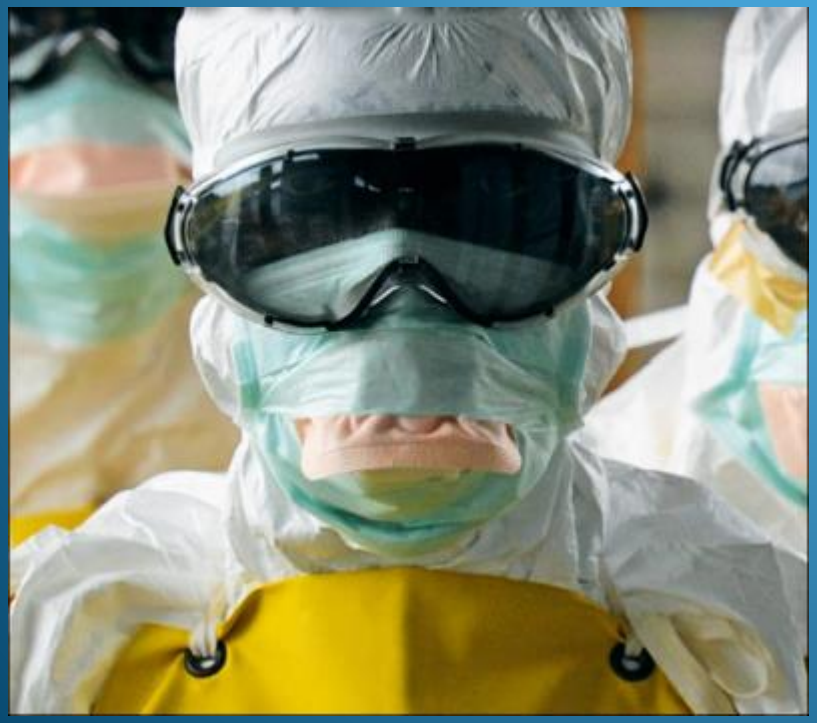
# TACKLING EBOLA

THE WEST AFRICAN LABORATORY RESPONSE



# PRESENTATION AT THE PATHOLOGY IS GLOBAL SYMPOSIUM

ROYAL COLLEGE OF PATHOLOGISTS
LONDON ,UNITED KINGDOM

BY

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1st NOVEMBER , 2016

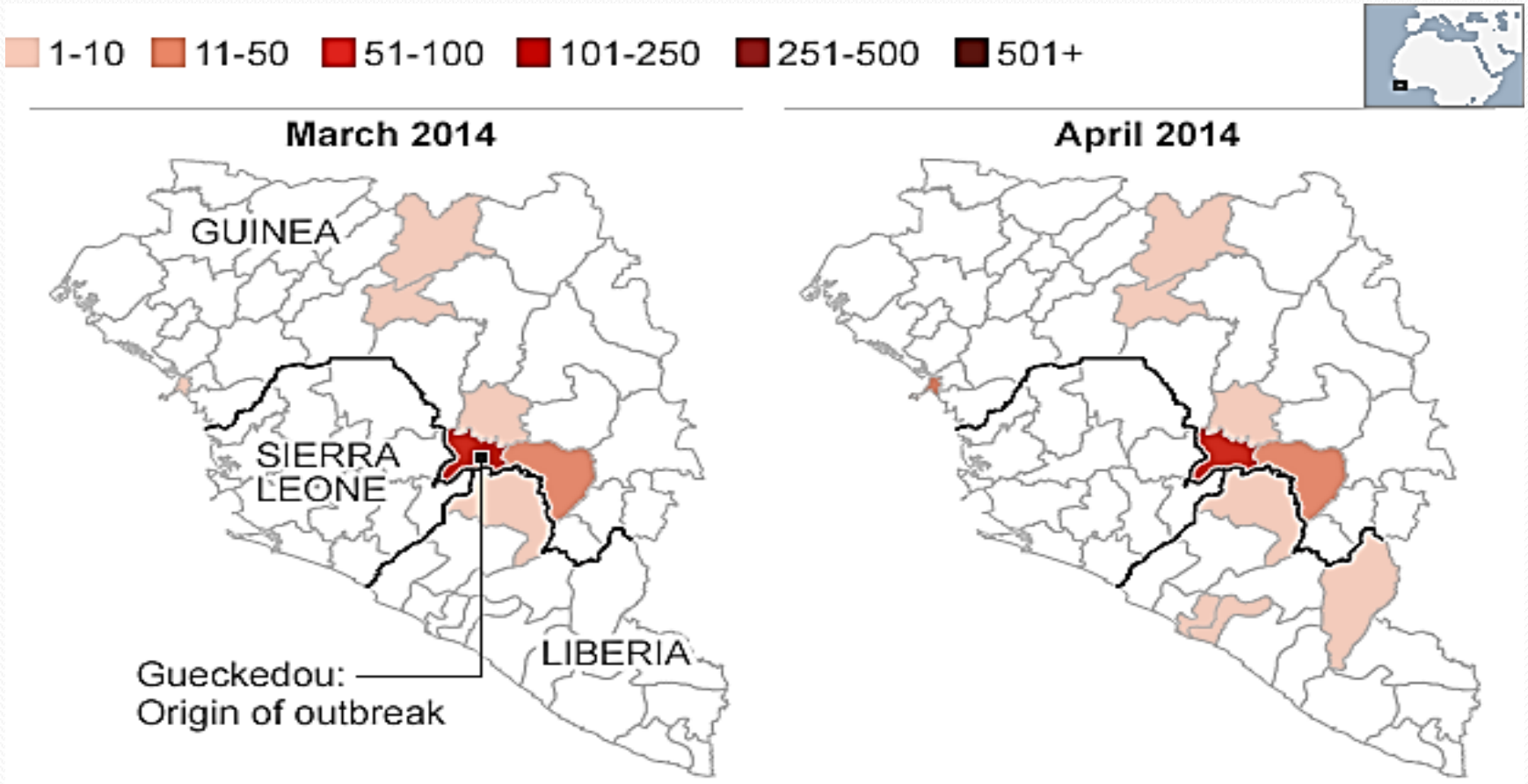
# OUTLINE OF PRESENTATION

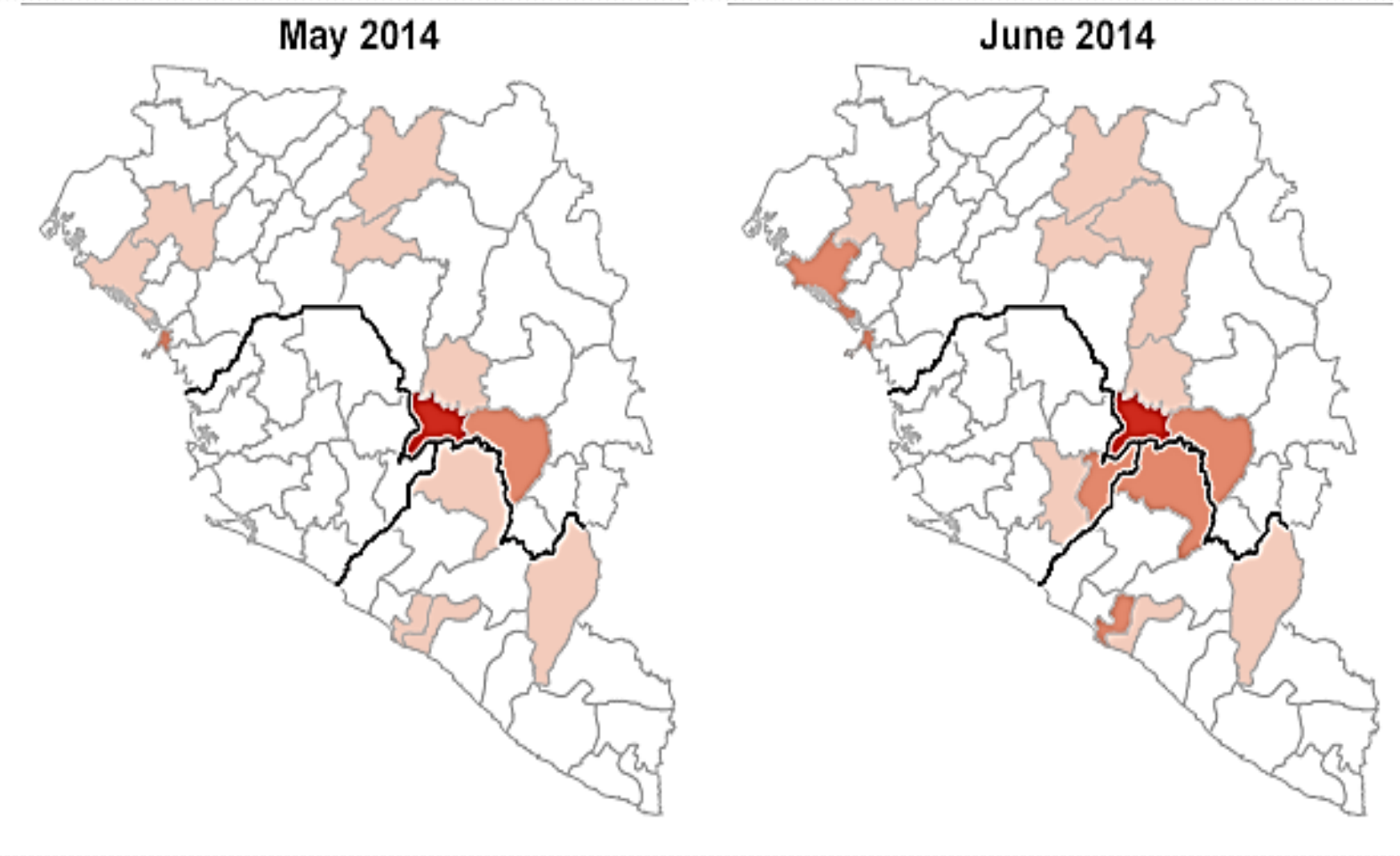
- **Origin ,spread and magnitude of Ebola Virus Disease (EVD) in West Africa**
