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# Disease resistance selection and gene scanning of Chinese shrimp, *F. chinensis*

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# Background



- *F. chinensis* is an important mariculture species with high commercial value in China;
- New varieties of 'Huanghai No. 1' and 'Huanghai No. 2' have alleviated and controlled the decline of shrimp farming caused by diseases, such as WSSV
- **Molecular marker-assisted breeding**

# Selective Breeding- WSSV Resistance



- **New variety, Huanghai-No.2**
- **Duration 1998-2008**
- **Mass selection + Multi-trait selection:**
  - **better growth performance**
  - **higher survival rate**
  - **longer survival time**



# 454 transcriptome sequencing

## Candidates for sequencing

1. Group S: 3 earliest moribund individuals;
2. Group R: 3 survivals;
3. Tissue: muscle and hepatopancrease for total RNA extraction



454/Roche

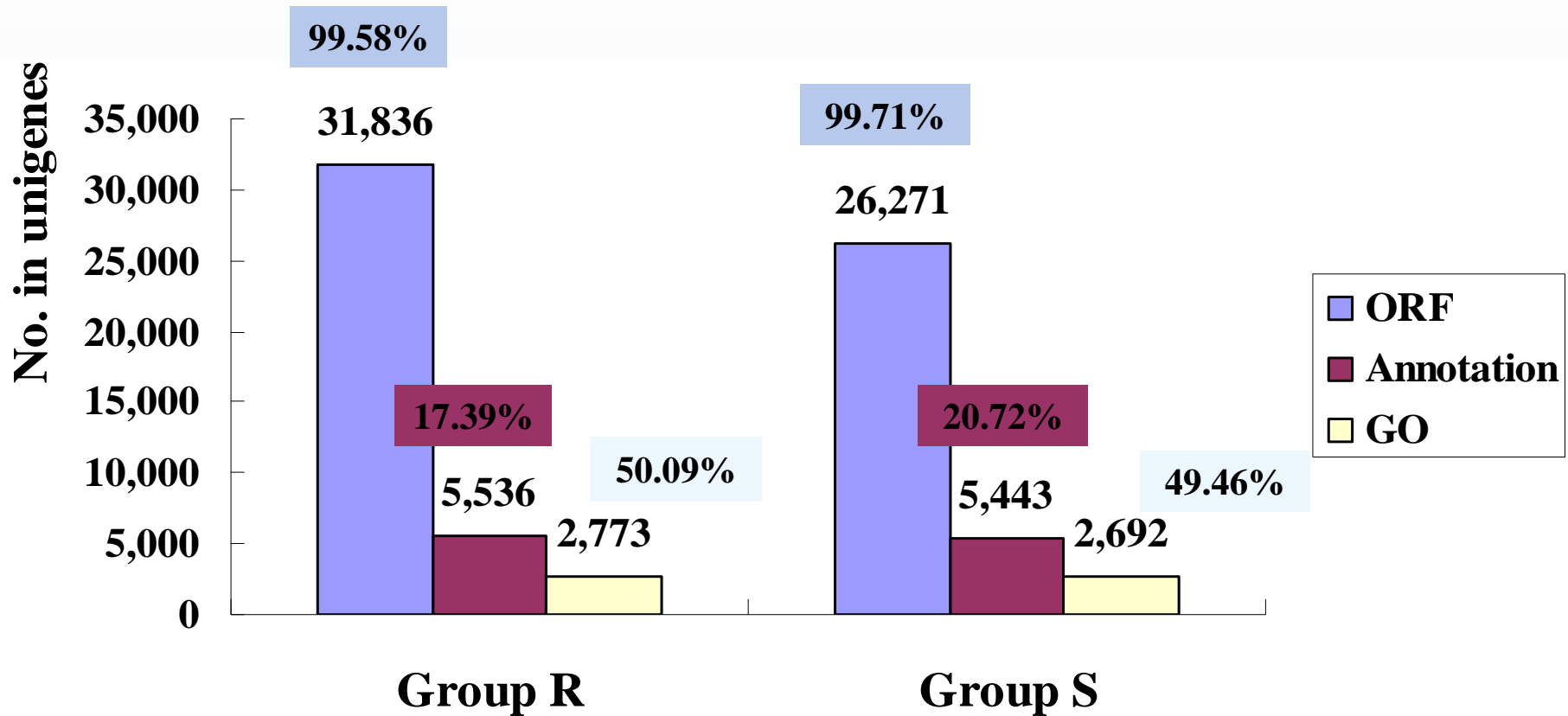
transcriptome sequencing

Chinese National Human Genome Center at Shanghai, Oct. 2008

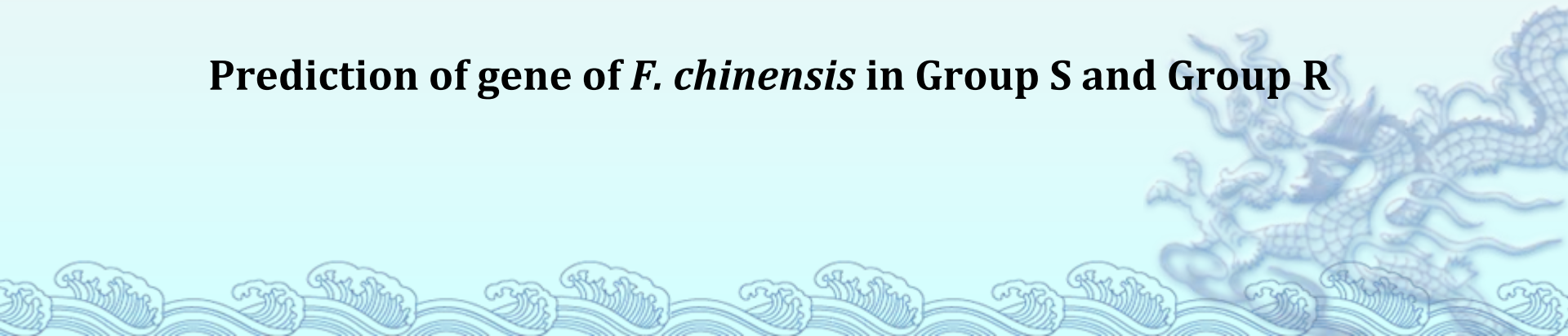
# Basic result of cDNA 454 sequencing-in all

Items	Results
Reads No.(ave len)	497,846 (219bp)
Reads No. after trimming adapters and low value reads (ave len)	451,637 (212bp)
Total bases	95,583,417 bp
reads assembled in contigs	421,516
Contigs No.	18,560
Length of all contigs	6,544,518 bp
Max length of Contig	4,086bp
Mean length of Contigs	353bp
Singletons No.	30,121
Length of all singletons	5,773,404 bp
Length of all contigs and singletons	12,317,922 bp

shrimp genome ~2 billion bp



**Prediction of gene of *F. chinensis* in Group S and Group R**



# Specific sequences in Group S and R

Group		Contigs	Singletons	Sum
R	Total	11,750	20,219	31,969
	Specific	4,258	12,532	16,790
	With annotation	968		
	With GO	427		
S	Total	11,218	15,129	26,347
	Specific	3,748	8,825	12,473
	With annotation	1156		
	With GO	465		

BLASTN (alignment ratio > 35% and  $e$  value >  $1e-10$ )

