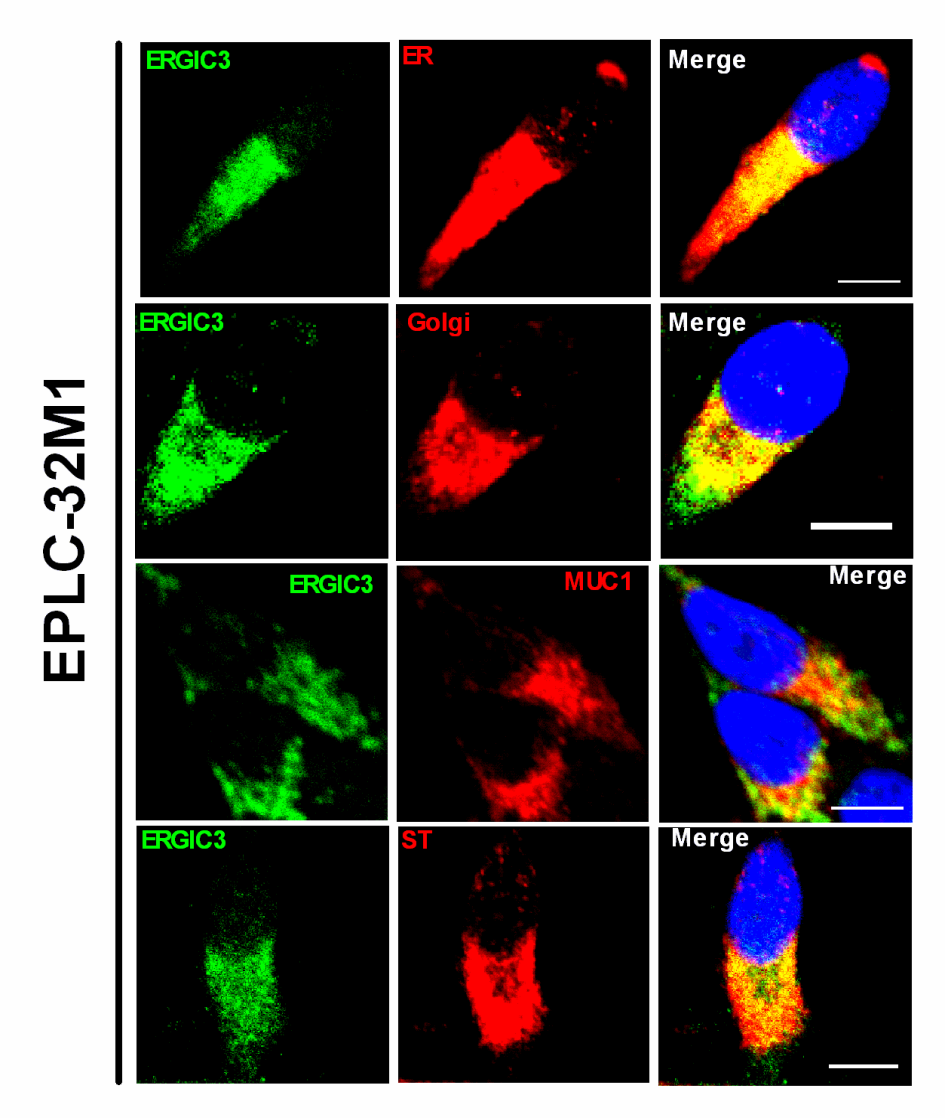
Positive rate:88.57%
AC>SCC

Score: −, no staining; +, <30%; ++, 31–60%; +++, >60% positive cells

2.1.4 Location of ERGIC3

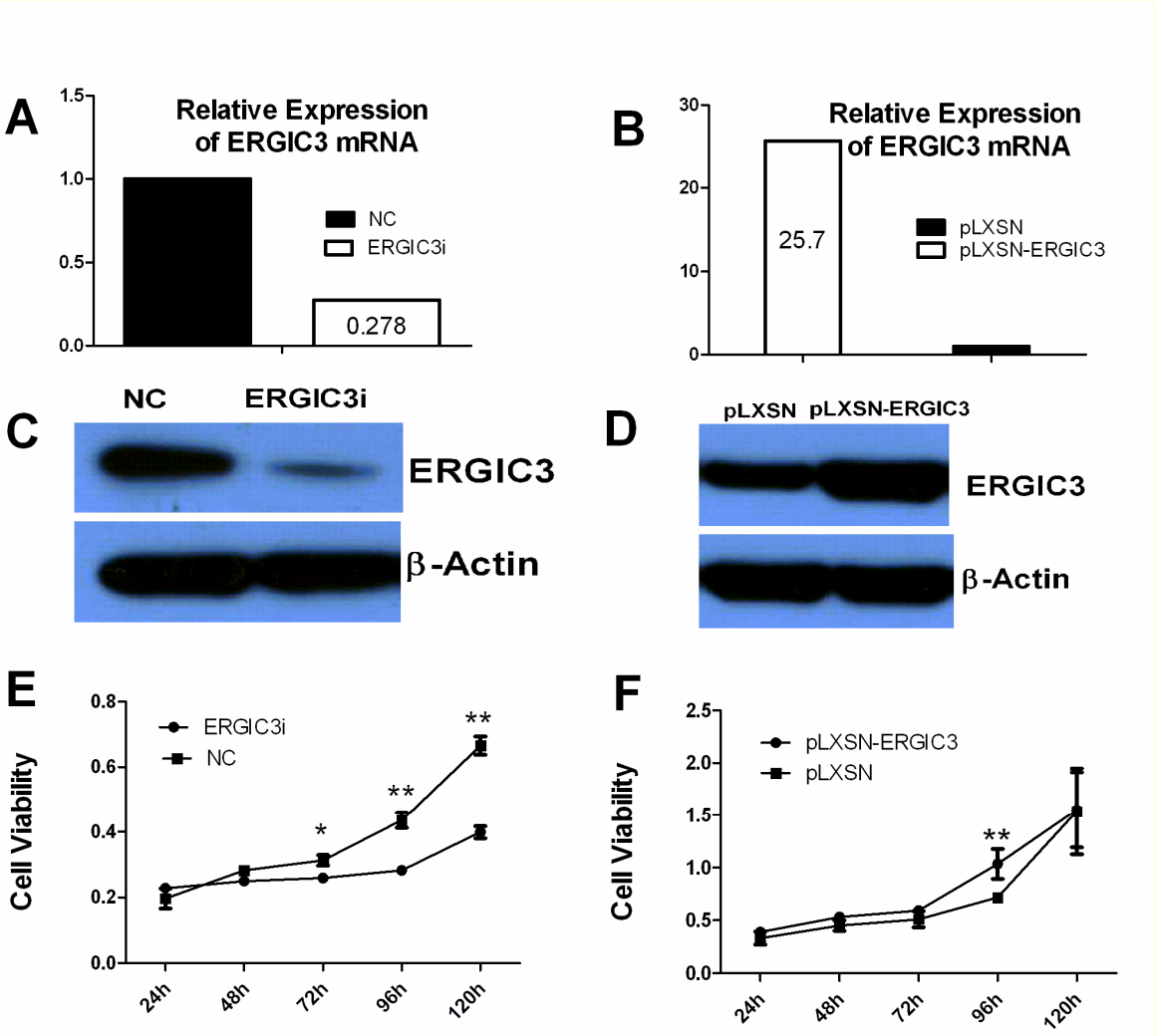