- **Causes of spread of the virus**
- **Laboratory diagnosis at initial stage of epidemic**
- **EVD laboratories in West Africa during the epidemic**
- **Laboratory operations : diagnostic methods , coordination ,challenges and outcomes**
- **EVD sequencing and effects on control of the epidemic**
- **Studies on convalescent plasma therapy**
- **Bio banking and bio security**
- **Recommendations**

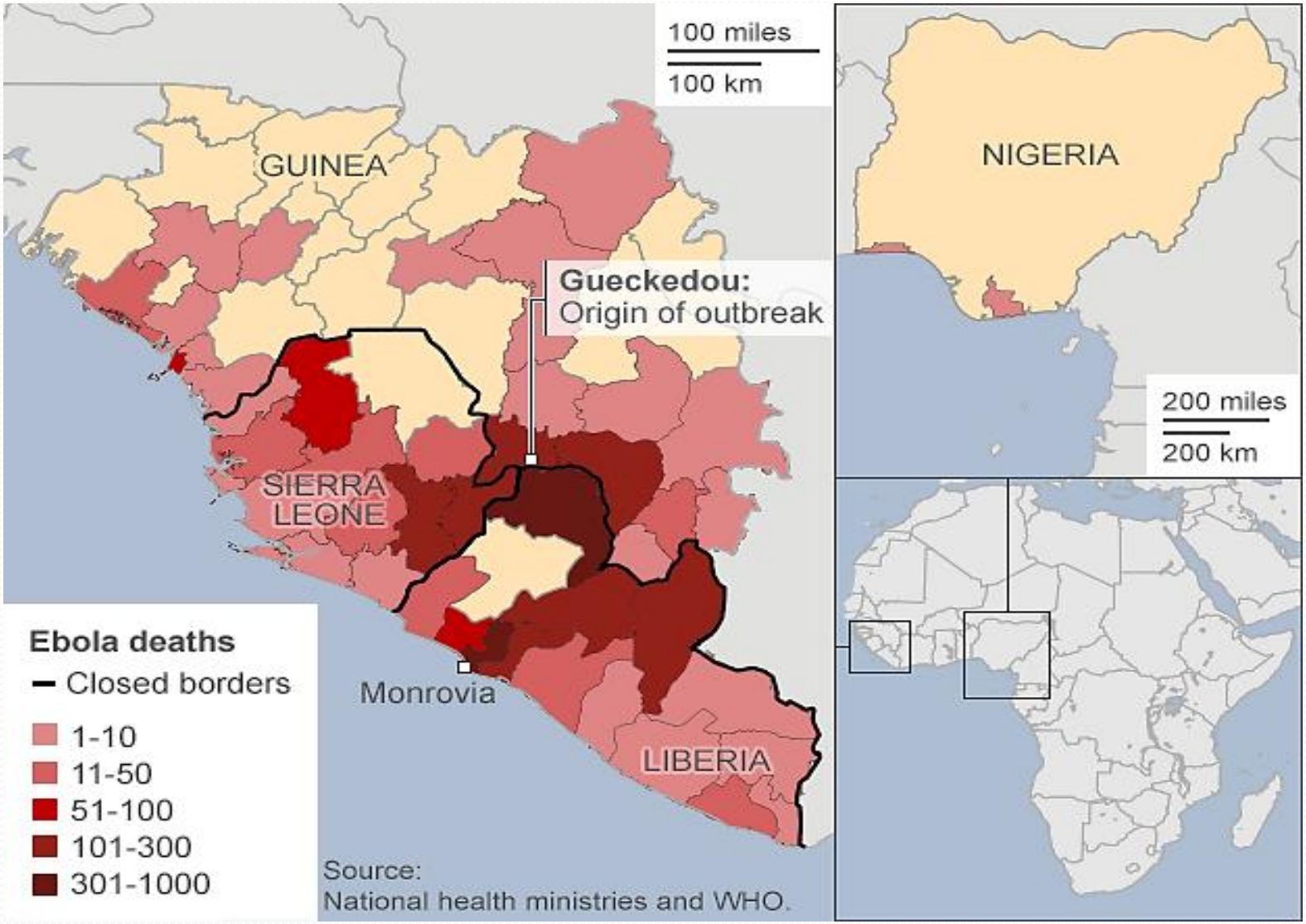
# ORIGIN, SPREAD AND MAGNITUDE OF THE EPIDEMIC

