

# 腸病毒71型免疫組合物及其用途

## Immunogenic Compositions and Uses Thereof

This invention relates to immunogenic composition against  
EV71 infection and related methods  
(US 9,051,361 B2)

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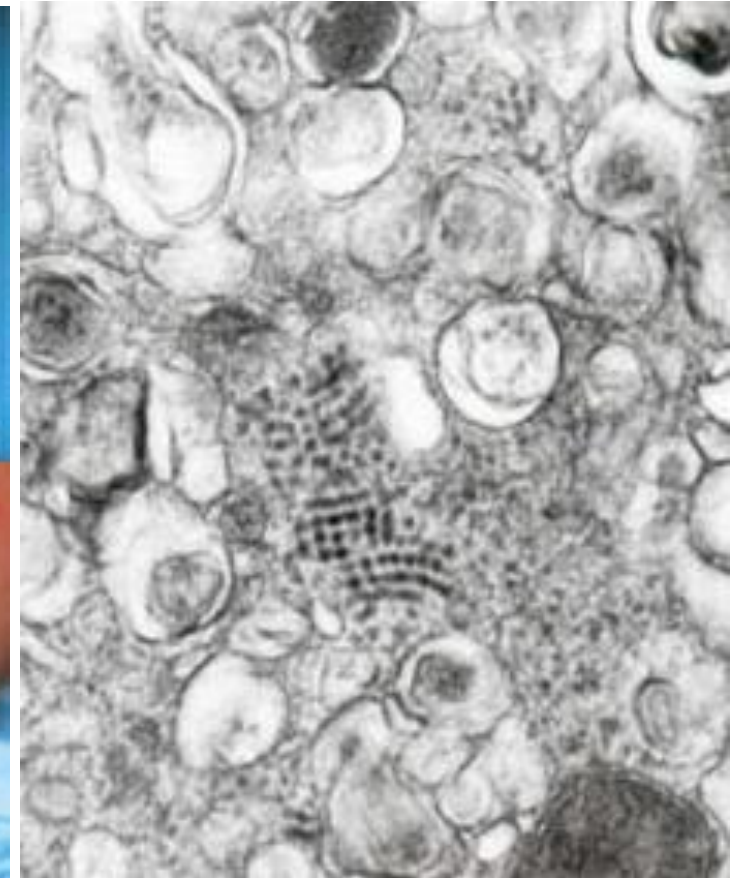
# Hand, foot and mouth diseases (HFMD)

Hand, foot and mouth disease (HFMD) is a self-limiting infection characterized by vesicular exanthema of the hands, feet, mouth and buttocks caused by human enterovirus infections of infants and young children.

Papules and vesicles



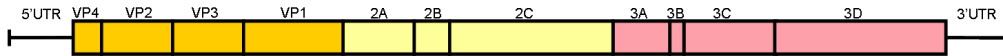
Herpangina



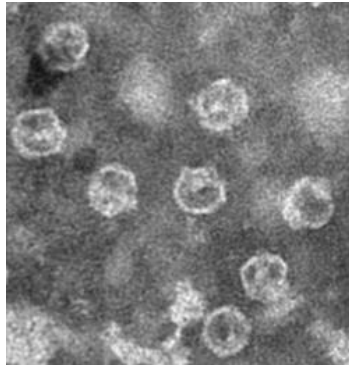
# Enterovirus 71 (EV71) immunogenic compositions

- EV71 is a RNA virus that belongs to the human enterovirus A species of the *Picornaviridae* family.
- EV71 is the predominant cause of HFMD and herpangina.
- NHRI has developed a method of producing a purified EV71 virus antigen, which used to make immunogenic compositions (inactivated EV71 whole-virion vaccine) against EV71 infection.

