

PROPOSED FORENSIC INVESTIGATION OF WUHAN LABORATORIES

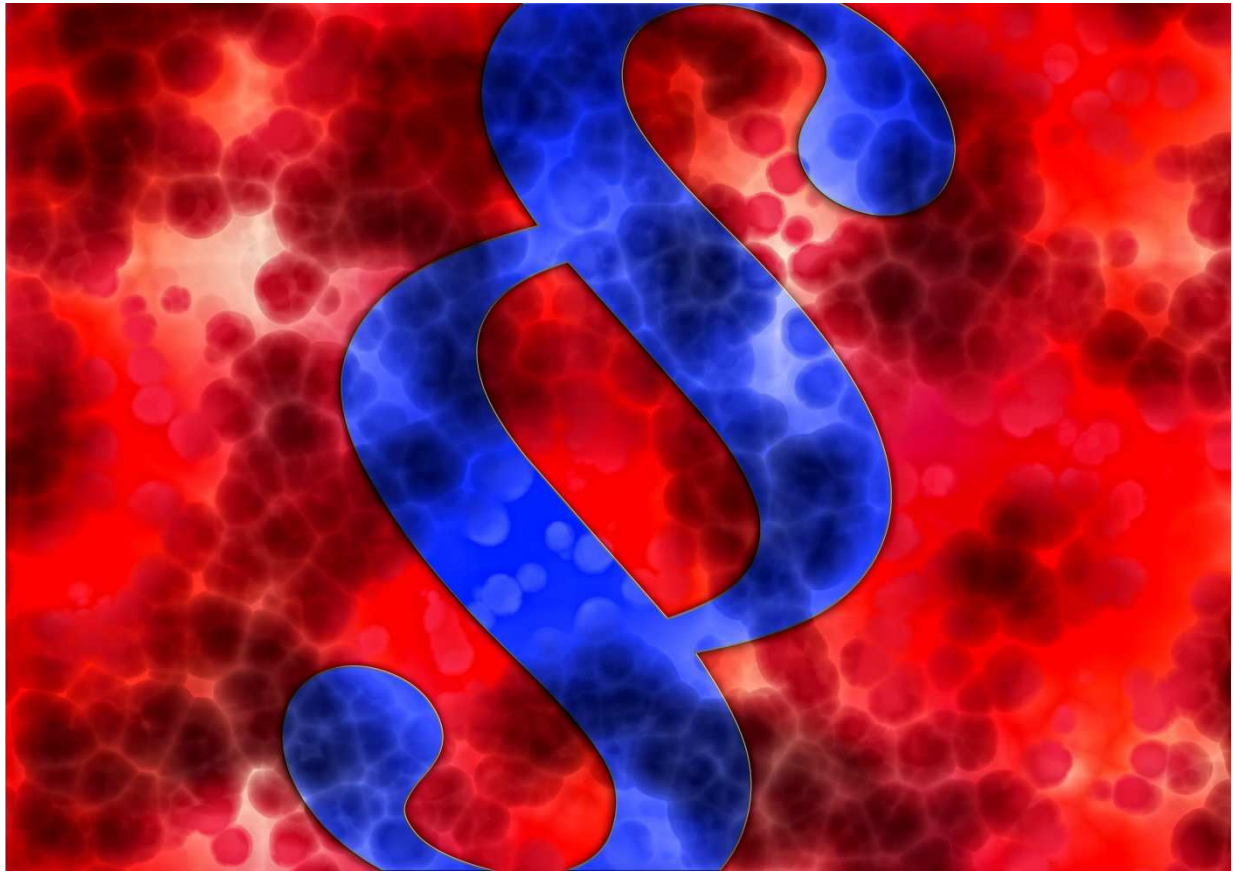


Image provided generously by Piqsel

WILLIAM BOSTICKSON AND YVETTE GHANNAM (2021)

This is the **first part of a series of reports** based on our previously unpublished investigations into the origins of SARS-COV-2. We wish to thank all the independent researchers who have contributed to this investigation, especially members of the DRASTIC Collective, many of whom wish to remain anonymous for reasons of security and privacy.

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A. BACKGROUND

It has become increasingly clear to many observers that the international community lacks transparent evidence from the World Health Organization (WHO), the Chinese authorities and laboratories operated by the Wuhan Institute of Virology (WIV) amongst others, to clarify certain hypotheses about the origins of SARS-CoV-2.

When there is a public perception in the US, that more than **500,000** people have died, but public health organizations do not seem to care enough to investigate the exact origin of this pandemic, then surely it must become the responsibility of law enforcement agencies to conduct an unbiased investigation on behalf of US citizens.

Concern has been raised that research scientists in China were carrying out unsafe research activities on SARS-CoV like novel viruses and in one way or another did not take the appropriate actions to work safely with SARS-CoV-2 or a precursor virus (Cohen & Kupferschmidt, 2020; Subbaraman, 2020).

With the advent of technological tools related to genomic manipulation, federal agencies need to update their protocols to investigate war crimes and bioterrorism. The new tendency of using biological weapons is related to the field of virology, which is one that few law enforcement officers have training in, since it is a complex scientific field. It would be devastating to US citizens, the US national economy and US security to allow foreign nations to dominate the US without a physical war through asymmetric warfare and deception involving viral pathogens. It is proposed that an international team of experts in biodefense visit the Wuhan Laboratories and conduct a clean, fair and un-biased investigation into the origins of SARS-CoV-2.

However, at this time, there is no agency in the United States that can fully deal alone with the weaponization of biological warfare agents (BWAs) or even biological infections naturally occurring in areas of genetic engineering, synthetic biology techniques, and complex genetic manipulations as currently may be the case with SARS-CoV-2. (Konda et al., 2020; Sharma, et al., 2020). If genetic manipulation actually did occur, then it will be extremely difficult to prove it without access to the relevant local databases and laboratory records of experiments involving genetic manipulation.

Genomic database investigations have traditionally been in the hands of health professionals (Kulynych & Greely, 2017), however, most health professionals with expertise in these areas of knowledge do not feel comfortable participating in warfare or weaponization investigations, especially if it is linked in any way to their research, due to fear of losing their jobs or funding to continue pending research studies.

The FBI, a pioneer agency in conducting DNA testing (2020), matches a person's DNA to one of the largest DNA databases named the **Combined DNA Index System (CODIS)** for a particular subject. Unfortunately, the FBI does not have as yet databases to investigate genomic manipulation which would be extremely useful for investigations where perpetrators may have used animal vectors to develop a bio agent to be subsequently used against humans (if this was the case, for example with SARS-CoV-2). Additionally, the FBI would need time to train their staff to be proficient in working with novel genomic databases.

Despite the difficulties involved, researchers have a responsibility to carry out a thorough analysis of all possible SARS-CoV-2 origins to the best of their abilities. This is especially important in light of the tragic human cost of SARS-CoV-2 and in order to better prevent future pandemics.

Thus, the question of the natural or synthetic origin of SARS-CoV-2 deserves to be examined in detail based on available evidence. It would be important to know exactly what types of genetic manipulations were carried at all Wuhan Laboratories. This is not to suggest that all genetic manipulations are worthy of suspicion. Indeed, genetic manipulations (i.e. Ace binding on the S spike, furin cleavage, furin trimerization motifs, ORFs, binding sites, RBD residues sites, RGD and LDI integrin binding) help scientists to understand the mechanism of replication and the emergence of viruses and their use for the development of vaccines (or antivirals) (Baric, 2006; Coutard et al., 2020; Vankadari, 2020).

On January 15, 2021, the US Secretary of State issued a press statement criticizing China's lack of transparency and lack of cooperation with the WHO, calling for a *"transparent and thorough investigation into the origin of COVID-19"* ([United States Department of State, 2021a](#)). The statement claimed that:

*"The U.S. government does not know exactly where, when, or how the COVID-19 `virus—known as **SARS-CoV-2**—was transmitted initially to humans. We have not determined whether the outbreak began through contact with infected animals or was the result of an accident at a laboratory in Wuhan" which "could resemble a natural outbreak if the initial exposure included only a few individuals and was compounded by asymptomatic infection. Scientists in China have researched animal-derived coronaviruses under conditions that increased the risk for accidental and potentially unwitting exposure"*

The statement also included a fact sheet (United States Department of State, 2021b), detailing intelligence findings concerning the Wuhan Institute of Virology, which mentioned:

1. Sick WIV researchers with COVID-19 like symptoms in the fall of 2019, before any confirmed cases.
2. A lack of transparency concerning WIV gain of function research on RaTG13 or similar unpublished viruses dating back to 2016.
3. WIV classified research projects involving experimental laboratory animal studies in collaboration with secret Chinese military projects since 2017.

The statement concludes that China “continues today to withhold vital information that scientists need to protect the world from this deadly virus, and the next one” and “reiterates the importance of unfettered access to virus samples, lab records and personnel, eyewitnesses, and whistleblowers to ensure the credibility of the WHO’s final report” (United States Department of State, 202a).

The statements demand that WHO investigators must enjoy “complete, transparent access to the research labs in Wuhan, including their facilities, samples, personnel, and records” and that if the investigation into the origin of SARS-COV-2 is going to achieve public credibility, then:

“WHO investigators must have access to the records of the WIV’s work on bat and other coronaviruses before the COVID-19 outbreak. As part of a thorough inquiry, they must have a full accounting of why the WIV altered and then removed online records of its work with RaTG13 and other viruses.” (United States Department of State, 202b).

In light of the above, the authors of this report propose nothing more nor less than a transparent and open investigation into the activities of laboratories in Wuhan in order to ascertain whether or not SARS-COV-2 emerged due to their sampling of bat *betacoronaviruses* and experiments performed on these deadly pathogens over the last decade at BSL laboratories in China and the USA.

B. PROPOSED INVESTIGATION

1. FBI HANDBOOK SUGGESTIONS

In the 2011 and 2018 FBI, CDC and DOJ handbook used by the FBI's WMD Directorate, WMD Biological Countermeasures Unit and WMD Operations Unit, essential elements of any criminal investigate procedure necessary in the event of bioterrorism include gathering of evidence, chain of custody issues, the need for authenticated original documents and accurate translations thereof, as well as witness statements. (DOJ & FBI, 2011, p. 33). The handbook emphasizes that while investigating bioterrorism, investigators are unable to predict exactly which "nuance or piece of information will be the crucial break needed to identify, arrest, and convict those responsible for the criminal act" (DOJ & FBI, 2011, p. 36). Thus, especially in the case of a foreign country such as China, intelligence gathering and secure storage of a wide range of data is of the utmost importance.

In "A Guide to Investigating Outbreak Origins: Nature versus the Laboratory", Dr. Richard Pilch , Director of the CBWN Program at the James Martin Center for Nonproliferation Studies (CNS) at MIIS and Filipa Lentzos warn of the growing danger of laboratory accidents due to the expansion of BSL Laboratories and the increase in "higher risk experiments". They conclude that "a deliberate biological attack may resemble an outbreak of natural or accidental origin, and a natural or accidental outbreak may be misattributed as an attack" Pilch et al. (2020).