2.1.4 Location of ERGIC3

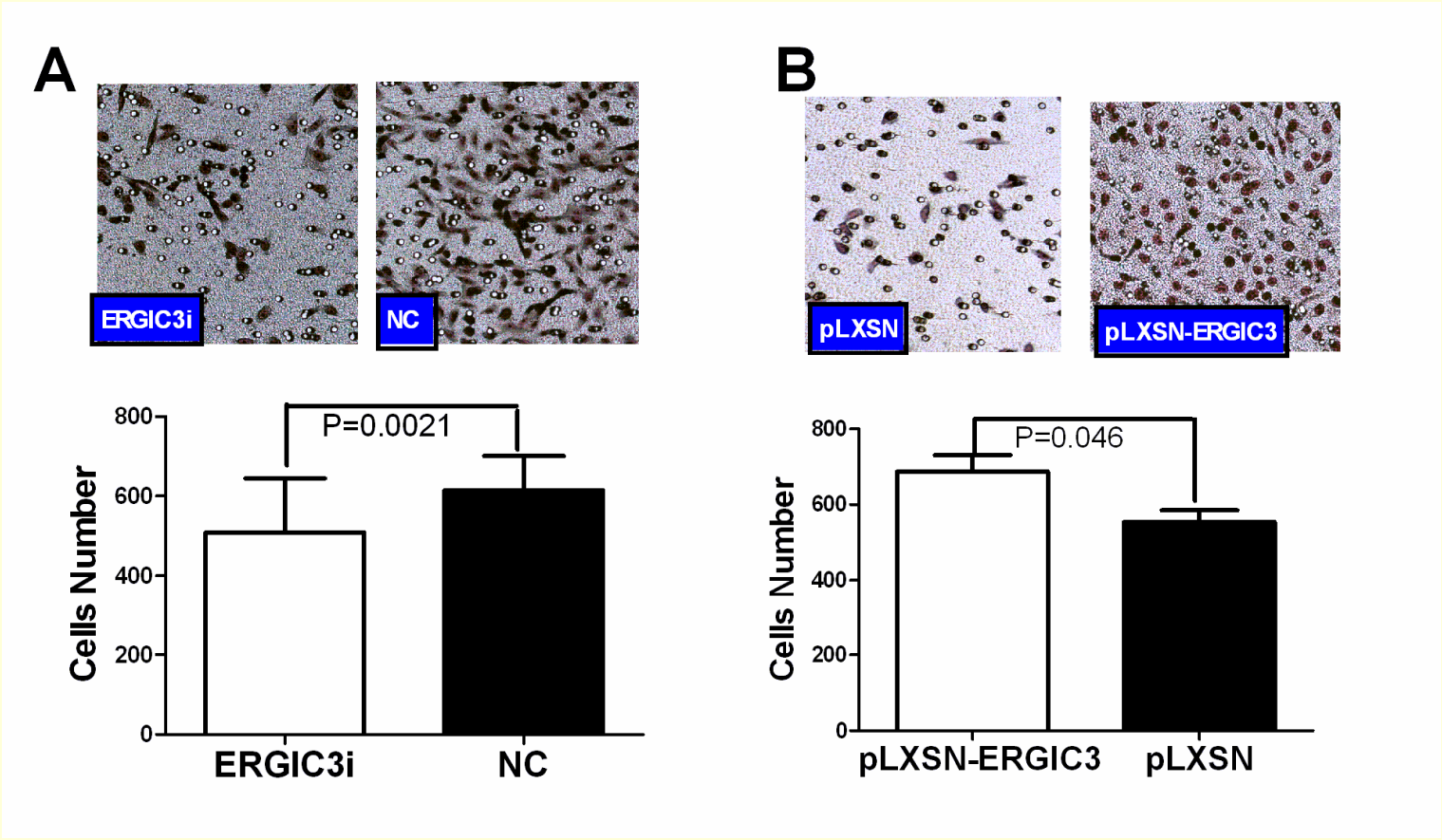


2.2 Functions of ERGIC3 in lung cancer

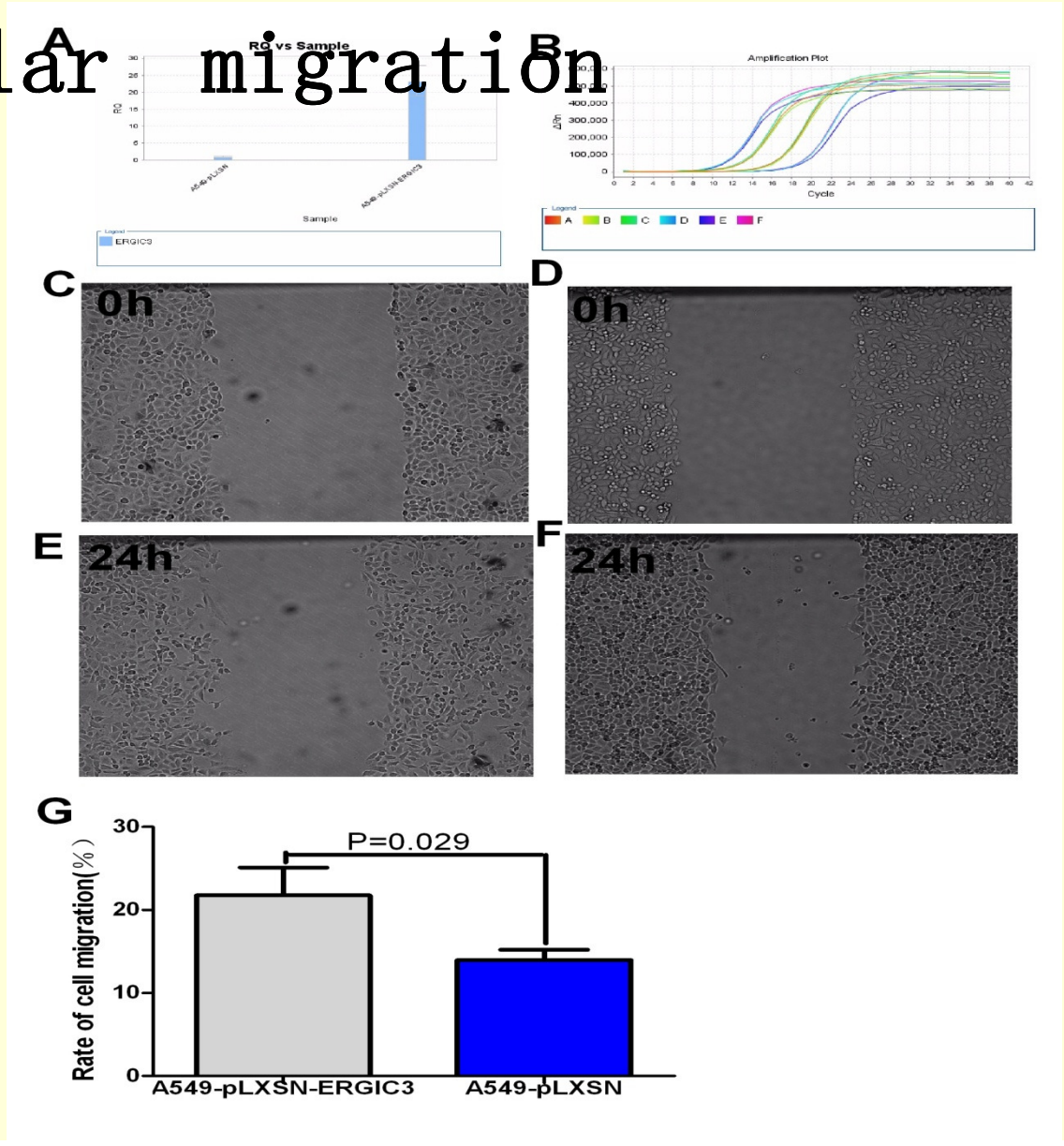
2.2.1 ERGIC3 promotes cellular growth



2.2.2 ERGIC3 promotes cellular migration



2.2.3 Stable overexpression of ERGIC3 promotes cellular migration



3、Summary

- Library of differentially expressed genes in lung cancer.
- Potential targets for diagnostic & therapeutic usage.
- Two lung cancer associated genes.
- ERGIC3 may be a new biomarker for lung cancer.
- ERGIC3 may be a novel candidate oncogen for lung cancer.

Thank you!