# *The European Diphtheria Surveillance Network (EDSN): a strong model to combat a rare disease and use resources efficiently, share knowledge openly, and give support effectively*

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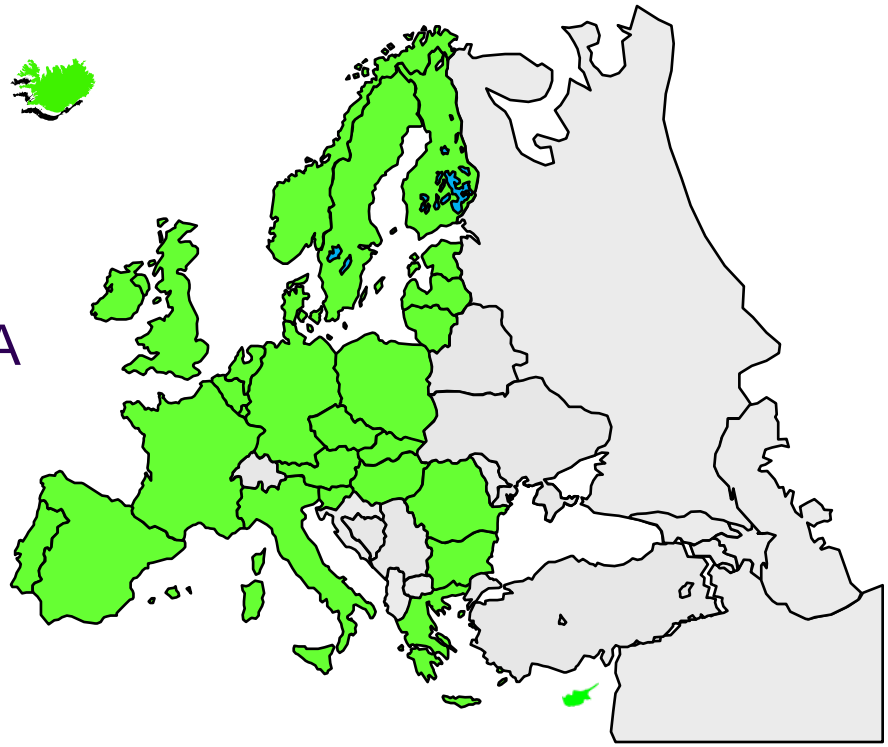
# Diphtheria perceived as low risk


- However, one of the largest epidemics was observed in 1990s in WHO EU region
- Sporadic cases of *C. diphtheriae* and *C. ulcerans* detected in some EU countries
  - Recent large outbreaks are ongoing in other WHO regions (India, Indonesia, Sudan)
- Latvia continues to experience highest incidence in the entire WHO EU region
- But waning immunity and minimal awareness causes diphtheria to be a threat and could return in epidemic proportions to Europe

**Good surveillance and laboratory detection of organisms is crucial for accurate diagnosis and a strong EU network will maintain these specialist skills**

- At request of WHO EURO, ELWGD established in 1993
- DIPNET was DGSANCO-funded between 2001 – 2009
- ECDC inherited network in 2010 and now expanded to all EU/EEA countries
- Laboratory activities tendered to HPA, London
- Continues to integrate microbiologists and epidemiologists



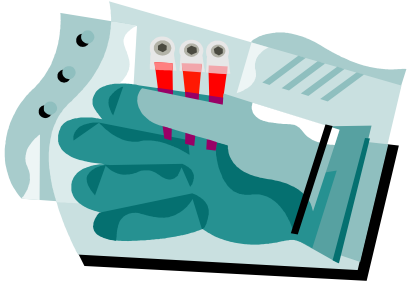
# European Diphtheria Surveillance Network – *Laboratory activities*

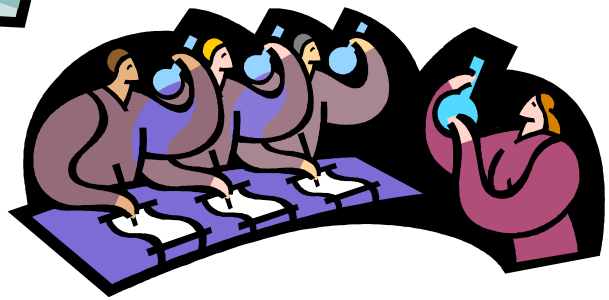

*'Coordination of a European laboratory network to strengthen laboratory diagnostics for diphtheria surveillance'*