2. INVESTIGATION METHODOLOGY

In their recent paper (Pilch et al., 2020) propose a step-by-step methodology to investigate the origins of SARS-COV-2, which is rooted in tried and trusted epidemiological principles based on case studies of previous outbreaks, namely: the 1979 anthrax outbreak in Sverdlovsk, the 1993 hantavirus outbreak in the US and the 2007 Foot-and-mouth disease at the Pirbright Institute in the UK in 2007. Based on these cases, Pilch et al. (2020) break down the methodology needed to investigate outbreak origins, into the following discrete steps, illustrated in Figures 1 and 2 below:

- Descriptive and Analytical Epidemiology
- Assessment of the “Epidemiological Triangle” and the Genome itself
- Further Analysis of Samples
- Assessment of the overall Laboratory Risk by determining:
 - ✓ the aims of the laboratory research,
 - ✓ samples or specimens stored at the laboratory
 - ✓ the nature of any high risk experimental studies

Step 1. Descriptive Epidemiology

- Multiple, geographically-dispersed index cases are identified
- Infecting agent is a traditional biological warfare agent
- Infecting agent is unusual for location or time of year
- Symptoms are unusual or unexpected (e.g., pulmonary symptomatology)
- Animal populations are affected in concert with humans
- Animal effects are unusual or unexpected for the species

Step 2. Analytical Epidemiology

(a) Epidemiological Triangle Assessment

- Lack of recognizable animal-human interface
(e.g., exposure to sick animal, tick bite)
- Epidemiological traceback of multiple cases to a common location or exposure

(b) Genome Assessment

- Infecting agent genome matches known weapons strain
- Infecting agent genome displays “frozen evolution”
- Infecting agent genome has been engineered / edited

Figure 1: Investigation Methodology A (Page 22). (Pilch et al., 2020)

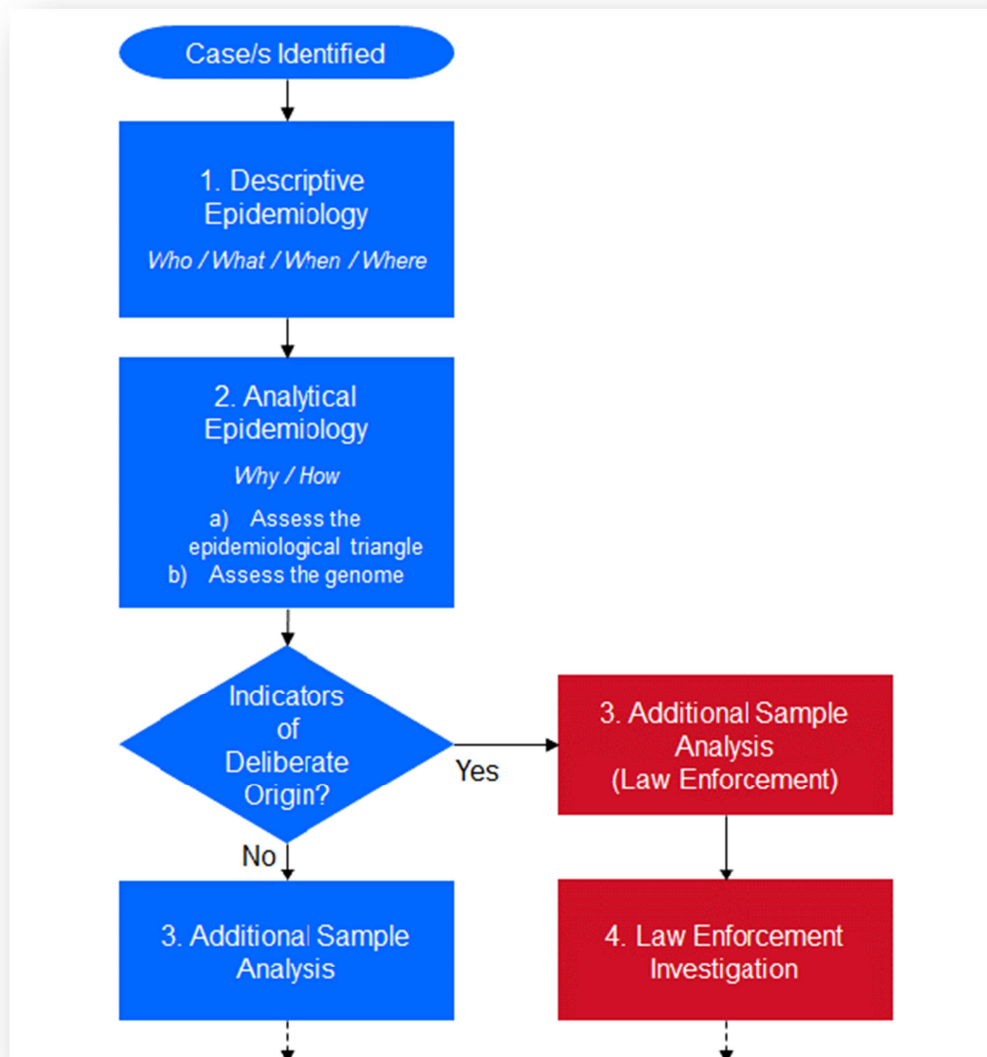


Figure 2. Investigation Methodology B (Page 23) (Pilch et al., 2020).

Once the overall laboratory safety profile has been determined, investigation would proceed to On-Site Laboratory Assessment in order to assess the probability of a deliberate outbreak versus an accidental one. Pilch et al. (2020) conclude that if the Laboratory Risk Assessment shows the need for deeper investigation, then a “comprehensive onsite assessment” would be required in line with WHO International Health Regulations (IHR) 2.0 policies.

This would involve unrestricted access by an international investigative team to the laboratory in question, its safety records and its staff, including details of ‘sample receiving and accessioning logs’ (Pilch et al., 2020). Further items to be investigated would include but not be limited to those illustrated in the following image from their recent report (Figure 3):

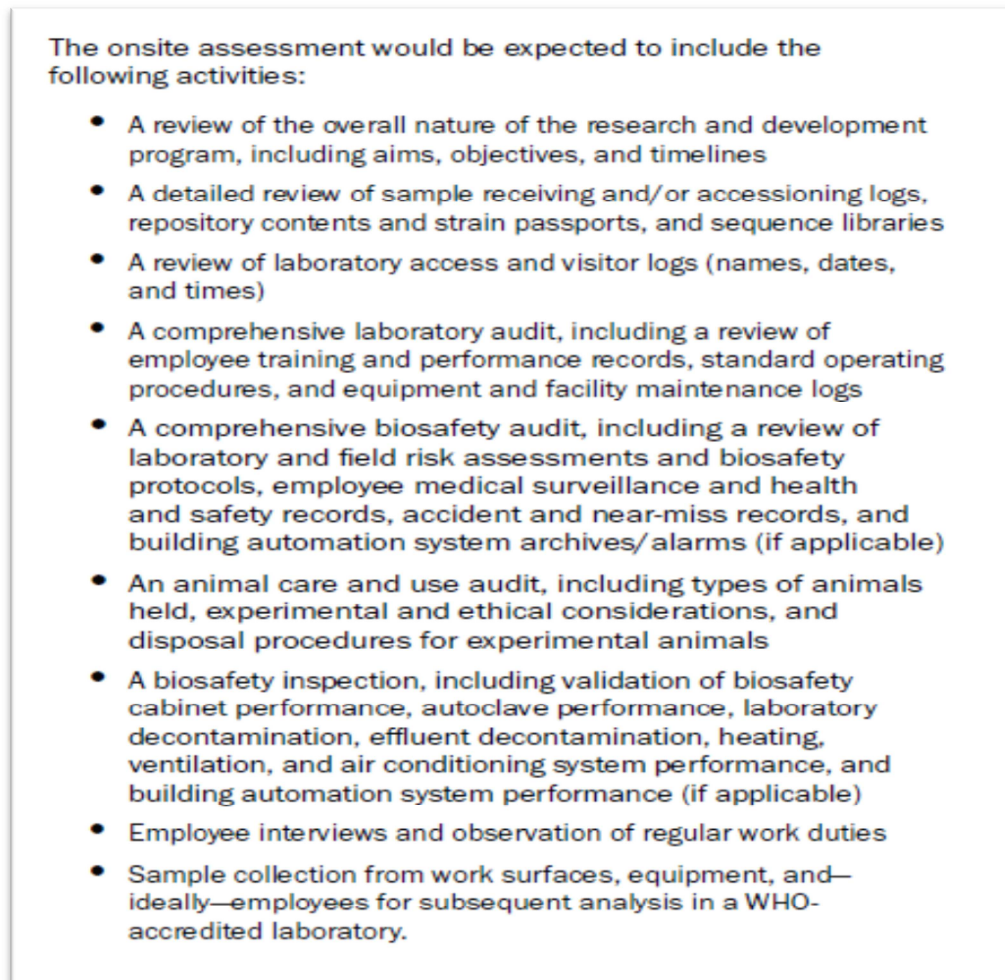


Figure 3: Onsite Assessment (Page 19) (Pilch et al, 2020)

Overall, Pilch et al (2020) provide a clear and effective strategy for investigating a possible laboratory leak, whether accidental or deliberate from laboratories in Wuhan, as illustrated here: (Figure 4).