- The Ebola virus disease in West Africa has been the worst both in magnitude and geographical spread in history and was responsible for over 28,000 infections and 11,000 deaths.
- The initial cases were diagnosed in March 2014 in the Guckedou province in Guinea close to the boarders with Liberia and Sierra Leone.
- The virus spread to these neighboring countries by June 2016 reaching a peak in all geographical divisions by December 2014,

# ORIGIN OF EVD OUTBREAK IN WEST AFRICA




May 2014
June 2014


100 miles
100 km
GUINEA
Gueckedou:
Origin of outbreak
SIERRA LEONE
Monrovia
LIBERIA
Ebola deaths
Closed borders
1-10
11-50
51-100
101-300
301-1000
Source:
National health ministries and WHO.
NIGERIA
200 miles
200 km

# COUNTRIES AFFECTED BY CURRENT EBOLA OUTBREAK

| | Country | Cases | Deaths | Last update<br>On 16 September 2015 by WHO |
|---|---|---|---|---|
|  | Liberia | 10,672 | 4,808 | |
|  | Sierra Leone | 13,756 | 3,953 | |
|  | Guinea | 3,792 | 2,530 | |
|  | Nigeria | 20 | 8 | |
|  | Mali | 8 | 6 | |
|  | United States | 4 | 1 | |
|  | Italy | 1 | 0 | |
|  | United Kingdom | 1 | 0 | |
|  | Senegal | 1 | 0 | |
|  | Spain | 1 | 0 | |
| | Total | 28,196 | 11,306 | as of 13 September 2015 |

# CAUSES OF RAPID SPREAD OF EBOLA IN WEST AFRICA

- Urban characteristics, population mobility
- Altered viral characteristics and clinical presentation
- Cultural practices and resistance
- Post conflict economic circumstances and social environment
- Home treatment of patients by health workers
- Limited infrastructure and disaster preparedness
- Insufficient human resource capacity

# CAUSES OF RAPID SPREAD OF EBOLA IN WEST AFRICA

- Delayed community engagement and empowerment
- Lack of financial resources
- Ill defined coordination, communication and collaboration
- Limited laboratory capacity, infrastructure and equipment
- Late declaration by WHO of the international nature of epidemic
- Inadequate PPE and poor IPC practices by health workers

# A COMPLEX EBOLA EPIDEMIC RESPONSE

- Several key players including the governments and line ministries, who, the world bank, united nations agencies and other international agencies like DFID, local NGO and the local community.
- Coordination of the response was undertaken by the national Ebola response centres.
- The technical arm of the response was effected by different pillars.
- Case management, psychosocial, surveillance, communications, safe burials, social mobilization, and the laboratory.

# INITIAL EVD LABBORATORY DIAGNOSIS

- There was a paucity of laboratories capable of diagnosing Ebola virus disease in West Africa before the outbreak.

- The first case of Ebola in Guinea West Africa was diagnosed by a European mobile laboratory in located in France on 23rd of March 2014.

- In Sierra Leone the first case was diagnosed in the Lassa fever laboratory which was established by Tulane university, USA about a decade prior to the EVD epidemic.