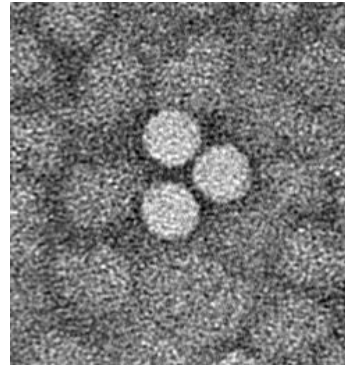
Virus genome 7.4kbp



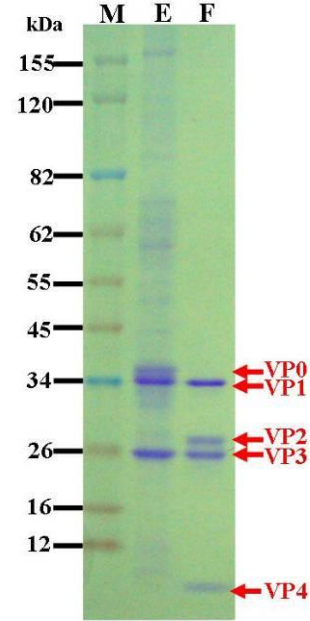
Empty particle



Full particle



4-12% SDS-PAGE  
Coomassie Blue



Structure proteins

- VP0 ~38 kDa
- VP1 ~36 kDa
- VP2 ~28 kDa
- VP3 ~27 kDa
- VP4 ~8 kDa



# NHRI PIC/S GMP facility



NHRI has established a **PIC/S GMP facility** to product viral vaccines.

# Pilot scale production of highly efficacious and stable EV71 vaccine

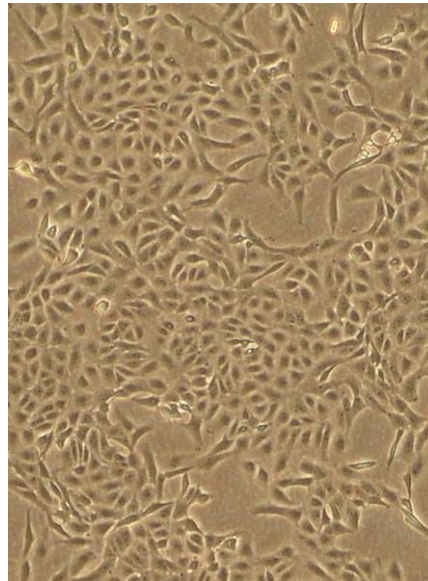
- The EV71 vaccine (E59 strain) is obtained from the Taiwan CDC.
- Master and working Vero cell and virus seed banks are established following cGMP guidelines, characterized to fulfill the requirements for the manufacture of biological products.