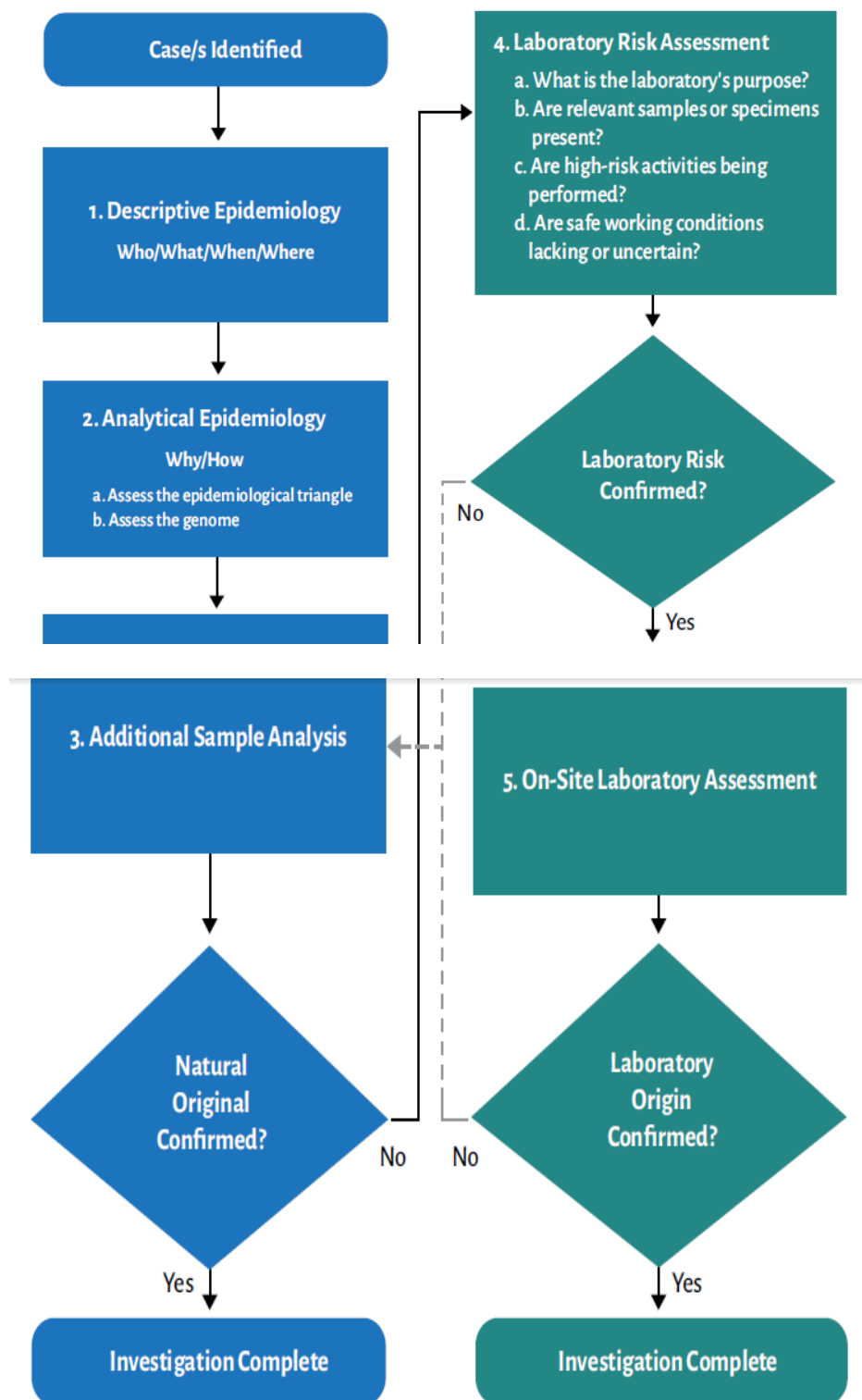


Figure 4: Investigation Flowchart: (Page 5) (Pilch et al, 2020).

3. BIOSAFETY QUESTIONNAIRE TOOLS

4. FAO LMT

The FAO Food and Agriculture Organization of the United Nations (FAO) published a Laboratory Safety Mapping Tool (LMT) in 2016 that allows laboratory safety data to be captured by external investigators (Figure 5) or via independent self-assessment over 1-2 days (FAO, 2017).

On-site LMT assessment protocols

- **Objective**
 - Understand the lab
 - Understand the staff
- **Time**
 - 1-2 days
- **Method**
 - Meetings with staff
 - Onsite lab visits
- **Output**
 - LMT assessments
 - Formal report



Figure 5: On-Site LMT Visit (Blacksell, 2012)

For any forensic investigation of laboratories in Wuhan, these tools would be an excellent starting point to help map areas of concern. The first questionnaire to be used would be the LMT Core tool (**Appendix 14: FAO Laboratory Mapping Tool-Core LMT-Core**) which would help to effectively evaluate the laboratory operation environment.

5. FAO LMT-S

Secondly, the safety LMT module (Appendix 15: FAO Laboratory Mapping Tool-Safety Module LMT-S) focuses on the environmental safety of laboratories and the occupational risks.

Both of these questionnaires can be used either as a hand completed excel file (Figures 5b and 5c) or as a mobile application (Figures 5d and 5e) to generate a “map” of BSL risk factors (FAO, 2017).

FAO Laboratory Mapping Tool (LMT)

Excel file

A: Date: 09/09/11 Name assessor A: Assessor's affiliation: NYL Self assessment Y

B: Date: 06/02/12 Name assessor B: R. Pin Dlop Assessor's affiliation: FYI Self assessment N

C: Date: 25/06/11 Name assessor C: Laurence Mleout Assessor's affiliation: FYI Self assessment N

Laboratory: /BMT Address/contact details/phone: Rwanda

Lab affiliation: ☐ Public/Government ☐ Private ☐ University ☐ Other

Main lab activity: ☐ Sub-national (District or Provincial) ☐ National ☐ Regional ☐ Not applicable

Lab admin level: ☐ Check here if O/E and/or FAO or other Partnership or Collaborating Centre

For each of the 108 subcategories, one out of four options can be selected

Scores

SELECT ONLY 1 OPTION per row THAT BEST DESCRIBES THE SITUATION and complete "Assessments Scores" for assessment C (Current assessment) scoring either 4,3,2 or 1 (there are 3 columns for 3 different assessments: previous A, previous B, current C), or check "no info" (column J) if not available.

If situation stands between two scores, please select one score and describe the reason for hesitation in the column for comments (column K).

If a given score is 1/-2 or 3 compared to the previous score, please provide a comment (column K).

| # | Category | Assessments scores | | | | K Comments |
|---|---------------------|---|--|--|---|---------------|
| | | A | B | C | Score | |
| 1 | Geographic location | Isolated compound outside of an residential area | Isolated compound in low populated area | Single building in low populated area | Building within residential area | 4 3 2 1 |
| 2 | Geographic location | Proper conservation + good (24 h) + Restricted access to building by way of fencing and/or physical site | Restricted access, doors are locked + guarded at the entrance for 24 h | Doors are closed but not locked + not guarded by post + guard is not always present | Easy access to laboratory compound even by night + no guard / doors are open + no guard present | 4 3 2 1 |
| 3 | Geographic location | Access to highway, airport, harbour or station within 30 minutes, or helicopter service on-site | Access to highway, airport, harbour or station within 60 minutes | Access to road, but sometimes involves (traffic, road condition, flooding) | Regular initiative in service to transport means (traffic, bad road, support in trip) | 4 3 2 1 |
| 4 | Laboratory budget | Lab is financially autonomous, lab funds from public source or self-generated | Lab is almost financially autonomous, lab funds from public source or self-generated (50%) AND development programme | Lab has no financial autonomy, lab funds from public source or self-generated (25%) AND dependent on development partner (40%) | Lab has no autonomous budget situation, exclusively dependent on external funding source | 4 3 2 1 |
| 5 | Laboratory budget | Lab budget allows ample opportunities for research (10 public/microscopy) budget routine, equipment maintenance | Lab budget allows a little research (5 to 10 public/microscopy) research (in the lab context), but mainly routine | Lab budget is insufficient for research, but results from ongoing work is published in national journals/bulletin or regularly presented | No research activity, no publication, no bulletins or regularly presented | 4 3 2 1 |
| 6 | Laboratory budget | Lab budget sufficient for | Lab budget sufficient for | Lab budget sufficient for | Lab budget sufficient for | 4 3 2 1 |

Scores for the 5 areas, the 18 LMT categories and an overall score will be automatically calculated and graphics generated in the "Summary"

Figure 5b: FAO Laboratory Mapping Tool (LMT) (Blacksell, 2012)

Example LMT Excel file

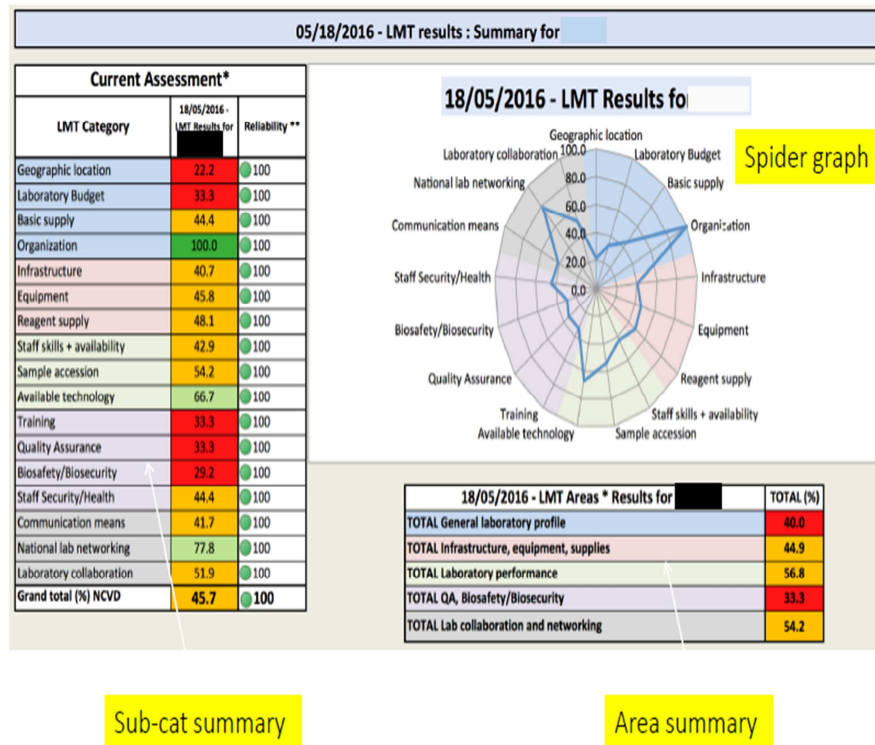


Figure 5c: “Map” generated from Excel LMT questionnaire (Blacksell, 2012)

LMT App - Some screenshots

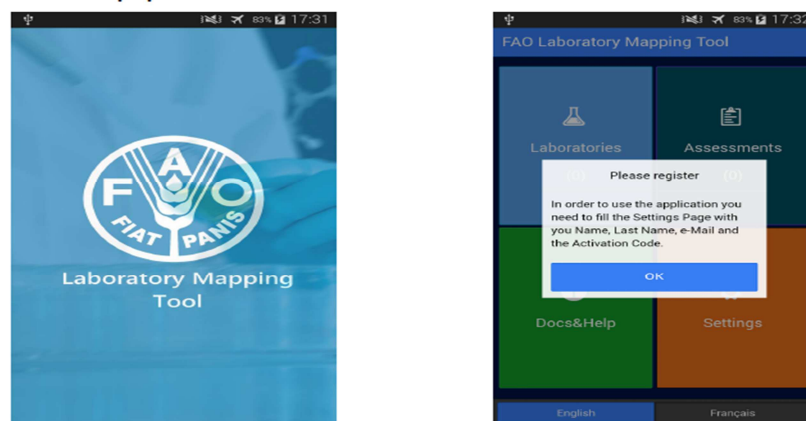


Figure 5d: Screenshots of the LMT Biosafety App (Blacksell, 2012)

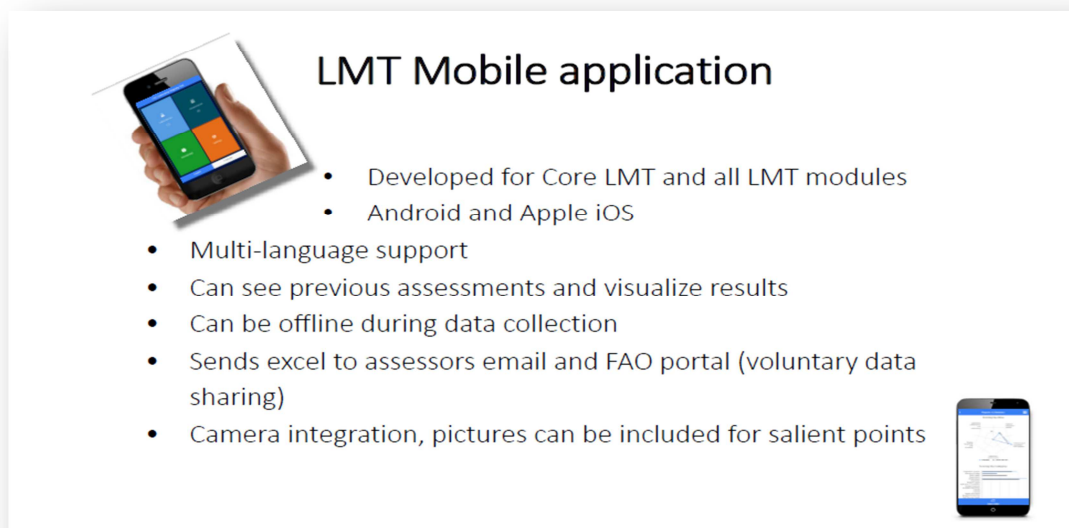


Figure 5e: LMT Mobile Application (Blacksell, 2012).