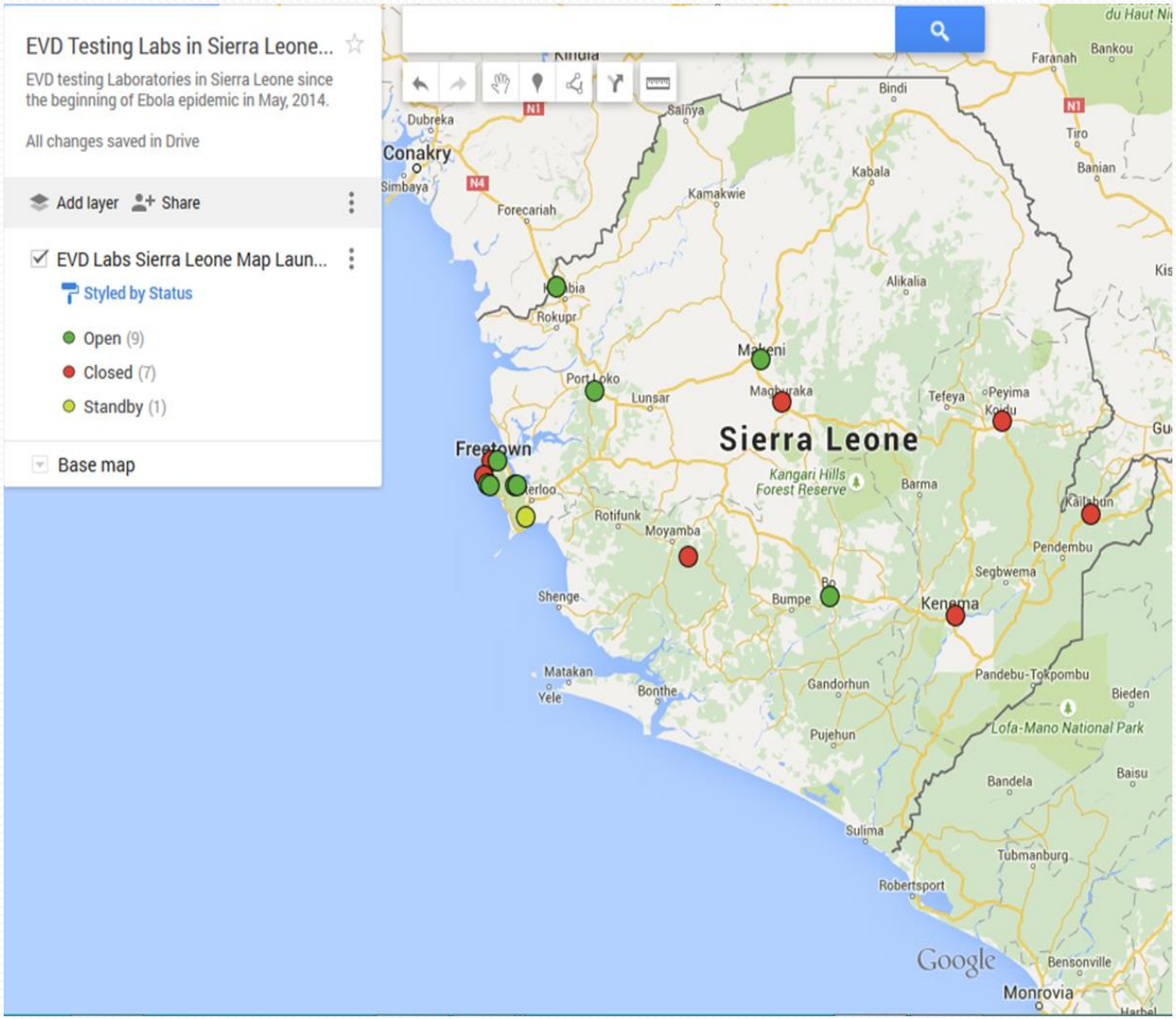
- In Nigeria diagnosis was done within the first 24 hours at the virology department of the College of Medicine - University of Lagos.

- However, as the epidemic spread within the various countries, considerable difficulties emerged including transportation of samples on rugged roads from far away distances and a marked increase in the number of sample thus delaying the response due to prolonged turnaround times.

- The capacity of these laboratories in the most affected countries was soon outstretched with the rapidly advancing epidemic thus necessitating the deployment of several mobile laboratories countrywide to shorten the turnaround times and improve efficiency.

# RUGGED ROADS IN EASTERN SIERRA LEONE

# EVD Labs in Sierra Leone



# OUTBREAK RESPONSE IN SIERRA LEONE 2014-2015 ( MAY)

| 16 INTERNATIONAL LABS | |
|---|---|
| Area | Labs |
| WESTERN | 9 |
| EASTERN | 1 |
| NORTHERN | 4 |
| SOUTHERN | 2 |

| 10 Countries | |
|---|---|
| Area | Labs |
| South Africa | 1 |
| USA | 2 |
| Canada | 2 |
| Italy | 2 |
| Germany | 1 |
| China | 2 |
| Netherlands | 2 |
| Nigeria | 1 |
| United Kingdom | 3 |

- There are small laboratories, designed specifically to handle viral haemorrhagic fever pathogens, with the capability to be deployed rapidly to the source of an outbreak.

- The equipment consists of one collapsible class three cabinet and two smart cyclers, which can all be transported on a normal commercial flight.

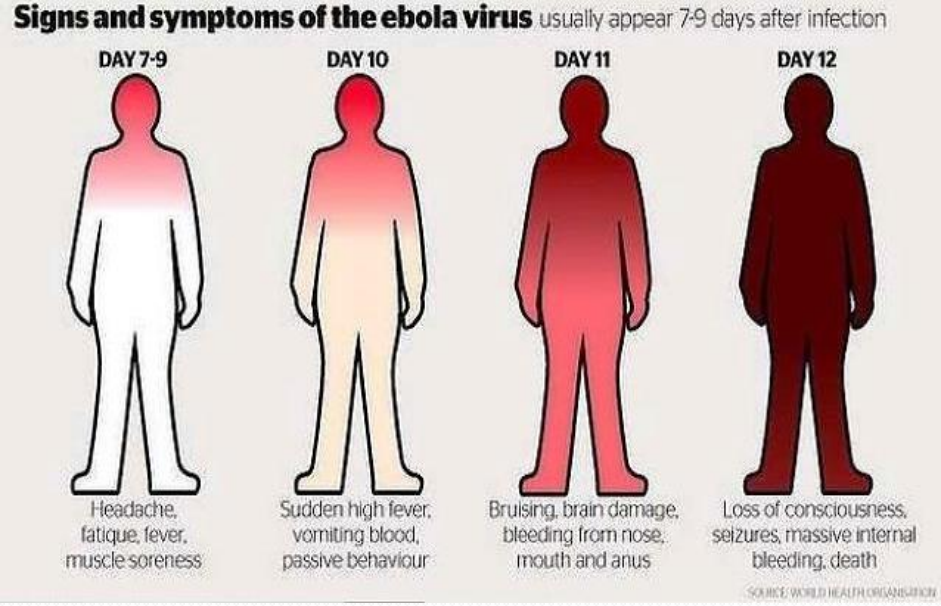
# ROLE OF LABORATORY IN THE OUTBREAK

1. Diagnosis of suspected cases satisfying case definition.
2. Testing of all corpses during the epidemic
3. Testing prior to discharge
4. Genomic sequencing
5. Viral persistence studies
6. Convalescent blood/plasma studies