- WP1: Coordination of the laboratory surveillance network of diphtheria
- WP2: Organisation of EQA scheme for the reference laboratory diagnostics of diphtheria
- WP3: Evaluation and assessment of serological immunity methods and EQA scheme of diphtheria (subcontracted to Institut Superiore di Sanita, Italy)
- WP4: Provision of hands-on practical laboratory training workshops in diagnostic methods

# EDSN Activities to date

- **Two annual meetings**
  - –June 2010 and March 2011
- **Two diagnostic EQAs**
  - –June 2010 and May 2012
- **One serology EQA**
  - –January 2012
- **Two training workshops**
  - –July 2010 and November 2011





# Laboratory Diagnostics Workshops (WP4)

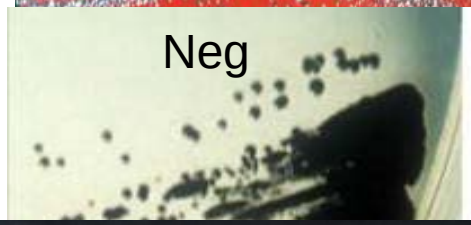
- July 2010, London: 4 ‘newcomer’ participants, Belgium, Hungary, Malta and Luxembourg
- November 2011, Athens: 14 participants, based on 2010 EQA results and ‘newcomers’

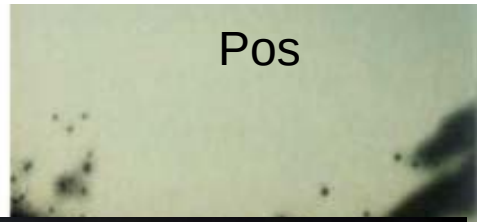
**Three day workshop, topics covered;**

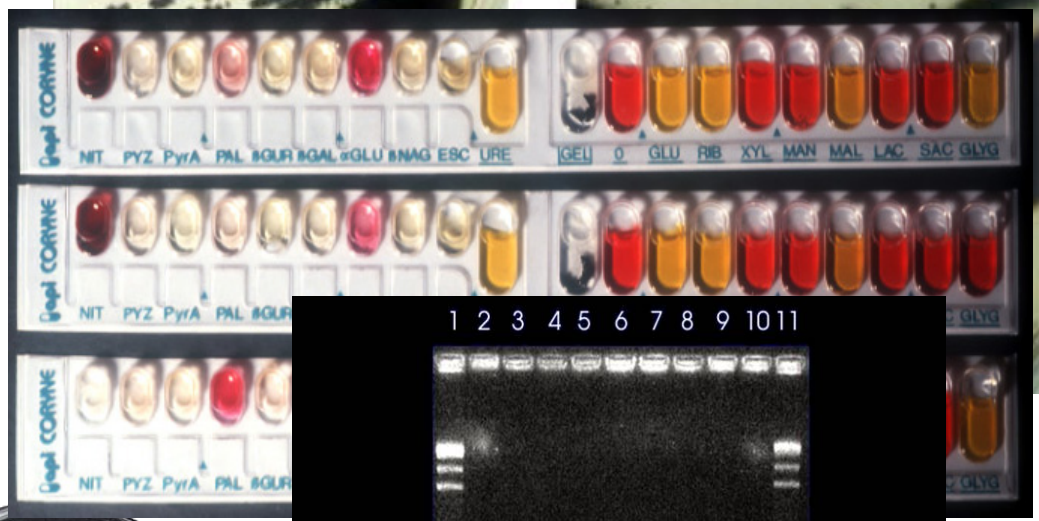
– clinical, epidemiology and microbiological talks
– primary culture, screening tests & identification methods
– phenotypic and molecular toxigenicity testing
– demonstration of serological assays
– discussions on screening throat swabs and problems acquiring reagents

# Diphtheria diagnostics: Key lab tests

- Gram positive rods
- Black colonies on Hoyles/Tellurite media
- Essential tests: Catalase pos, Cystinase pos, Pyrazinamidase neg
- Four *C.diphtheriae* biovars: *mitis, gravis, belfanti, intermedius*
- Diphtheria toxin is major virulence factor
  - –Elek test
  - –PCR




# Laboratory Diagnosis of Diphtheria Workshop November 2011 – Athens,

# Laboratory diagnostic EQAs (WP2):
## Preparation & despatch


•Laboratory Questionnaire sent April 2010 to all 30 participants

– Level of Reference, Laboratory Diagnosis, Toxigenicity Testing, Serological Assays, Culture Collections, Antibiotic Sensitivity, Epidemiological Typing, EQA Participation