6. QUANHDIP (ECL BIORISK)

Another useful laboratory biosafety assessment tool is called QUANDHIP (Appendix 13: Quality Assurance Exercises and Networking on the Detection of Highly Infectious Pathogens). This was based on input from 29 BSL3 laboratories and six BSL4 laboratories in Europe (Appelt et al, 2020). The questionnaire tool (**Appendix 13**) is designed to assess potential gaps that could lead to safety and security breaches at BSL Labs (Lloyd et al, n.d).

This “Integrated European Checklist for Laboratory Biorisk Management in Handling of High Consequence Risk Group 3 and 4 Agents” (ECL-Biorisk) checklist is freely available for download and use (EMERGE – QUANDHIP, 2016), and enables BSL3 and BSL4 laboratories to carry out an external or internal assessment of biocontainment requirements by checking ECL list items which cover dedicated to 14 Bio Risk categories (EMERGE – QUANDHIP, 2016):

“to identify risks and introduce infrastructure and operational controls that prevent unintentional and intentional release of pathogens” EMERGE - QUANDHIP. (2016)

7. EPIDEMIOLOGICAL ASSESSMENT TOOL

Grunow & Finke (2002) also developed an epidemiological assessment tool to identify or rule out BW in the event of an unusual infectious disease outbreak, using specific criteria. Dembek et al, (2006) examine the Grunow & Finke (2002) epidemiological assessment tool, and discuss “epidemiological indicators that should be considered during outbreak investigations” and how to apply these indicators during:

- bioterrorism incidents
- accidental release of a pathogen
- outbreaks of infections alleged to have been deliberately initiated,

The assessment tool was designed to provide a retrospective epidemiological analysis of ‘unusual’ epidemics and the politico-military, socioeconomic, medical, epizootological, epidemiological and environmental situation in the outbreak region”. It also helps investigators evaluate the probability of a natural or artificial outbreak by analyzing the 11 listed criteria (Dembek et al, 2006). The 11 criteria to be evaluated are as follows (Grunow & Finke, 2020) and are quoted verbatim:

1. Biorisk.

“Are BW agents available, with the means for distribution, and the will to use them? Or can an outbreak be explained by natural biological hazards?”

2. Biothreat.

“Does a biological threat exist with a group possessing a BW agent and threatening to use it?”

3. Special aspects.

“Is there plausible evidence of deliberate pathogen manipulation?”

4. Geographic distribution.

“Is the disease’s geographic distribution probable given its locale? With the advent of a non-endemic pathogen, a thorough evaluation should include epidemiological, epizootic, ecological, microbiological and forensic analysis.”

5. Environmental concentration.

“Is there a high environmental concentration of the pathogen?”

6. Epidemic intensity.

“Is the course of illness relative to disease intensity and spread in the population expected in naturally occurring illness?”

7. Transmission mode.

“Was the path of disease transmission naturally occurring? The appearance of a naturally occurring epidemic alone does not rule out deliberate use of a biological agent.”

8. Time.

“Was the calendar time of the epidemic abnormal?”

9. Unusually rapid spread.

“Was the spread of the epidemic unusually rapid?”

10. Population limitation.

“Was the epidemic limited to a specific (target) population? (If certain individuals had forewarning of a BW attack, they might protect themselves, as compared to naïve target populations.”

11. Clinical.

“Were the clinical manifestations as expected for the disease?”

During the initial stage of any forensic investigation into laboratories in Wuhan, all four of the evaluation tools mentioned above (FAO LMT, FAO LMT-S, QUANDHIP –ECL BioRisk, The Grunow & Finke (2002) epidemiological assessment criteria) would be useful adjuncts to the proposed investigation methodological framework developed by Pilch et al. (2020).

8. QUALITY CONTROL AND ASSURANCE ASSESSMENT

Specifically, the putative laboratory leak or lab acquired infection hypothesis can best be explored by getting access to the Human Resources (HR) files of the employees, blood samples of laboratory workers for antibody tests, and the laboratory supply records and their relationship to the Wuhan Laboratories.

As the Wuhan Institute of Virology enjoyed funding from the NIH, it is also important to closely investigate any electronic or written communication between them relating to any reports of problems of cell contamination, infections or accidents involving laboratory staff. Generally speaking, internal Chinese language documents between the WIV and the WCDC or CCDC or other Chinese authorities will need to be carefully inspected for similar issues. For example:

- Monthly reports
- Project reports
- Status updates
- Project proposals
- Lab notebooks
- Safety audit reports
- Safety incident reports
- Safety procedures.
- Inventory list of pathogens
- Environmental audit reports
- Environmental incident reports
- Facilities improvement projects
- Accident notification protocols
- Emergency procedures and notifications
- Facilities and equipment maintenance logs and records
- Purchasing records by department for supplies, for example, PCR oligonucleotides
- Purchasing records by department for new equipment, such as HEPA filters

Another possible avenue of investigation would be to look into funding trends at Wuhan Laboratories since their certification at BSL1-2-3-4 levels and if there was any significant decrease in funding which may in turn have affected the proper safety functions while conducting research into coronaviruses for vaccine production up to 2019. This could be via records obtained from the Chinese Academy of Science ARP integrated financial system and the government accounting system (WHIOV, 2019c).

A complete investigation would need to ascertain if anyone brought cell cultures from the USA to China or vice versa with the purpose of producing a vaccine for SARS. It would also need to determine if something went wrong in the cell lines during advanced vaccine production. A thorough investigation would also need to determine if scientists at Wuhan Laboratories become aware of such irregularities and neglected to report them due to fear of punishment by Chinese authorities or fear of losing funding for their laboratories.

9. UTILITY RECORDS (WATER AND ELECTRICITY)

It will also be important to obtain a detailed record of water and electricity services at the Wuhan Laboratories since that would reveal useful information about the proper use of the BSL-1-2-3-4 laboratories when staffs were at work or off work. Laboratory equipment such as autoclaves, incubators, PCR machines, etc., usually will be in use even when the staff is not at work. According to the proposed use, some pathogens required overnight incubation periods, and laboratory freezers and refrigeration need to be set at different temperatures (i.e., reagents or vaccines). As is the case with viral vaccines, they need to be appropriately stored in refrigerators to maintain product quality (Driggers, 2019; Parker, 2011). For more information about quality control, see (FDA ORA, 2019; WHO, 2014; Pharmaceutical guidelines, n.d; US Department of Health and Human Services, 2009; American Society for Microbiology, 2009; CDC/NIH, 2020)

10. PHONE RECORDS AND EMAILS

Additionally, the investigation should check all phone records and emails to determine whether normal functions were ever interrupted for safety events inside the laboratories dealing with research into coronaviruses. Indeed, according to a report posted by NBC News Investigations (2020) which analysed Wuhan cell phone data, it has been suggested that an October shutdown at the Wuhan Institute of Virology BSL3 or 4 Laboratories may have occurred, possibly due to a laboratory incident (Dilanian, Arrow, Kuben, Lee, Jones & Bodo, 2020).

The report published by the NBC News Verification Unit and based on commercially-available location data, claimed that there was no mobile phone activity in a high-security portion of the Wuhan Institute of Virology between October the 7th and October the 24th 2019. The report concludes that a 'hazardous event' may have taken place there between October 6th and October 11th 2019 (Shen, 2020).

It is indeed fortunate that in the USA, FOIA requests can be made and as mentioned earlier in this report, email communications from Peter Daszak have been released showing that he acted with a certain lack of integrity when inviting other scientists to sign a public letter to the Lancet condemning so-called conspiracy theories about laboratory leaks (Malkan, 2020; Suryanarayanan, 2020b).

Furthermore, some “Letters of Preservation” already have been issued to scientists involved in collaboration with WIV and more will be issued in the coming weeks:

(See **Appendix 8 - PROPOSED RECIPIENTS of LETTERS of PRESERVATION**).

It is also recommended that researchers working for EcoHealth Alliance, such as Peter Daszak, Jonathan Epstein, Kevin Olival, William Karesh, Tracy Goldstein, Alice Latinne, be invited to answer questions about their collaboration with WIV and asked to hand over their digital devices for inspection and analysis by forensic experts.

Unfortunately, such transparency is inconceivable under the current regime in China, so access to servers and the personal computers and mobile phones of Chinese researchers will be a necessary task, whether remotely or physically (Ferran & Fujita, 2013; Hsu, 2014; Reuters, 2014) to facilitate a complete and effective investigation (Gartland, 2020; Greenwald, 2014)

Specifically, as many of the Chinese researchers mentioned in this report travel abroad to international conferences, it is recommended that their digital devices be examined on entry to the USA or other countries and that they be questioned about their research activities prior to the outbreak in Wuhan. Some names of interest can be found in **Appendix 8 (Letters of Preservation)**, which include on the Chinese side:

- Changchun Tu (Nanjing Military Command Laboratory)
- Zhengli Shi (WIV)
- Peng Zhou (WIV)
- George Gao Fu (CCDC)
- Professor Zhang Yongzhen (Fudan - Shanghai)
- Tian Jun-Hua (WCDC)
- Ge Xing Yi

11. AUTOCLAVES AND DISINFECTANT RECORDS

Other important requirements for any investigation would be to collect photographs and written protocols of how employees carried out molecular biology procedures under BSL (levels 1-4), and to evaluate the use of autoclaves, including purchasing and maintenance records and the records of electrical services during work hours at the Wuhan Laboratory.