# EBOLA VIRUS DISEASE CASE DEFINITION

1. Elevated by temperature or subjective fever or symptoms

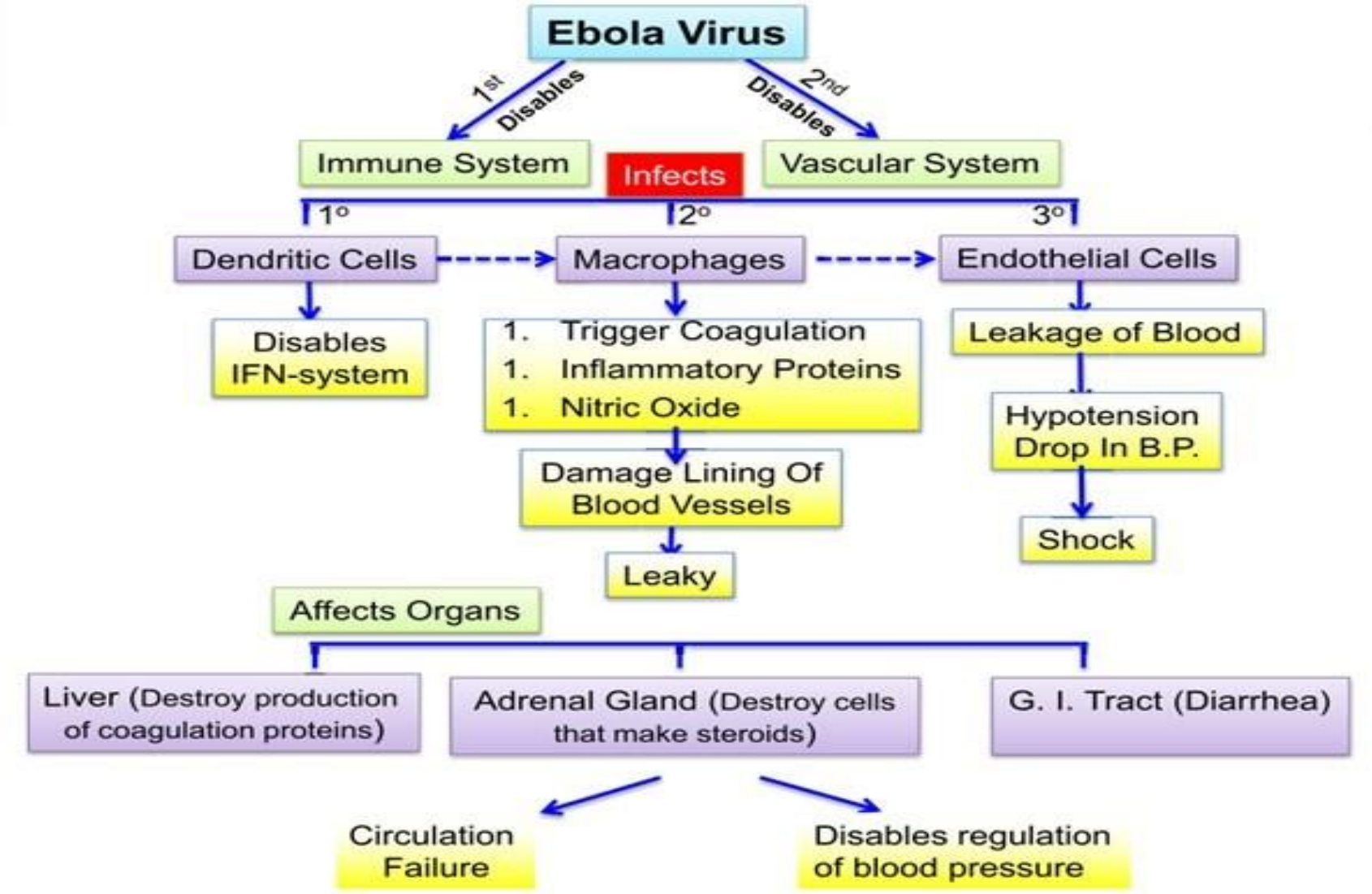
- Severe headache
- Fatigue
- Muscle pain
- Vomiting
- Diahorroea
- Abdominal pain
- Unexplained haemorrage




