

Name: Using paramagnetic particles and PNA for  
isolation and electrochemical detection  
of DNA corresponding influenza virus

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# VIET NAM

- First appearance of influenza virus: 12/2003.
- Up to now: 3 pandemics caused 95 cases of H5N1 and 43 people were dead.
  - First pandemic 12/2003 - 27/4/2004: 57 provinces and cities, 43.9 million of domestic fowls were dead.
  - Second pandemic 4/2004 - 11/2004: 17 provinces and cities, 84.078 thousand of domestic fowls were dead.
  - Third pandemic 12/2004 – 5/2005: 36 provinces and cities.

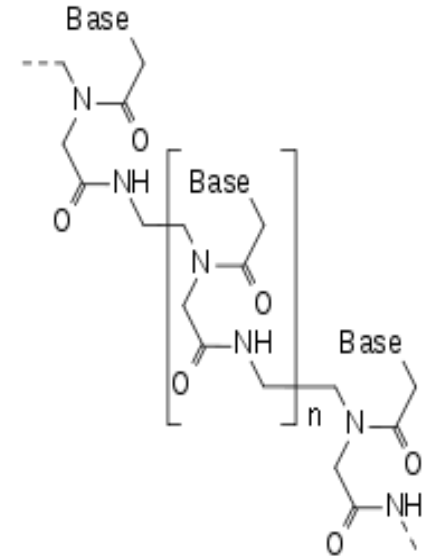
# VIET NAM



Places suffered from H5N1 pandemic in Vietnam

# PNA

- ❖ PNA has a backbone made from repeating N-(2-aminoethyl)glycine units linked by peptide bonds. The different bases (purines and pyrimidines) are joined to the backbone by methylene or carbonyl linkages.



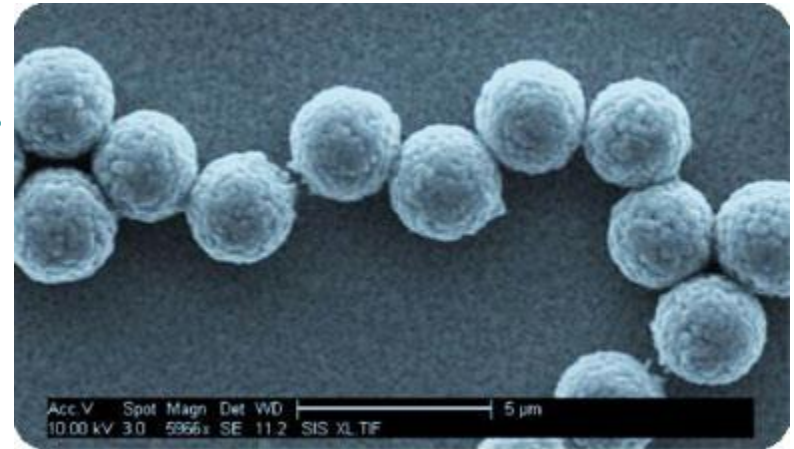
- ❖ PNA/DNA is better thermal stability than DNA/DNA
- ❖ PNA has various application such as: antigen and antisense therapy; PNA as molecular biology and functional genomics, PNA as a probe for diagnosis and detection, and PNA as biosensor.

# PNA in gene therapeutic

- ❖ PNA offer the ability to be used as adapters, linking plasmid vectors to peptides, proteins, drugs, and molecular tracers.
- ❖ PNA can be also used to form PNA/DNA duplex with DNA influenza virus. PNA/DNA duplex formation lowers ability of replication of DNA influenza virus.

# Paramagnetic particles

❖ Small size but large surface (2 nm-10  $\mu\text{m}$ ), different variant of modification.



❖ Their ability to facilitate bioactive molecules binding

❖ Advantages of paramagnetic particles: easy using, short time.

# **METHOD**



# Method

- ❖ Automatic pipetting station EP Motion 5075 (Eppendorf, Germany) was used for fully automated isolation process of target DNA sequence (5'-CCTCAAGGAG-3') corresponding to influenza virus by using Oligo dt(25) and PNA (5'-AAAACTCCTTGAGG-3').



- ❖ Square wave voltammetry, square wave voltammetry coupled with adsorptive transfer technique, and differential pulse voltammetry method were used for electrochemical detection of nucleic acids.