A further avenue of investigation would be to investigate the supply chain of laboratory disinfectants such as Microchem Plus to evaluate if they were diluted by suppliers, as occurred in Romania with Hexi Pharma SRL which closed after the Bucharest Colectiv fire because of diluted disinfectants during 2015-2016 (Andreea, 2020). It was found that Hexi Pharma had diluted the active ingredient in least ten specific products by up to 90% in some cases compared to the details printed on the labels (Saddar, 2016).

The Wuhan Institute of Virology recommended the use of Micro-Chem Plus at 5% dilution, which is in line with international standards, as observed in a 2018 paper co-authored by Zhengli Shi "Evaluation of MICRO-CHEM PLUS as a Disinfectant for Biosafety Level 4 Laboratory in China" (Zhang et al., 2018). In fact, the researchers tested Micro-Chem Plus as a method for both infectious liquid waste treatment before autoclaving and shower cycle decontamination of their BSL4 positive pressure suits, using Bat Sars-Like Coronavirus WIV-1 absorbed into filter paper as a test pathogen (Zhang et al, 2018).

The reason the researchers tested Micro-Chem Plus was possibly due to the one year delay in the inauguration of the WIV BSL4 Laboratory, after a French Investigation (Izambard, 2020) discovered safety issues involving disinfectants before its opening:

"Inauguration of WIV P4 Lab was delayed from 2016 to 2017 due to use of bleach in decontamination showers by WIV staff, making it necessary to repair the stainless steel materials affected by corrosion" (Izambard, 2020).

As mentioned above, from 2018 to 2020, WIV was using Micro-Chem Plus in its BSL3 and BSL4 Labs based on recommendations by Professor Shi in her 2018 paper (Zhang et al., 2018), which is generally in line with best practices at US BSL3 and BSL4 Labs (Klaponski et al, 2011; Parkes et al, 2103) where it is also used for chemical shower decontamination and in “dunk tanks”, for example at the Fort Detrick ABSL-4 (Lackemeyer et al, 2014) and at the National Emerging Infectious Diseases Laboratories (NEIDL) BSL4 at Boston University (Henderson & Mellouk, 2014). Micro-Chem Plus was not however licensed for use in Europe or the UK at the time (Parkes et al, 2013).

A more detailed discussion of Micro-Chem Plus and problems with disinfectant use in BSL Laboratories is included in another section of this report which will be published in due course. However, suffice to say that the use of Micro-Chem Plus in BSL4 Laboratories was certainly not the panacea that WIV considered it to be and can be plausibly be considered one of the possible causes for a laboratory leak.

In fact, WIV appears to have become aware of the problems with using Micro-Chem Plus in 2020 when they filed for a patent, “CN112262846A”, on the 13th of November 2020, with the title “Object surface disinfectant for high-grade biosafety laboratory and preparation method thereof”. This patent proposes a new disinfectant formula designed to specifically address a series of observed problems at their BSL3 and BSL4 Laboratories, including “solving corrosion issues which led to leaks of pathogens” (Google Patents, 2020a).

Object surface disinfectant for high-grade biosafety laboratory and preparation method thereof

Abstract

The invention discloses a material surface disinfectant for a high-grade biosafety laboratory and a preparation method thereof. The physical surface disinfectant not only has a killing effect on highly pathogenic microorganisms such as Ebola virus, SARS coronavirus, novel coronavirus, AIDS virus and the like, but also obviously reduces the corrosion effect of the disinfectant on metal, particularly stainless steel by adding nano magnesium into the disinfectant, thereby avoiding biological safety accidents caused by the leakage of the highly pathogenic microorganisms after metal components of biological safety protection facility equipment in a high-grade biological safety laboratory are corroded, simultaneously reducing the aggregation of the nano magnesium by magnetizing the nano magnesium in the disinfectant to enable the nano magnesium to have magnetism, finally forming a stable dispersion system in the disinfectant, obviously increasing the stability of the disinfectant during storage, and keeping the metal corrosion prevention effect of the disinfectant.

CN112262846A

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Inventor: 吴佳, 袁志明, 唐浩, 刘军, 秦颖, 刘毅, 王林

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Figure 6a: Patent CN112262846A “Object surface disinfectant for high-grade biosafety laboratory and preparation method thereof” (Google Patents, 2020a).

The patent was filed by the following scientists at WIV:

Wu Jia, Yuan Zhiming, Tang Hao, Liu Jun, Qin Hao, Liu Yi and Wang Lin

Investigators should ensure that they are questioned closely on the research which led up to the filing of this patent with particular attention being paid to reports on corrosion and failure of HEPA filters at WIV laboratories, relevant photographic evidence, microscope analysis, as well as other safety incident reports..

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Application CN202011271611.1A events ⓘ

2020-11-13 • Application filed by [中国科学院武汉病毒研究所](#), [朗力生物医药\(武汉\)有限公司](#)

2020-11-13 • Priority to [CN202011271611.1A](#)

Figure 6b: Patent CN112262846A “Object surface disinfectant for high-grade biosafety laboratory and preparation method thereof” (Google Patents, 2020a).

This patent is worth examining in some detail as it reveals some quite startling facts:

1. Abstract (Google Patents, 2020a)

*“The invention discloses a material surface disinfectant for a high-grade biosafety laboratory and a preparation method thereof. The physical surface disinfectant not only has a killing effect on highly pathogenic microorganisms such as Ebola virus, SARS coronavirus, novel coronavirus, AIDS virus and the like, but also obviously reduces the corrosion effect of the disinfectant on metal, particularly stainless steel by adding nano magnesium into the disinfectant, **thereby avoiding biological safety accidents caused by the leakage of the highly pathogenic microorganisms after metal components of biological safety protection facility equipment in a high-grade biological safety laboratory are corroded.**”*

2. Detail (Google Patents, 2020a)

“in the existing disinfectants, the disinfectants with low corrosivity to metals such as stainless steel and the like have poor disinfection effect...and the disinfectants with killing effects on the highly pathogenic microorganisms have different degrees of corrosion effects on the metals such as the stainless steel and the like, and **can cause the corrosion of metal components such as the stainless steel and the like after long-term use, so that the biosafety protection effect of the facility equipment is reduced, the service life of the facility equipment is shortened, economic loss is caused, and even the highly pathogenic microorganisms are caused to escape into the external environment of the laboratory, and further the loss of lives and properties of people is caused, and serious social problems are brought.** Therefore, it is necessary to develop a disinfectant which can effectively kill highly pathogenic microorganisms and has no corrosivity or low corrosivity on metals such as stainless steel, and the like.”

3. Technical problem to be solved (Google Patents, 2020a)

“Aiming at the defects of the prior art, the invention provides a material surface disinfectant for a high-grade biosafety laboratory and a preparation method thereof. The disinfectant not only solves the high-efficiency killing effect on highly pathogenic microorganisms, but also solves the problems that the existing disinfectant has strong corrosivity on metal, particularly stainless steel materials, reduces the integrity of high-level biosafety laboratory facilities and equipment, and causes biosafety accidents caused by the leakage of the highly pathogenic microorganisms.”

4. Advantageous effects (Google Patents, 2020a)

- “The corrosivity of the disinfectant on metal, particularly stainless steel materials, can be obviously reduced”
- “The integrity of high-grade biosafety laboratory facility equipment is protected”
- **“The leakage of highly pathogenic microorganisms is avoided”**
- **“Further biosafety accidents are avoided”**

The WIV environmental assessment report (see Appendix 2 - Wuhan Institute of Virology Environmental Assessment Report) shows that Micro Chem Plus was adopted as the chemical bath agent for BSL-4 entry and exit in 2018.

2018 年 5 月 28 日-29 日，中国科学院武汉国家生物安全实验室项目实验室以及环保设施正常运行。

动物房内饲养动物数量：兔子 30 只。

| 表 1 项目生产负荷统计一览表 | | | Cell culture medium, fetal bovine blood cleansing, medical alcohol, 84 disinfectant, MicroChemPlus disinfectant, hydrogen peroxide, formaldehyde |
|-----------------|--------------|--|--|
| 实验室类别 | 实验项目 | 主要使用试剂 | |
| BSL-4 | 高致病病毒感染实验 | 细胞培养基、胎牛血清、医用酒精、84 消毒液、MicroChemPlus 消毒液、过氧化氢、甲醛 | |
| BSL-3 | 高致病细菌与病毒感染实验 | 细胞培养基、胎牛血清、医用酒精、84 消毒液、过氧化氢、甲醛 | |

Figure 7: Wuhan Institute of Virology Environmental Assessment Report (Appendix 2)

Another question that would need to be clarified during any forensic investigation would be regarding the frequency of HEPA filter replacement at Wuhan laboratories. From the data regarding tenders, WIV had one hazardous waste class, “used HEPA filters”, so they seem to have been replaced once a year, as the filters are relatively costly. Although WIV replaced HVAC, some documents refer to “maintenance” of HEPA filters rather than replacement which would a safer method.

Furthermore, investigators should be given full access to any obsolete fixed assets disposed of by WIV in 2020, especially for testing of corroded HEPA filters and other laboratory equipment. On January 8th, 2020, the WIV announced a tender for the “**Recycling and Disposal of the First Batch of Obsolete Fixed Assets in 2020 by Wuhan Institute of Virology, Chinese Academy of Sciences**” (WHIOV, 2020):

“The Wuhan Institute of Virology now needs to recover and dispose of a batch of scientific research and office equipment. Recycling companies with relevant qualifications are invited to participate in the quotation. The first package: a batch of scientific research and office equipment in Xiaohongshan Park. The second package: a batch of scientific research and office equipment in Zhengdian Park” (WHIOV (2020).

In light of the above, investigators should interview “Teacher Zhang” (Xiaohongshan Park. Tel: 027-87199190) and “Teacher Jin” (Zhengdian Park. Tel: 027-51861006) who were in charge of WIV asset disposal and obtain a complete list of items disposed of to cross check with the records of the waste disposal company awarded the contract, probably Hanshi Environmental Engineering Co., Ltd (Xinhua, 2020), which was the only company permitted to dispose of medical waste in Wuhan (Cao, 2020; Li, 2020).

12. LABORATORY CONTAMINATION MONITORING

Prevention of contamination is adversely affected by inherent limitations in the accurate detection of contamination via PCR testing, which is both time consuming and often fails to identify unknown pathogens (Xiao et al., 2019). Thus, Xiao et al. (2019) recommend the use of next generation sequencing (NGS) to effectively and accurately monitor the presence of both known and unknown viruses in laboratories.

If Wuhan laboratories subject to investigation used next generation sequencing routinely to monitor contamination in their laboratories, then electronic records will be available for investigators to query either remotely or in situ.

Regarding next generation sequencing (NGS), WIV must provide documentation on all data handling and storage, including but not limited to interpreted variant call files, .bam, .fastq and .vcf files, in order to facilitate reanalysis of primary data (Australia - Department of Agriculture, 2020).