2. An epidemiological risk factor twenty (21) days before onset of symptoms

3. Symptoms mimic cholera ,malaria typhoid, Lassa fever which are epidemic in West Africa

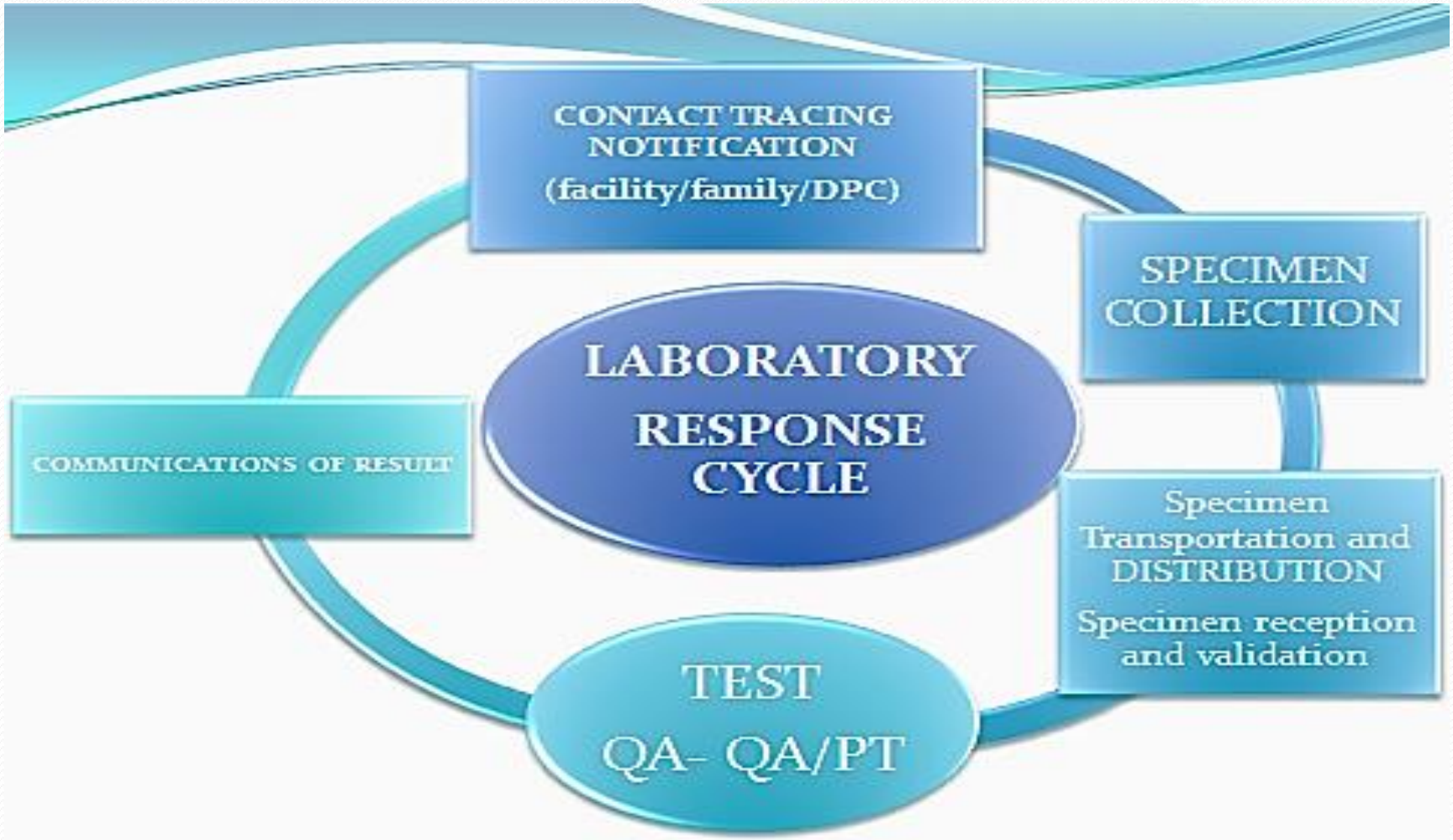
# PATHOGENESIS OF EBOLA VIRUS DISEASE



- The laboratory plays a critical role in combating the Ebola virus disease. Laboratory results are required for vital decision making such as quarantine of households and communities, treatment and discharge of patients.

- The work of these laboratories was facilitated by ensuring an efficient pre-analytical phase and post-analytical phase: collection, packaging and transport of samples and the dissemination of the result to various stake holders for action.

- The laboratory technical working groups in the affected countries in collaboration with the visiting teams served as the laboratory pillar to devise strategies to ensure an efficient laboratory response, build on successes, solve mitigating factors and challenges.


CONTACT TRACING
NOTIFICATION
(facility/family/DPC)
SPECIMEN
COLLECTION
LABORATORY
RESPONSE
CYCLE
COMMUNICATIONS OF RESULT
Specimen
Transportation and
DISTRIBUTION
Specimen reception
and validation
TEST
QA- QA/PT

PRE-ANALYTICAL

- **SPECIMEN MANAGEMENT**
- COLLECTION
- PACKAGING
- TRANSPORT

DIRECTION OF HOLDING and Treatment unit
CASE MANAGEMENT
SURVEILLANCE/trend

ANALYTICAL

- **TESTING LAB ALGORITHM**
- NICD
- KERRYTOWN
- CDC-USA
- CDC-CHINA

POST ANALYTICAL

- **RESULT DISSEMINATION**

# Laboratory Response Progress and Challenges 1

| Response Process Flow | Activity Achieved | Evidence | CHALLENGES/GAPS |
|---|---|---|---|
| 1) Specimen Management | -Develop specimen collection stock list and distribution protocol.<br><br>-Develop human resource plan and conduct training for all 4 regions. | -Trained specimen collectors and Laboratory Liaison Officers.<br>-National use of chain of custody.<br>-Mapping laboratory to facility to improve TAT. | **-Limited cold chain in remote regions.**<br><br>**-Lack of Bio-security and Bio-risk policy.**<br><br>**-Need Bio-Banking for specimen storage** |
| 2)Specimen Transportation | -Engage RSLAF to support specimen courier.<br>-Established travel routes. | -Improved specimen transit.<br>-Biosecurity of specimen. | **-Timely Vehicular maintenance.** |
| 3)Laboratory Testing Quality Assurance (QA) | -Implement 2 rounds of External QA<br>-Conduct 3 quality audits<br>-Conduct 1 Transition audit<br>-Usage of National Malaria testing protocol. | -Participation in WHO EQA PT<br>-Laboratory utilization of improved testing protocol.<br>-Malaria Parasite testing as part of management. | **-Independent Laboratory actions on testing protocol.**<br>**-Prioritisation of research studies.**<br>**-Shortage of local personnel being trained.** |

# Laboratory Response Progress and Challenges 2

| Response Process Flow | Activity Achieved | Evidence | CHALLENGES/GAPS |
|---|---|---|---|
| **4) Result Dissemination** | **-Establishment of Laboratory unit at Command Centre.**<br>**-Development of drop down result template.**<br>**-Development of result reporting 1st loop to facility.** | -Harmonised reporting result template.<br>-National leadership in result analysis. | **-Uncontrolled result sharing breaching patient confidentiality.** |
| **5) Coordination** | **-Establishment of laboratory Technical working group. Weekly technical team meetings**<br>**-Coordination of training**<br>**-Development of laboratory transition to MOHS**<br>**-Linkage with case management on facility operations**<br>**-World Bank funding for laboratory services through WHO.** | -Lab Coordinating centre OFFICE at NERC<br>-Supportive supervision to define training needs. Phased closure of laboratories in line with case management scaling down.<br>-National Transition plan developed in line with health sector plan and MRU initiatives. | **-Limited support for supportive supervision.**<br>**-Need to review MOU of laboratory partners.**<br>**-Unilateral actions of laboratory partners.** |

# TRAINING

- Human resource training plan were designed and rolled out nation wide with focus on reduction in rejection rates of samples collected and reduction of turn around times.

- Moreover there was an urgent need to implement infection, prevention and control, training and undertake monitoring and support supervision in order to reduce mortality amongst laboratory workers.