All these specific contigs were considered very meaningful in analyzing the mechanism of WSSV resistance in shrimp.



# SNP reads (variation) in total reads

SNP	reads	Total	≥6	5	4	3	2	1
Same s	mis-sense and nonsense types of SNPs may reveal the truth of WSSV resistance in shrimp							
Mis-sense		34,642	1,704	440	763	1,593	4,243	25,899
Nonsense		1,478	42	21	28	58	151	1,178
Noncoding		18,275	799	229	437	927	2,482	13,401
Total		71,724	4,241	975	1,668	3,509	9,100	52,231
Indel		31,769	1,060	302	604	1,418	3,733	24,652

Note: based on 18,560 contigs of all reads, including Group R and S.



# Identifying of SNPs associated with WSSV resistance

SNP Locus ID	Genotype	Group	Number of Ind.	$\chi^2$ value	df value	<i>P</i>
C1401-67TA	A/A	S	1	9.31	1	**
		R	12			
C6625-56CT	C/T	S	10	4.5	1	*
		R	22			
C7695-204AG	A/G	S	13	10.29	1	**
		R	1			
C7911-209CT	T/T	S	30	4.26	1	*
		R	16			
C9733-231TA	T/A	S	4	4	1	*
		R	0			
C12698-488CA	A/A	S	34	5.67	1	*
		R	17			
C283-145AG	G/G	S	26	12.50	1	**
		R	6			
C364-89AT	A/A	S	46	5.73	1	*
		R	72			
C12355-592CT	T/T	S	78	8.26	1	**
		R	46			
C12355-592CT	C/T	S	49	6.62	1	*
		R	78			

# Gene associated with WSSV resistance

Contig	Annotation	Species	E
Contig1401	fructose 1,6-bisphosphate aldolase	<i>Bombyx mori</i>	2E-37
Contig6625	deoxycytidylate deaMase	<i>Crassostrea gigas</i>	2E-12
Contig7695	u5 small nuclear ribonucleoprotein 200 kda helicase-like protein	<i>Equus caballus</i>	9E-35
Contig7911	glycine decarboxylase, isoform CRA_a	<i>Mus musculus</i>	1E-50
Contig9733	DEAD(Asp-Glu-Ala-Asp)box polypeptide 39	<i>Macrobrachium nipponense</i>	0
Contig283	effector caspase	<i>Penaeus monodon</i>	3E-36
Contig364	predicted: peptide transporter family 1-like	<i>Apis mellifera</i>	4E-16
Contig12355	40S ribosomal protein S2	<i>Perca flavescens</i>	1E-105

# Further Cooperation with UGent

**Gene discovery in shrimp related to:**

✓ **EMS**

✓ **WSSV resistance**

✓ **Sex determination**

✓ **Temperature tolerance**

✓ **Other economic important gene**





**Thank you  
for your  
attention**