13. ILLUMINA MACHINE CROSS CONTAMINATION

Contamination of reagents and cross contamination across samples is of course a long-recognized issue in laboratories. Massive sequencing projects and databases based on next-generation sequencing (NGS) suffer even more from contamination issues as contaminant sequence reads are often buried in the sample reads, making them hard to detect and eliminate. Because NGS library construction protocols often use multiple PCR amplification steps, which create high concentrations of DNA, contamination increases (Ballenghien et al., 2017).

The presence of contaminating organisms in high throughput sequencing data can take place via “upstream contamination” of the tissue samples themselves before being processed in the laboratory, or “downstream contamination” caused by environmental contamination during laboratory processing of the same samples (Sangiovanni et al, 2019).

Besides contamination within samples, there may also be cross-contamination among samples. Ballenghien et al (2017) showed that it is essential to analyze NGS datasets for contamination given their findings that “the vast majority of cross-contamination events are ascribable to sequencing centres”. This kind of laboratory specific contamination has been clearly demonstrated in sequencing centres, revealing “specific signatures of contaminating genomes as ‘time stamps’” (Tae et al, 2014).

Analysis of the “unmapped reads” can often detect novel viral sequences which are “hidden by common contaminants of NGS experiments” (Laurence et al, 2014). A case in point would be the Bangladeshi Nipah Strain contamination found in some early Wuhan covid-19 patient samples (Chakraborty, 2020) obtained from Wuhan Jinyintan Hospital, by WIV, as discussed earlier in this report.

Ballenghien et al (2017) describe contamination occurring from unexpected viral infection of cell lines, as was the case with xenotropic murine leukaemia virus-related virus

(XMRV) in human prostate cell lines. The virus infected other cell lines in culture but without “overt phenotypes”, thus giving rise to undetected contamination of cell lines.

Contamination may also occur unexpectedly during the sequencing process, due to the increased sensitivity of cutting edge sequencing technology which allows even small amounts of contaminating nucleic acids to show up in processed datasets (Ballenghien et al, 2017).

Regarding contamination at sequencing centres, Robinson et al (2017) demonstrated that bacterial species detected from RNA and DNA sequencing were associated with specific sequencing centers.

After HeLa-derived human papillomavirus 18 (H-HPV18) was discovered in non-cervical cancer samples in TCGA RNA-seq, Selitsky et al (2020) decided to use virus detection software “VirDEtect” to evaluate this H-HPV18 contamination by looking at laboratory processing variables including time of sequence generation.

Ballenghien (2017) investigated the prevalence of cross-contamination among samples from different species of animals processed in the same laboratory involving subcontracted transcriptome sequencing. They found that 80% of samples from a large-scale sequencing experiment had evidence of cross-contamination from the sequencing centre where libraries were constructed involving PCR amplification:

“we identified a minimum of 782 events of between-species contamination, with approximately 80% of our samples being affected. An analysis of laboratory metadata revealed a strong effect of the sequencing center: nearly all the detected events of between-species contamination involved species that were sent the same day to the same company” (Ballenghien, 2017)

Contamination may take place either during several different processes, including “the library preparation stage through physical transfer of material” and the sequencing stage

“through mis-tagging, when the identifier assigning a read to its source sample is in error” (Ballenghien, 2017).

In a recent study by Abouelkhair (2020), next-generation sequencing data from SARS-CoV-2 infected patients was downloaded from the NCBI SRA datasets and subjected to fastv and Kraken 2 analysis:

“sequence data analysis was performed on public Illumina HiSeq/MiSeq libraries from the NCBI SRA database (Bioproject PRJNA605983) sequenced from bronchoalveolar lavage fluid from five patients (WIV02, WIV04, WIV05, WIV06, and WIV07) with pneumonia at the early COVID-19 outbreak in Wuhan, China”

Abouelkhair (2020) detected multiple non-SARS-CoV-2 genome sequences in the early Wuhan COVID-19 patient BALF samples:

- Nipah virus
- Influenza type A (H7N9) virus
- Human immunodeficiency virus
- Rhabdovirus
- Human metapneumovirus
- Human adenovirus
- Human herpesvirus 1
- Coronavirus NL63
- Parvovirus
- Simian virus 40
- Hepatitis virus

Although some of these non-SARS-CoV-2 sequences are indeed evidence of co-infections in the patients, in several cases they were interpreted erroneously as such.

For example, the Nipah sequences detected in the patient’s BALF, only appear in the metagenomes from a distinct cluster of runs (122-125) on the Illumina HiSeq 3000, not from the Illumina MiSeq, which tends to imply lab contamination, as can be seen in Figure 8 below.

The samples were listed in two separate groups, with the alleged co-infections only appearing from one group, again suggesting laboratory contamination involving fragments of cloned virus which remained attached to the clones,

Group 1. WIV02, WIV04, WIV06, WIV07

Group 2. WIV02-2, WIV04-2, WIV05, WIV06-2, WIV07-2

| SRA | Instrument | Run | Name | | | | | | | | |
|------------|---------------------|-------------|---------|------------|----|------|-----|----|-----|-------|--|
| SRX7730885 | Illumina MiSeq | SRR11092058 | WIV02 | ICU4G | S1 | L001 | | R1 | 001 | fastq | |
| SRX7730885 | Illumina MiSeq | SRR11092058 | WIV02 | ICU4G | S1 | L001 | | R2 | 001 | fastq | |
| SRX7730886 | Illumina MiSeq | SRR11092057 | WIV04 | ICU6G | S2 | L001 | | R1 | 001 | fastq | |
| SRX7730886 | Illumina MiSeq | SRR11092057 | WIV04 | ICU6G | S2 | L001 | | R2 | 001 | fastq | |
| SRX7730887 | Illumina MiSeq | SRR11092056 | WIV06 | ICU9G | S3 | L001 | | R1 | 001 | fastq | |
| SRX7730887 | Illumina MiSeq | SRR11092056 | WIV06 | ICU9G | S3 | L001 | | R2 | 001 | fastq | |
| SRX7730879 | Illumina MiSeq | SRR11092064 | WIV07 | ICU10G | S4 | L001 | | R1 | 001 | fastq | |
| SRX7730879 | Illumina MiSeq | SRR11092064 | WIV07 | ICU10G | S4 | L001 | | R2 | 001 | fastq | |
| SRX7730880 | Illumina HiSeq 3000 | SRR11092063 | WIV02-2 | v300043428 | | L04 | 121 | | | 1 fq | |
| SRX7730880 | Illumina HiSeq 3000 | SRR11092063 | WIV02-2 | v300043428 | | L04 | 121 | | | 2 fq | |
| SRX7730881 | Illumina HiSeq 1000 | SRR11092062 | WIV04-2 | v300043428 | | L04 | 122 | | | 1 fq | |
| SRX7730881 | Illumina HiSeq 1000 | SRR11092062 | WIV04-2 | v300043428 | | L04 | 122 | | | 2 fq | |
| SRX7730882 | Illumina HiSeq 3000 | SRR11092061 | WIV05 | v300043428 | | L04 | 123 | | | 1 fq | |
| SRX7730882 | Illumina HiSeq 3000 | SRR11092061 | WIV05 | v300043428 | | L04 | 123 | | | 2 fq | |
| SRX7730883 | Illumina HiSeq 3000 | SRR11092060 | WIV06-2 | v300043428 | | L04 | 124 | | | 1 fq | |
| SRX7730883 | Illumina HiSeq 3000 | SRR11092060 | WIV06-2 | v300043428 | | L04 | 124 | | | 2 fq | |
| SRX7730884 | Illumina HiSeq 3000 | SRR11092059 | WIV07-2 | v300043428 | | L04 | 125 | | | 1 fq | |
| SRX7730884 | Illumina HiSeq 3000 | SRR11092059 | WIV07-2 | v300043428 | | L04 | 125 | | | 2 fq | |
| SRX7730880 | Illumina HiSeq 3000 | SRR11092063 | WIV02-2 | v300043428 | | L02 | 126 | | | 1 fq | |
| SRX7730880 | Illumina HiSeq 3000 | SRR11092063 | WIV02-2 | v300043428 | | L02 | 126 | | | 2 fq | |
| SRX7730881 | Illumina HiSeq 1000 | SRR11092062 | WIV04-2 | v300043428 | | L02 | 127 | | | 1 fq | |
| SRX7730881 | Illumina HiSeq 1000 | SRR11092062 | WIV04-2 | v300043428 | | L02 | 127 | | | 2 fq | |

Figure 8: Illumina MiSeq and HiSeq Runs for Patient Sample Numbers.

Source: Francisco A. de Ribera (n.d.).

The same principle discussed above may also hold true for the “Adenovirus vaccine genetic sequences” found by Quay (2021) in early patient samples from five Wuhan covid-19 patients collected in December 2019 and sequenced by the Wuhan Institute of Virology using the Novogene Illumina Sequencing machine (Quay, 2021), Figure 9.

| Adenovirus sequences detected | GenBank URL | GenBank Biosample URL | GISAI ID | CoV-2 Isolate | Sequencing Institution | Clinical Information from GISAI |
|-------------------------------|----------------------------|------------------------------|----------------|---|--|---|
| >100 | SRX7730879 | SAMN14082200 | EPI_ISL_402130 | WIV07; Lineage B; mutations NSP3 D1761A, NSP4 T327I; passage original | Wuhan Institute of Virology, Chinese Academy of Sciences | 56 y, male, hospitalized, ICU10G, 20 Dec 2019 |
| >100 | SRX7730880 | SAMN14082196 | EPI_ISL_402127 | WIV02; Lineage B; mutations NSP16 D220N; passage original | Wuhan Institute of Virology, Chinese Academy of Sciences | 32 y, male, hospitalized, ICU4G, outbreak 19 Dec 2019 |
| >100 | SRX7730881 | SAMN14082197 | EPI_ISL_402124 | WIV04; Lineage B; no mutations; passage original | Wuhan Institute of Virology, Chinese Academy of Sciences | 49 y, female, hospitalized, ICU-6, outbreak 27 Dec 2019, Retailer at Huanan Seafood Wholesale Market, patient alive |
| >100 | SRX7730882 | SAMN14082198 | EPI_ISL_402128 | WIV05; Lineage B; NSP3 G1433S, NSP16 K160R; passage original | Wuhan Institute of Virology, Chinese Academy of Sciences | 52 y, female, hospitalized, ICU8G, outbreak 22 Dec 2019; recovered |
| >100 | SRX7730883 | SAMN14082199 | EPI_ISL_402129 | WIV06; Lineage B; no mutations; original passage | Wuhan Institute of Virology, Chinese Academy of Sciences | 40 y, male, hospitalized, ICU9G, 25 Dec 2019 |
| >100 | SRX7730884 | SAMN14082200 | EPI_ISL_402130 | WIV07; Lineage B; mutations NSP3 D1761A, NSP4 T327I; passage original | Wuhan Institute of Virology, Chinese Academy of Sciences | 56 y, male, hospitalized, ICU10G, 20 Dec 2019 |
| 7 small | SRX7730885 | SAMN14082196 | EPI_ISL_402127 | WIV02; Lineage B; mutations NSP16 D220N | Wuhan Institute of Virology, Chinese Academy of Sciences | 32 y, male, hospitalized, ICU, outbreak 19 Dec 2019 |
| 1 small one | SRX7730886 | SAMN14082197 | EPI_ISL_402124 | WIV04; Lineage B; no mutations; passage original | Wuhan Institute of Virology, Chinese Academy of Sciences | 49 y, female, hospitalized, ICU-6, outbreak 27 Dec 2019, Retailer at Huanan Seafood Wholesale Market, patient alive |
| Very few | SRX7730887 | SAMN14082199 | EPI_ISL_402129 | WIV06; Lineage B; no mutations; original passage | Wuhan Institute of Virology, Chinese Academy of Sciences | 40 y, male, hospitalized, ICU9G, 25 Dec 2019 |
| None | SRX8032202 | SAMN14479127 | EPI_ISL_412898 | hCoV-19/Wuhan/HB-CDC-HB-02/2019 | Hubei Provincial Center for Disease Control and Prevention | male, "traveled from Wuhan" |
| None | SRX8032203 | SAMN14479128 | EPI_ISL_402132 | Wuhan HB-CDC-HB-01/2019; Lineage B; mutation Spike F32I; original passage | Hubei Provincial Center for Disease Control and Prevention | 49 y, female, hospitalized |

Figure 9: Patient samples (Adenovirus sequences) Source: Quay (2021 Page 144).