# TRAINING OF SWABBERS DURING THE EBOLA EPIDEMIC

# DOFFING AND DISPOSAL OF PPE

# LABORATORY COORDINATION MEETING

# OPENING CEREMONY: TRAINING OF RAPID RESPONSE TEAM

# PRINCIPLES OF EBOLA DIAGNOSIS

- Clinical features of Ebola mimics malaria, typhoid fever, cholera and Lassa fever
- Confirmation by laboratory diagnosis
- PCR done within the 1st 72 hours to prevent false positive results
- Virus is detected readily in oral swabs of the diseased because of high viral load.
- Patients are discharged after two negative PCR results 72 hours apart
- In survivors, IgM levels rise after two weeks followed by IgM

# LABORATORY DIAGNOSTIC METHODS

- Quantitative PCR tests were used for diagnosis of Ebola.
- 
- These tests targeted mainly the conserved domains of the Ebola virus zaire gene, and structural elements: l , NP, VP40, GP.
- The kits used included Altona RT-PCR kits (Altona diagnostics GmbH, Hamburg, Germany) the DOD ez1 real-time RT-PCR, CDC's QRT-PCRs for the viral NP and VP40, and the biomerieux biofire film array assay (Biofire Defense, LLC, Salt lake city, Utah).
- Followed by Cepheid Xpert Ebola assay on the GeneXpert platform fevers ( Cepheid ,Sunny vale California}

# RAPID DIAGNOSTIC TESTS

- Rapid diagnostics tests for point of care testing for the Ebola were developed towards the end of the epidemic.

These include:

1. THE LATERAL FLOW ANTIGEN-CAPTURE
   LATERAL FLOW ANTIGEN –CAPTURE ASSAY
   (Corgenix, Inc Broomfield, Colorado , USA)
2. THE ORAQUICK EBOLA ANTIGEN TEST(Orasure Technologies, Inc, Bethelem, Pennsylvania,)

- Validation of the cogenix test in Sierra Leone revealed that it is as sensitive as gold standard, RT-PCR in diagnosing Ebola (Broadhust et al,2015).

- Test kits were never deployed during the epidemic for diagnosis of Ebola.
- These kits could be deployed in future outbreaks to triage suspected patients although positive and highly suspicious negative cases should undergo confirmation.
- Several pathogens can be captured on a single strip and could be useful for differential diagnosis for pathogens with similar clinical presentation. (E.G., Malaria parasites, Marburg virus, and Lassa fever virus).
- Challenges include waste management triage clinical staff and quality assurance

# GENETIC SEQUENCING DURING THE OUTBREAK

- Early sequencing was done by Gire et al at Harvard university in collaboration with the ministry of health and sanitation , Sierra Leone.
- 99 virus genomes from 78 patents diagnosed within the first three weeks of the outbreak were sequenced . Both inter host and intra host genetic variation and leading to elucidation of the origin and the determination of patterns of viral transmission in the initial weeks of outbreak in Sierra Leone
- The west African variant ( Makona) diverged from a central African strain a decade ago ,with zoonotic transmission in guinea and crossing into Sierra Leone at a funeral of a traditional healer who was infected in guinea.
- No further evidence of zoonotic transmission was discovered.

- Further analysis of 232 patients in Sierra Leone over the initial 7 months of import of EBV across national boarders including 86 previously released revealed genomes in the epidemic confirmed sustained human to human transmission with no evidence of export across national boarders (Park et al, 2015)

- Researchers in Liberia also linked majority of the cases to single introduction from neighboring countries and sustained human to human transmission. Thus confirming the use of sequencing in the determination of transmission chains and enhancing surveillance .(Ladner et al )

- Sequencing systems have been installed in the three most affected countries to undertake viral persistence studies , others including the documentation of modes of transmission in semen and breast milk.(Armando et al 2016,)