## Upstream: Serum-free roller bottle process

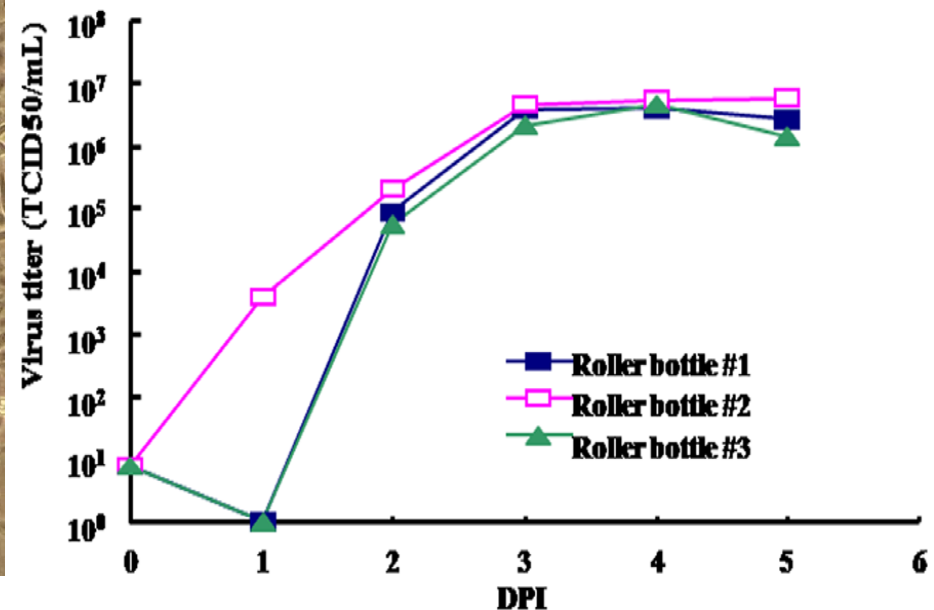
Incubator



Vero cell



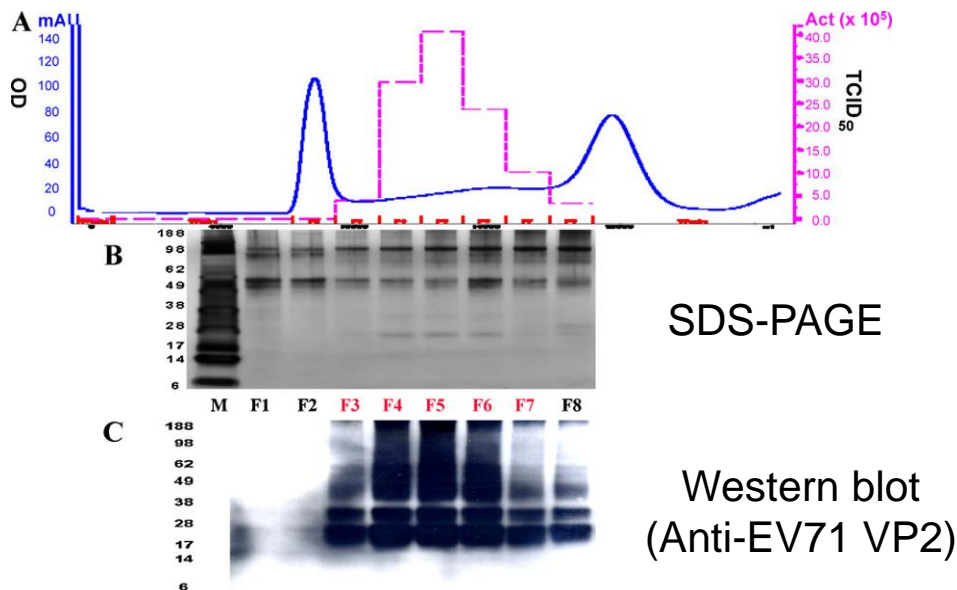
EV71 virus titers



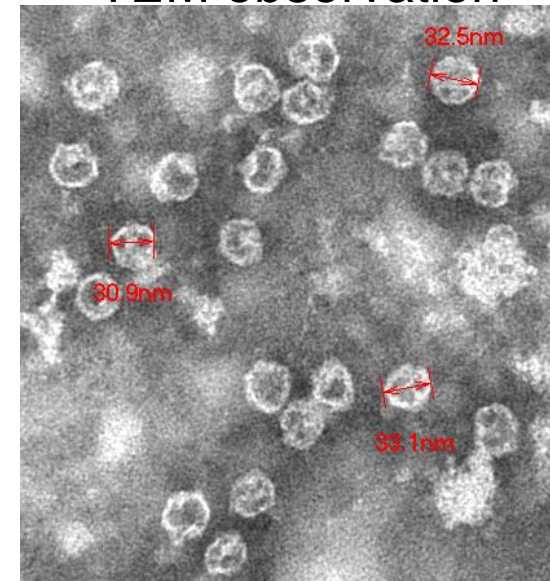
# Pilot scale production of highly efficacious and stable EV71 vaccine

- The production processes are investigated, characterized and quantified to establish the potential vaccine manufacturing process including the time for virus harvest, the membrane for diafiltration and concentration, the gel-filtration chromatography for the downstream virus purification.