Fuyutao (2020) discussed significant errors in early sampling data in Wuhan in December and January, 2020, and provides tables (Figure 10) showing which machines and institutions sequenced these early patient samples:

| Errors | Platform | Assembler | AgeGender | Virus Name 病毒株名 |
|--------|-----------------------------|---|-----------|----------------------------------|
| 0 | DNBSEQ | SPAdes v3.12.0 | 21F | BetaCoV/Wuhan/WH-03/2019 |
| 4 | Illumina MiSeq, MGISEQ 2000 | Geneious v11.0.3, MEGAHIT v1.2.9 | 32M | WIV02 |
| | | | 32M | BetaCoV/Wuhan/IVDC-HB-05/2019 |
| | | | 32M | BetaCoV/Wuhan/WH19005/2019 |
| 0 | Illumina MiSeq, MGISEQ 2000 | Geneious v11.0.3, MEGAHIT v1.2.9 | 40M | WIV06 |
| | Illumina NextSeq | | 41M1 | BetaCoV/Wuhan/IPBCAMS-WH-03/2019 |
| 0 | Illumina MiniSeq | Megahit v1.1.3 | 41M2 | Wuhan-Hu-1 |
| | DNBSEQ | SPAdes v3.12.0 | 43M | BetaCoV/Wuhan/WH-02/2019 |
| | DNBSEQ | SPAdes v3.12.0 | 44M | BetaCoV/Wuhan/WH-01/2019 |
| 0 | Illumina Miseq | CLC Genomics Workbench 12 and Geneious 12.0.1 | 49F | BetaCoV/Wuhan/HBCDC-HB-03/2019 |
| 0 | | | 49F | WIV04 |
| 6 | Illumina NextSeq | | 49F | BetaCoV/Wuhan/IPBCAMS-WH-02/2019 |
| 1 | Illumina Miseq | CLC Genomics Workbench 12 and Geneious 12.0.1 | 49F | BetaCoV/Wuhan/HBCDC-HB-01/2019 |
| 0 | | | 49F | BetaCoV/Wuhan/IVDC-HB-01/2019 |
| | | | 49F | BetaCoV/Wuhan/WH19001/2019 |
| 2 | MGISEQ 2000 | Geneious v11.0.3, MEGAHIT v1.2.9 | 52F | WIV05 |
| 0 | Illumina NextSeq | | 52F | BetaCoV/Wuhan/IPBCAMS-WH-04/2019 |
| | | | 56M | WIV07 |
| 4 | Illumina NextSeq | | 61M | BetaCoV/Wuhan/IPBCAMS-WH-05/2020 |
| | | | 61M | BetaCoV/Wuhan/IVDC-HB-04/2020 |
| | | | 61M | BetaCoV/Wuhan/WH19004/2020 |
| | Illumina NextSeq | | 65M | BetaCoV/Wuhan/IPBCAMS-WH-01/2019 |
| 0 | Illumina Miseq | CLC Genomics Workbench 12 and Geneious 12.0.1 | UM1 | BetaCoV/Wuhan/HBCDC-HB-02/2019 |
| | Illumina Miseq | CLC Genomics Workbench 12 and Geneious 12.0.1 | UM2 | BetaCoV/Wuhan/HBCDC-HB-04/2019 |
| 0 | | | UU1 | BetaCoV/Wuhan/WH19008/2019 |

Figure 4. Counting sequencing and/or assembly errors.

Figure 10: <https://virological.org/uploads/short-url/kUOjxvNkuOgQY5YazZFeRLTjwrQ.pdf>

Source: Fuyutao (2020)

From the information contained in the above Figures, Francisco A. de Ribera (n.d.) reordered the table of Quay(2020) in Figure 11, and it can be observed that the adenovirus is associated only with the WIV MiSeq (except for WIV07 (SRX7730879, SRR11092064))

| | HiSeq | MiSeq | HBCDC |
|---------------------|-------|----------|-------|
| WIV07 | >100 | >100 | |
| WIV02 | >100 | 7 small | |
| WIV04 = HBCDC-HB-01 | >100 | 1 smal | None |
| WIV05 | >100 | | |
| WIV06 | >100 | Very few | |
| HBCDC-HB-02 | | | None |

Figure: 11. Adenovirus Samples matched by Sequencer (Francisco A. de Ribera, n.d.)

This in turn suggests that the adenovirus itself was probably not in the patients, but rather the result of laboratory contamination, as it was only found in the datasets from one of the two laboratories.

The laboratories in question used an Illumina sequencing machine belonging to Novogene to process the Wuhan Hospital patient samples. Illumina sequencing machines are well known to give rise to cross-contamination:

“The Illumina MiSeq DNA sequencing system generates several gigabases of short reads per run with a relatively low error rate. It is characterized by systematic low level, within-run cross-sample contamination, an under-reported issue for this platform” (Brumme et al, 2020)

Laurence et al (2014) point out that Illumina machines often reveal unknown and previously undetected sequences which may be mistakenly identified as emerging from specimens rather than laboratory contamination:

“Unbiased high-throughput sequencing of whole metagenome shotgun DNA libraries is not limited to known sequences. Unlike most sequencing applications, it is highly sensitive to laboratory contaminants as these will appear to originate from the clinical specimens” (Laurence et al, 2014)

Despite possible yet time consuming strategies to remedy these issues involving qPCR analysis, sequencing run contamination remains an integral feature of Illumina machines due to their high sensitivity (Laurence et al, 2014).

This raises an interesting and novel avenue of inquiry for remotely investigating Wuhan laboratory viral sequences without having access to their laboratories. DRASTIC researchers found sequence traces of bat coronaviruses in the datasets uploaded to genetic databases by researchers from other Universities and organisations in Wuhan who used the same Illumina NGS sequencing machine (ST-J00123) owned by Novogene.

Novogene has offices in Beijing and Wuhan, and its Illumina NGS machines were shared by Universities and Laboratories in Hubei, such as BIG, CTGU, HBMU, HBUST, HUST, HZAU and WIV.

Indeed, it seems that WIV has left traces of their viruses in metagenomes of other institutions of Hubei (Zhang, 2021; Quay, 2021). Cases in point are the datasets “CRA001143” and “CRA001432” mentioned by Cai et al (2019) from the Hubei University of Science and Technology (HBUST) in their 2019 paper.

Unfortunately, the Genome Sequence Archive (GSA) for “CRA001143” was initially embargoed on BIGD until September 2022. An independent researcher from “DRASTIC”, Francisco A. de Ribera (n.d.) considers it possible that it contained runs from ST-J00123 (the Illumina sequencing machine used by WIV for RaTG13). After pressure from the journal publishers and requests from independent researchers, the “CRA001143” dataset has now been made available and is being analysed for evidence of contamination and any traces of bat coronavirus sequences. See below Figures 12 and 13.

To identify time-dependent DE miRNAs, we used the maSigPro package (Conesa et al., 2006) with default parameters. Gene ontology (GO) enrichment was also assessed using the enrichGO functions in R package clusterProfiler (version 3.6.0) (Yu et al., 2012) and the significance of the enriched GO terms was evaluated using a hypergeometric test with a false discovery rate <0.05 . Our sequencing data have been uploaded to the Genome Sequence Archive with accession numbers **CRA001143** and CRA001432.

Figure 12: Datasets “CRA001143” and “CRA001432” (Cai et al, 2019)

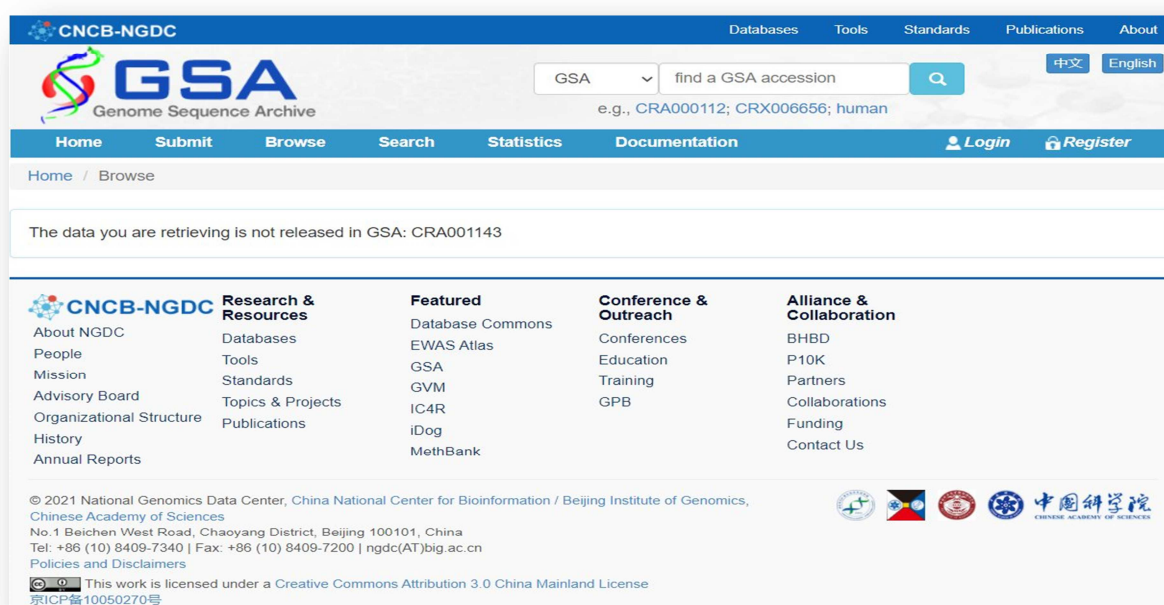


Figure 13: CRA001143 <https://bigd.big.ac.cn/gsa/browse/CRA001143> (BIGD-GSA, 2021)

A further example was revealed in another paper from researchers at the Huazhong University of Science and Technology (HUST) (Wu et al., 2020) which used the CRA001143 dataset:

"All data presented are freely available without restriction in the public open access database (GSA, CRA001143)" and "the high-throughput sequencing data reported in this study have been deposited in Genome Sequence Archive (GSA), CRA001143" (Wu et al., 2020). Their paper states that:

"The libraries were sequenced on a HiSeq X Ten sequencer with paired-end reads, 150-cycle sequencing run by Novogene" (Wu et al. 2020). Novogene is the owner of the Illumina machine shared by WIV, HZAU, HBUST, CGTU and HUST in Wuhan.