•Each panel consisted of six simulated throat swabs

– target organisms selected and checked by SDRS, HPA

– 'specimens' prepared and freeze-dried by Quality Assurance Lab, HPA

– panel checked for content by SDRS before shipping to participants

Participants sent back results to HPA for analysis

Fully concordant result = matched identification, biotype and toxigenicity

Acceptable result = did not match biotype

# Laboratory diagnostic EQA

## Performance of centres

### 2010 EQA

- Measured by a fully correct or acceptable result;
  - Only five centres produced acceptable results for all six strains
    - Denmark, France, Malta, Norway and the UK
- 156 available reports (6 strains, from 26 centres)
  - 21 **(14%)** unacceptable identification reports (at species level)
  - 16 **(10%)** unacceptable toxigenicity reports

### 2012 EQA

- Measured by a fully correct or acceptable result;
  - Ten centres produced acceptable results for all six strains
    - Austria, Cyprus, France, Iceland (ID only), Malta (ID only), the Netherlands, Norway, Slovakia, the UK and the USA
- 186 available reports (6 strains, from 31 centres)
  - 18 **(10%)** unacceptable identification reports (at species level)
  - 20 **(11%)** unacceptable

# Participant problems

- Centres still experience difficulties with the Elek test
  - Several countries received specialised media and reagents from HPA, which are becoming increasingly difficult to obtain
  - Performing test can be difficult to interpret – training workshops are key
  - False negative toxigenic results, would impact negatively on the speed of public health action and patient management

- Isolation and identification of target organism also problematic
  - Eleven countries reported worse results for the recent EQA, mostly due to incorrect identification
  - Identification systems should not be solely relied upon – if identity is <95%, additional tests may be required

# Serology EQA

## Coordinated by ISS, Rome, Italy


- Important for monitoring vaccine efficacy, individual & population immunity
- Sixteen centres tested blind panel of 150 sera
  - Using Vero cell TNT, DELFIA, MIA or ELISA
- Reference assay selected was the TNT from lab I (TNT currently gold standard)
  - Participants compared on quantitative and qualitatitve basis

- Performance of labs using the TNT was generally very good (n=4)
- *in vitro* methods such as dDA-DELFIA or MIA was also good (n=2)

# Conclusions

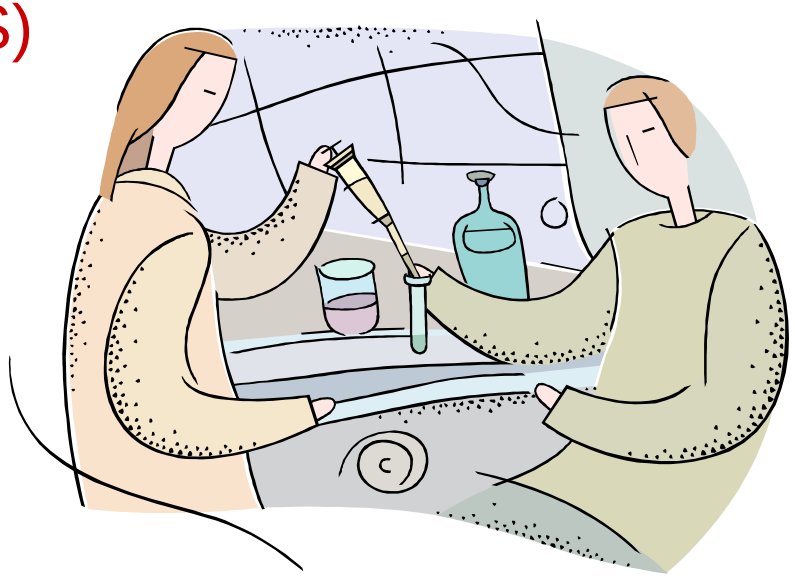

- The quality of surveillance data is strongly supported through regular EQA exercises so as to ensure prompt and accurate microbiological diagnoses
  - These EQA results indicated that further training and EQA exercises are essential to maintain expertise and assess capabilities in the EU

- Participating in training and EQAs gives confidence and encourages people to expand their diphtheria laboratory capabilities

- There still remains an urgent need to continue the network for laboratory diagnostics of diphtheria

# Acknowledgements

- EDSN participants
- Ida Czumbel (ECDC )
- Christina von Hunolstein (ISS)
- Androulla Efstratiou (HPA)



http://ecdc.europa.eu/en/activities/surveillance/edsn/pages/index.aspx