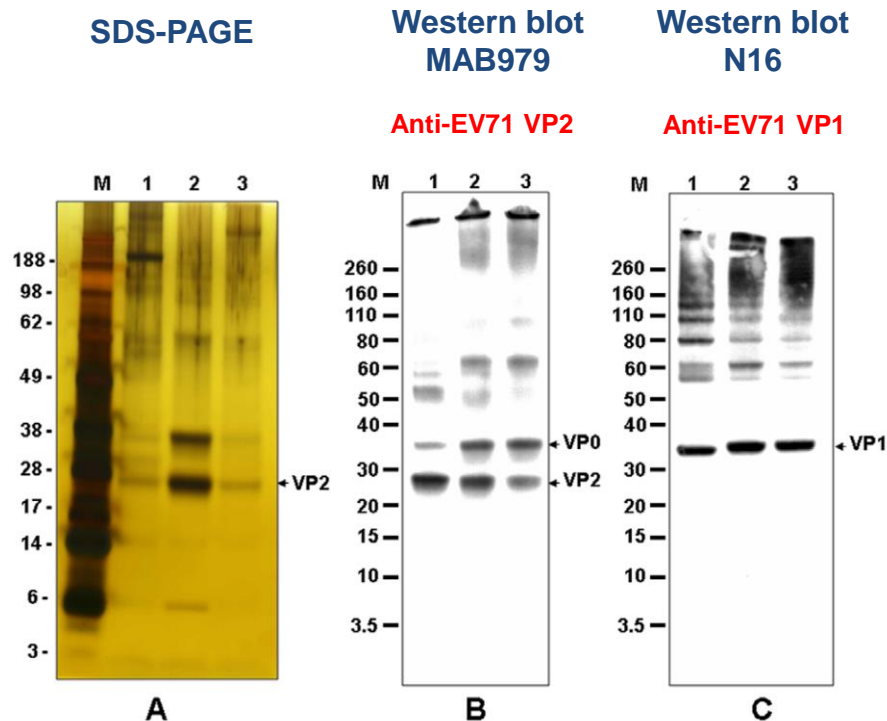
## Downstream: Gel-filtration chromatography



## TEM observation



# Stability profiles of different Lots of EV71 vaccine products



**M:**Novex Sharp Standard

**1:** Serum-contain process (stored for **26** months)

**2:** Serum-free process (stored for **13** months)

**3:** Serum-free process (stored for **4** months)

## Mouse model

Time		Neutralization Titer (GMT $\pm$ SE)	
		Lot #1	Lot #3
Initial	4°C	443 $\pm$ 279	233 $\pm$ 99
1 month	4°C	153 $\pm$ 52	265 $\pm$ 27
2 month	4°C	105 $\pm$ 89	246 $\pm$ 14
3 month	4°C	120 $\pm$ 58	400 $\pm$ 88
	25°C	269 $\pm$ 110	913 $\pm$ 125
6 month	4°C	79 $\pm$ 29	335 $\pm$ 83
	25°C	262 $\pm$ 122	850 $\pm$ 122
9 month	4°C	534 $\pm$ 90	1089 $\pm$ 187
12 month	4°C	301 $\pm$ 86	349 $\pm$ 85
18 month	4°C	534 $\pm$ 77	643 $\pm$ 97

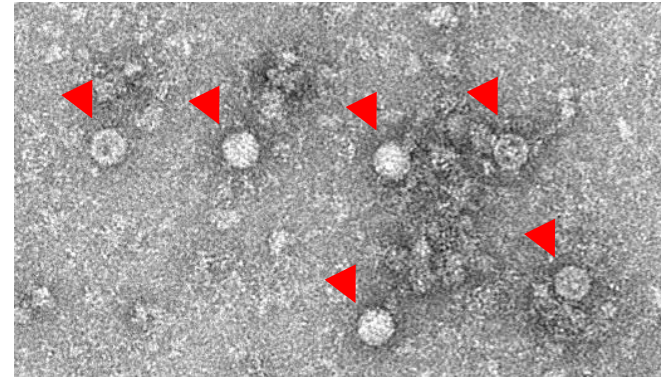


# EV71 vaccine clinical trial phase I

## EV71 vaccine product



## TEM observation



Study design (NIH Clinicaltrials.gov #NCT01268787)

Group	Dosage	Content	
		Total protein	Adjuvant $\text{AlPO}_4$
A05 (30 subjects)	0.25 mL	5 mcg	150 mcg
B10 (30 subjects)	0.5 mL	10 mcg	300 mcg