# **RESULT AND DISCUSSION**

# Result and discussion

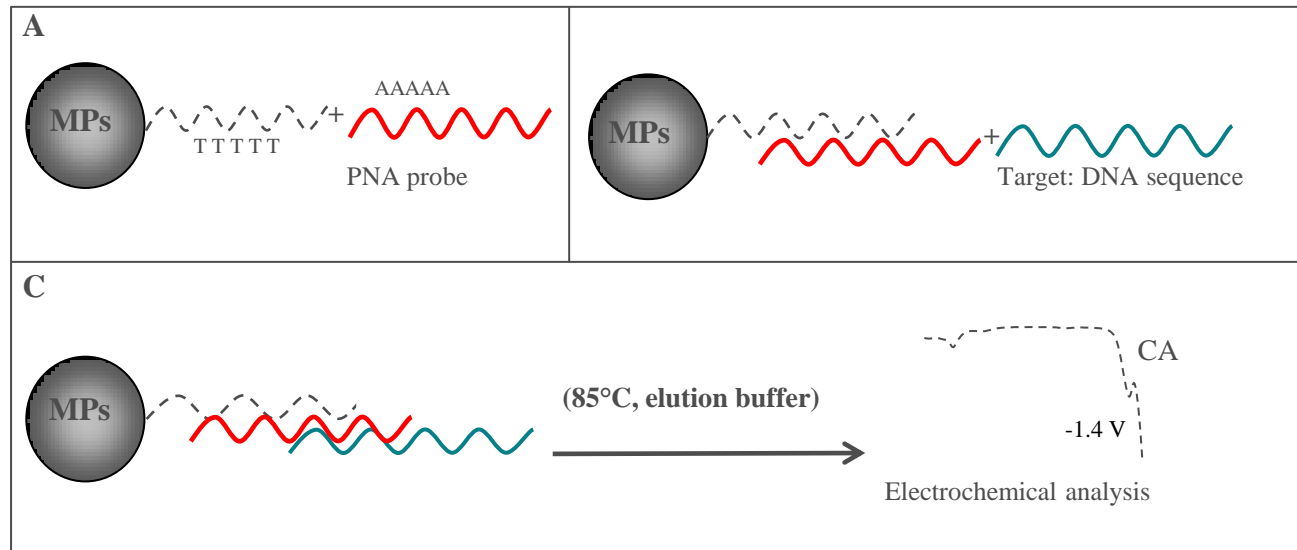
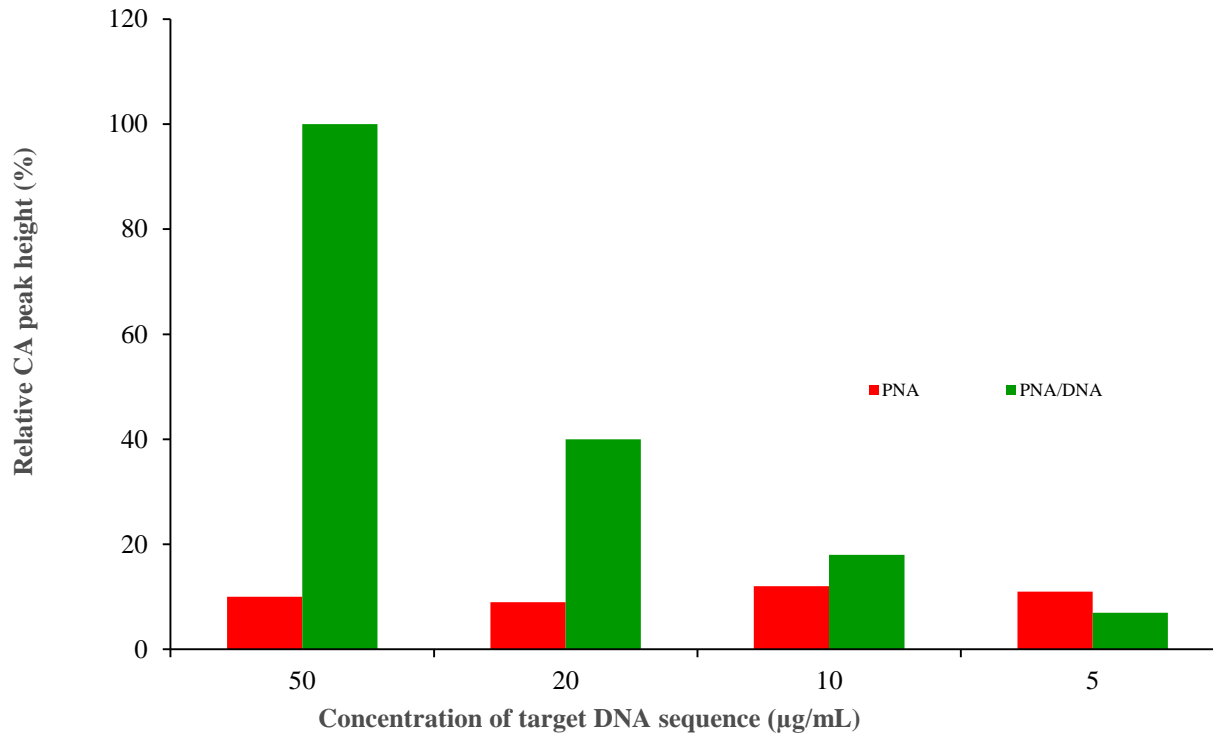


Figure.1: Scheme of isolation and detection of influenza derived oligonucleotide by MPs and PNA probe. A PNA binding MPs, B addition of DNA target sequence, C binding of DNA to MPs with PNA, D electrochemical detection of isolated product.

# Result and discussion



**Fig. 2:** Dependence of relative CA peak height (%) of PNA and PNA/DNA on concentration of applied target DNA influenza derived sequence ( $\mu\text{g/mL}$ ). Measurements were carried out by AdT SWV. Parameters of AdT SWV was: time of accumulation 120s; purge time 60s; frequency 280 Hz; initial potential 0 V; end potential -1.8 V; step potential 0.00495 V; amplitude 0.02505 V.

# Conclusion

- ❖ Electrochemical method is a powerful technique for nucleic acid determination.
- ❖ PNA can be used as biosensor for DNA target sequence because PNA shows ability to hybridize with DNA with high affinity and specificity.
- ❖ Paramagnetic particles and PNA as a probe can be used for isolation of DNA target sequence because this established technique can facilitate DNA isolation process.

# Thank you for your attention