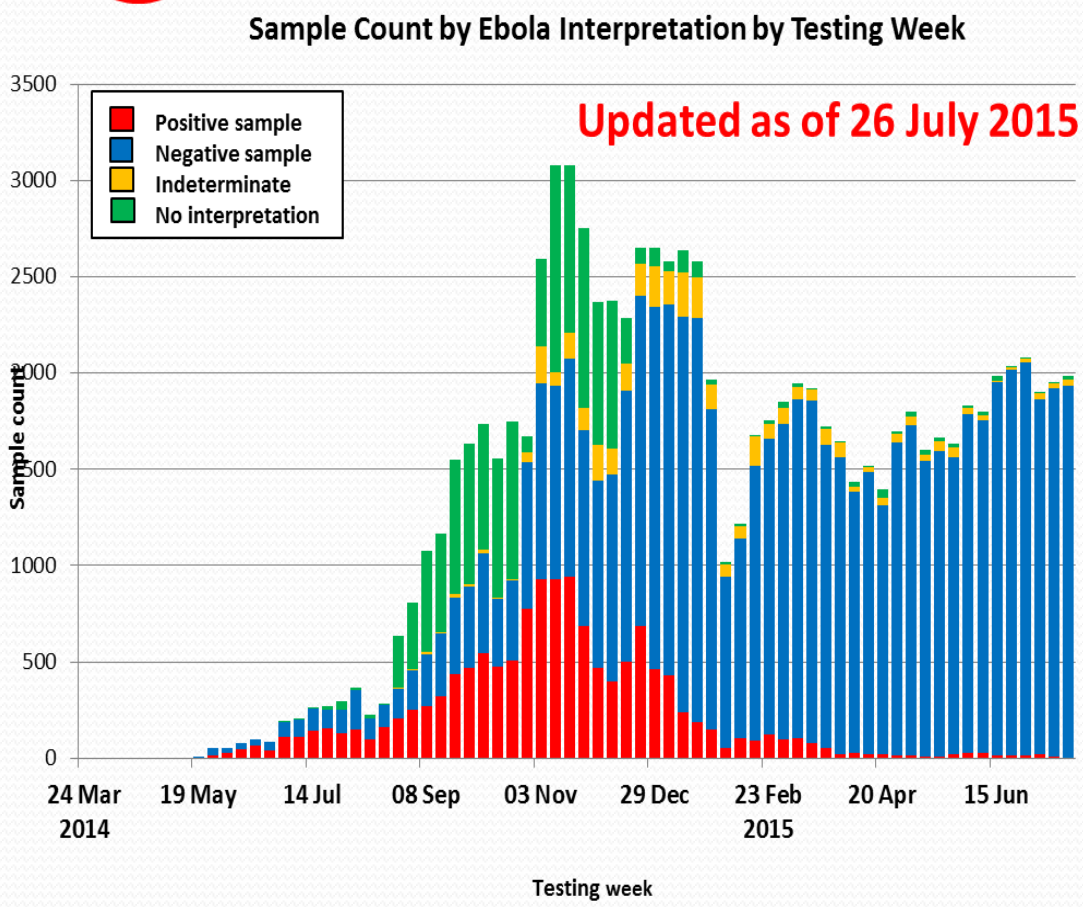
# BIOBANKING AND BIOSECURITY

- Samples of varied nature were collected in all West African countries
- These included  mainly swabs from corpses and blood samples
- There are still many samples in facilities that do not have appropriate levels of biosafety and biosecurity
- Sierra Leone government in collaboration with global emerging  pathogens consortium are in collaboration to secure these samples in an inventoried repository with adequate biosecurity measures.
- Inventories of samples shipped out of Sierra Leone is being undertaken with a view to creating offshore biobanks. Public Heath England is  taking  the lead

# Total Number of Specimens Tested

EDPLN

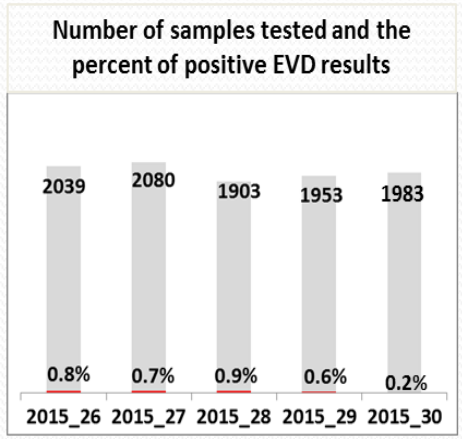
| No. Labs | No. of Samples | Total of Positive | Percentage Positive |
|---|---|---|---|
| 15 | 92,698 | 13,488 | 14.6% |




China-CDC Lab- Freetown
CPHR/DTRA - Freetown
Dutch Mobile lab – Kono
Dutch Mobile lab - Freetown
EU Mobile lab - Hastings
EMDF/NICD - Freetown
Nigeria Mobile Lab - Kambia
PH England Lab - Bombali
PH England Lab - Port Loko
US-CDC Lab – Bo
Italian Lab - Goedrich
Nigeria Mobile Lab – Freetown
PHA Canada - Kailahun
PHA Canada – Tonkolili
US DTRA – Moyamba

*For last five weeks:*

Average number of samples tested per week: 1992



# OBJECTIVES

- TO MAINTAIN AND SECURE A CENTRAL QUALITY BIOLOGICAL SPECIMEN REPOSITORY TO SUPPORT PUBLIC HEALTH RESEARCH DEVELOPMENT.
- TO STORE ALL EBOLA SPECIMENS SAFELY AND SECURELY
- TO DEVELOP FUNCTIONAL INVENTORY AND A SYSTEM FOR SPECIMEN MANAGEMENT SYSTEM (COLLECTION, TRANSPORTATION, ARCHEIVING AND RETRIEVAL OF STORED SPECIMENS (LABORATORY INFORMATION MANAGEMENT SYSTEM).
- TO DEVELOP POLICIES FOR DESTRUCTION, ACCESSING RETRIEVAL AND UTILIZATION, DISPOSAL OF STORED SPECIMENS
- ESTABLISH A NATIONAL GOVERNING BODY OVERSEEING BIO BANKING.
- ESTABLISH AND STRENGTHEN CAPACITY FOR MANAGEMENT, COORDINATION OPERATION AND MAINTAINING OF THE BIO BANKING.

# Three CP Studies in West Africa

**Ebola_Tx**

- Johan Van Griensven et al.
- ITM Antwerp + 15 others
- MSF Conakry Guinea

**EVD001**

- David Hoover et al.
- Clinical RM/Duke/UNC
- ELWA2 Monrovia Liberia

**Ebola_CP**

- Sahr Gevao, Calum Semple et al.
- Liverpool & 15 others
- MoHS & 34th Regiment Hospital Freetown

# RESULTS

- Griensven et al (2016) NEJM 374:33-42 transfusion of 84 patients with low levels of neutralising antibodies documented no improvement in survival.

- Plasma studies in Sierra Leone and Liberia suspended because of decrease in patient numbers

- Results of convalescent blood studied in Sierra Leone more encouraging and sent for publication.

# RECOMMENDATIONS

- Mobile laboratories donated to the affected countries should be integrated into the public health response systems to build indigenous capacity and research capability for future outbreak response to Ebola and other VHFS.
- Replicate the Sierra Leone biosecurity model in Guinea and Liberia to create functional on and off shore biobanks .
- Integrate training on biosecurity, IPC measures ,diagnosis and control of VHFS in training curricula of health personnel.
- Leverage on collaboration built with international organisations and agencies during the outbreak to build long term partnerships to prevent a recurrence of this human tragedy.
- Build a robust surveillance system and health infrastructure to detect and respond early to VHF and other epidemics.

# ACKNOWLEDMENT

1, WEST AFICAN COLLEGE OF PHYSICIANS EXPRET COMMITTEE

- Arising from recognition of the scale of the morbidity and mortality from outbreak, WACP in July 2014 constituted a multidisciplinary expert committee to advise it on the multifaceted challenges and issues related to the control of VHFS in West Africa.

- The committee had a broad mandate that encompassed emergency response to the on-going outbreak as well as preparedness for future outbreaks of VHFS.

- Made recommendations to country chapters on the constitution of Ebola response committees, and the development of response plans for the guidance of affected and at-risk countries

2. LABORATORY TECHNICAL WORKING GROUP, SIERRA LEONE.

# REFERENCES

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2 Broadhurst et al ( 2016)ReEBOV Antigen Rapid Rest Kit for point-of-care and laboratory testing for Ebola virus disease , A field evaluation study . Lancet Aug 29:386(9996)867-74

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5,Park et al (2015) Ebola Virus Epidemiology , Transmission and Evolution during the seven months in Sierra Leone . Cell 1516-1526 161(7)

# THANK YOU
# FOR LISTENING