Vaccination: Day 0, Day 21

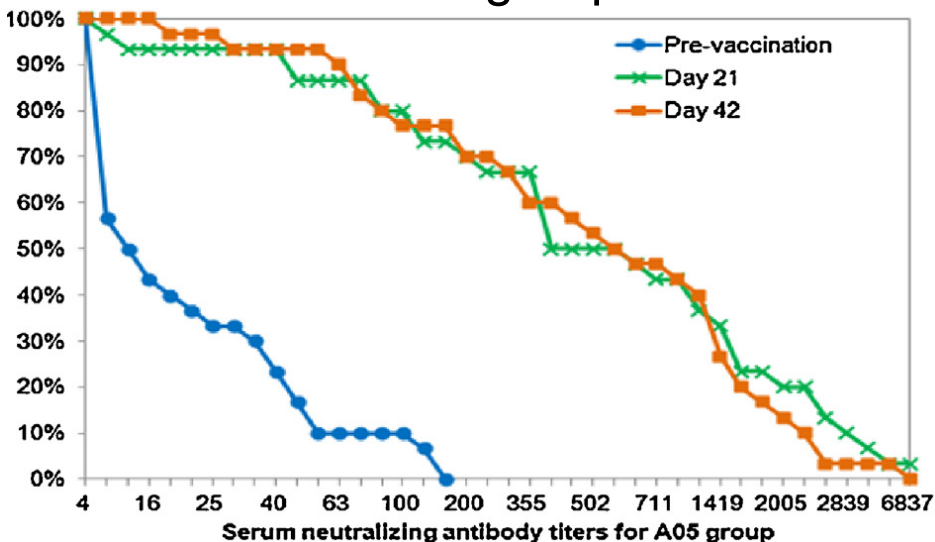
Clinical trial sites: Taipei Veterans General Hospital  
National Taiwan University Hospital



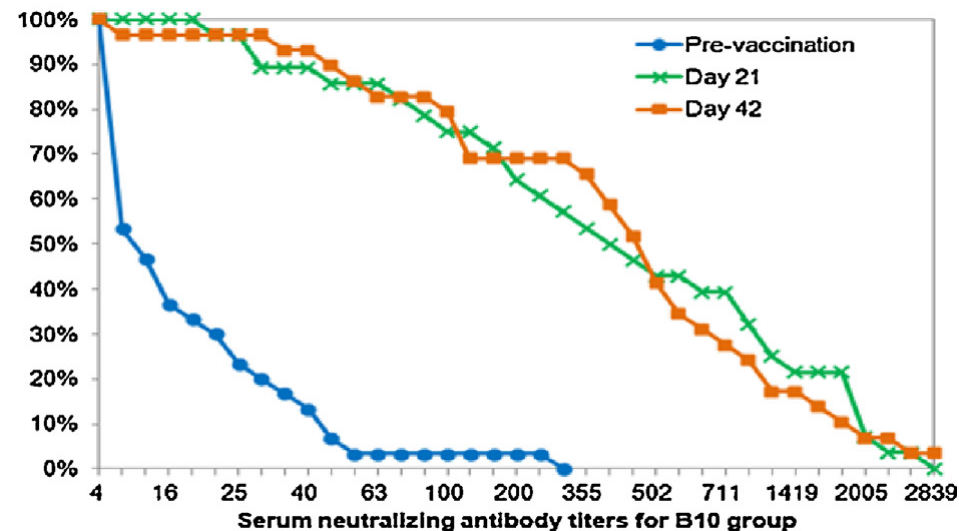
# Results of human clinical trial phase I

## Reverse cumulative distribution of serum neutralizing antibody titers in subjects

A05 group



B10 group



Results: The 5 mcg and 10 mcg EV71 vaccines are generally safe and immunogenicity in healthy adults.

NHRI has transferred raw materials and technologies to two Taiwan local vaccine companies.

# Acknowledgements



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衛生福利部 疾病管制署

CENTERS OF DISEASE CONTROL



國家衛生研究院

National Health Research Institutes



衛生福利部食品藥物管理署

Food and Drug Administration



財團法人醫藥品查驗中心

Center for Drug Evaluation, Taiwan



臺北榮民總醫院

Taipei Veterans General Hospital



國立臺灣大學醫學院附設醫院

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