CRA001143 seems to have been created (but not released) on a similar date as the RaTG13-labelled amplicons (27-Sep-18 to 30-Sep-18). Another CRA with a close ID, CRA001148, is within a project whose "submission date" is 2018-10-07.

The CRA datasets could contain runs from the Novogene Illumina ST-J00123 as both institutions (HBUST) and HUST in Wuhan are known for sharing the Novogene Illumina machine with WIV. Thus, if these datasets can be analyzed, they may well reveal sequence contamination from WIV work with RaTG13 or the 7896 clade, which could for example, allow independent researchers to assemble RaTG13 from the sequence in the datasets.

Bearing this in mind, Francisco A. de Ribera (n.d), a researcher from DRASTIC has generated a partially complete list of sequencing runs on the Novogene ST-J00123 Illumina HiSeq 300 machine used by different laboratories in Hubel Province:.

| Instrument | Run | FCID | Lar | Abbrevi | Reference | BioProject | SRP | SRX | SRR | Key |
|------------|-----|-----------|-----|---------|---------------------|-------------|-----------|------------|-------------|---------------------------------------|
| ST-J00123 | 27 | H7WFNBBXX | 5 | HZAU | | | | | | |
| ST-J00123 | 28 | | 3 | HZAU | | PRJNA338015 | SRP081118 | | | |
| ST-J00123 | 28 | H7Y52BBXX | 6 | HZAU | Zhao et al. (2020) | | | | | MH_young_leaf_Input_1 |
| ST-J00123 | 30 | H7Y75BBXX | 6 | HZAU | | | | | | |
| ST-J00123 | 31 | | 1 | HZAU | | PRJNA338015 | SRP081118 | | | |
| ST-J00123 | 38 | | 1 | HZAU | | PRJNA396502 | SRP114409 | | | |
| ST-J00123 | 44 | | 1 | Other | | | | | | Patent CN107190003A |
| ST-J00123 | 44 | | 3 | WIV | Xie et al. (2018) | PRJNA393936 | SRP111649 | | | |
| ST-J00123 | 44 | hcc5vbbxx | 4 | HZAU | Zhao et al. (2020) | PRJNA597475 | SRP238686 | SRX7437747 | SRR10763648 | MH63_young_leaf_DNAmeth_1 |
| ST-J00123 | 44 | hcc5vbbxx | 4 | HZAU | Zhao et al. (2020) | PRJNA597475 | SRP238686 | SRX7437748 | SRR10763649 | MH63_young_leaf_DNAmeth_P1 additional |
| ST-J00123 | 44 | hcc5vbbxx | 4 | HZAU | Zhao et al. (2020) | PRJNA597475 | SRP238686 | SRX7437755 | SRR10763656 | ZS97_young_leaf_DNAmeth_1 |
| ST-J00123 | 65 | HF7TNBBXX | 8 | HZAU | | | | | | |
| ST-J00123 | 65 | | 8 | WIV | Xie et al. (2018) | PRJNA393936 | SRP111649 | | | |
| ST-J00123 | 67 | | 2 | HZAU | | PRJNA383075 | SRP104117 | | | |
| ST-J00123 | 67 | | 3 | HZAU | | PRJNA383075 | SRP104117 | | | |
| ST-J00123 | 67 | | 6 | HZAU | | PRJNA383075 | SRP104117 | | | |
| ST-J00123 | 67 | | 8 | HZAU | | PRJNA383075 | SRP104117 | | | |
| ST-J00123 | 68 | | 6 | HZAU | | PRJNA383075 | SRP104117 | | | |
| ST-J00123 | 68 | | 7 | HZAU | | PRJNA383075 | SRP104117 | | | |
| ST-J00123 | 78 | HKWJNBBXX | 4 | HZAU | Yang et al. (2019b) | PRJNA531553 | SRP192698 | SRX5692740 | SRR8907014 | GEMS28 |
| ST-J00123 | 78 | HKWJNBBXX | 5 | HZAU | Yang et al. (2019b) | PRJNA531553 | SRP192698 | SRX5692470 | SRR8906763 | BY807 |
| ST-J00123 | 78 | HKWJNBBXX | 6 | HZAU | Yang et al. (2019b) | PRJNA531553 | SRP192698 | SRX5692474 | SRR8906759 | ZHENG32 |
| ST-J00123 | 78 | HKWJNBBXX | 7 | HZAU | Yang et al. (2019b) | PRJNA531553 | SRP192698 | SRX5692476 | SRR8906757 | M97 |
| ST-J00123 | 78 | HKWJNBBXX | 8 | HZAU | Yang et al. (2019b) | PRJNA531553 | SRP192698 | SRX5692473 | SRR8906760 | TY4 |
| ST-J00123 | 79 | HL3W5BBXX | 2 | HZAU | Yang et al. (2019b) | PRJNA531553 | SRP192698 | SRX5692475 | SRR8906758 | R15 |
| ST-J00123 | 79 | HL3W5BBXX | 4 | HZAU | Yang et al. (2019b) | PRJNA531553 | SRP192698 | SRX5692469 | SRR8906764 | U8112 |
| ST-J00123 | 84 | hmn7ybbxx | 6 | WIV | Zhou et al. (2020) | PRJNA606165 | SRP249482 | SRX7724752 | SRR11085797 | RaTG13 |
| ST-J00123 | 85 | HMMKLBXX | 2 | HZAU | Yang et al. (2019b) | | | | | seedling leaf,ChIA-PET rep2 |
| ST-J00123 | 85 | HMMKLBXX | 6 | HZAU | Yang et al. (2019b) | PRJNA531751 | SRP191927 | SRX5666326 | SRR8879428 | seedling leaf,ChIA-PET rep1 |
| ST-J00123 | 88 | | 1 | WIV | Li et al. (2020) | PRJNA606159 | SRP249478 | | | |
| ST-J00123 | 88 | | 3 | WIV | Li et al. (2020) | PRJNA606159 | SRP249478 | | | |
| ST-J00123 | 89 | | 5 | HZAU | Li et al. (2019b) | PRJNA380842 | SRP102637 | | | |
| ST-J00123 | 89 | | 5 | WIV | Li et al. (2020) | PRJNA606159 | SRP249478 | | | |
| ST-J00123 | 89 | | 6 | WIV | Li et al. (2020) | PRJNA606159 | SRP249478 | | | |
| ST-J00123 | 89 | | 7 | WIV | Li et al. (2020) | PRJNA606159 | SRP249478 | | | |
| ST-J00123 | 89 | | 8 | WIV | Li et al. (2020) | PRJNA606159 | SRP249478 | | | |
| ST-J00123 | 91 | | 1 | WIV | | PRJNA649646 | SRP274224 | | | |
| ST-J00123 | 93 | HTY5YBBXX | 6 | HZAU | Zhao et al. (2020) | PRJNA597475 | SRP238686 | SRX7437763 | SRR10763664 | Nip_young_leaf_DNAmeth_1 |
| ST-J00123 | 93 | HTY5YBBXX | 8 | HZAU | Zhao et al. (2020) | PRJNA597475 | SRP238686 | SRX7437764 | SRR10763665 | Nip_panicle_DNAmeth_1 |
| ST-J00123 | 104 | H2KK3BBXY | 7 | CTGU | | | | | | |
| ST-J00123 | 104 | H2KK3BBXY | 7 | CTGU | | | | | | |
| ST-J00123 | 104 | H2KK3BBXY | 7 | CTGU | | | | | | |
| ST-J00123 | 104 | H2KK3BBXY | 7 | CTGU | | | | | | |
| ST-J00123 | 105 | | 6 | HZAU | | PRJNA554517 | SRP214970 | | | |
| ST-J00123 | 119 | H75C5BBXY | 2 | CTGU | | | | | | |
| ST-J00123 | 119 | H75C5BBXY | 2 | CTGU | | | | | | |
| ST-J00123 | 119 | H75C5BBXY | 2 | CTGU | | | | | | |
| ST-J00123 | 119 | H75C5BBXY | 3 | CTGU | | | | | | |
| ST-J00123 | 119 | H75C5BBXY | 3 | CTGU | | | | | | |
| ST-J00123 | 119 | H75C5BBXY | 4 | CTGU | | | | | | |
| ST-J00123 | 119 | H75C5BBXY | 6 | CTGU | | | | | | |
| ST-J00123 | 119 | H75C5BBXY | 7 | CTGU | | | | | | |
| ST-J00123 | 119 | H75C5BBXY | 8 | CTGU | | | | | | |

Figure 14: ST-J00123 Illumina HiSeq 300 Runs Source: Francisco A. de Ribera (n.d.).

By requesting all output files from Illumina HiSeq 300 machine ST-J00123 and analysis of all raw sequence reads (of any institution) from the Illumina HiSeq 3000, traces of sequences of bat viruses that WIV were experimenting on can be detected and analysed.

Thus, despite the time elapsed since the putative laboratory leak, thanks to the shared Illumina sequencing machines in Wuhan, investigators will be able to “illuminate” many details of WIV research into bat viruses between 2012 and 2021.

This investigation of contamination in uploaded and publicly available datasets can best be undertaken using a free software tool called “DecontaMiner” developed by Sangiovanni et al (2019), available at:

<https://bio.tools/DecontaMiner>

<https://github.com/amarinderthind/decontaminer>

<http://www-labgtp.na.icar.cnr.it/decontaminer/>

“DecontaMiner” is described in “From trash to treasure: detecting unexpected contamination in unmapped NGS data (Decontaminer)” (Sangiovanni et al, 2019) as a free software solution to investigate:

“the presence of contaminating sequences in NGS data and analyze the sequences rejected during the alignment to the reference genome, the so called unmapped reads”.

DecontaMiner can use fastq, fasta or bam format files, but specifically for investigative purposes:

“as a detection tool to identify contaminating sequences among the unmapped reads, generally stored in a bam file”.

For example, using Decontaminer, investigators could remotely search published SRA datasets for any traces of 7896 Clade Viruses sequences, specifically for the recently revealed aa sequence:

NSKAKDENGQYFYRLFRKSKLLPFQRDVSNVTYGSKGNDGCNPSEADCYWPLLKY
GFTGSVSQDYQPYP

14. SAMPLE STORAGE MANAGEMENT SYSTEM AT WIV

Evidence from a Beijing based company, “Orient International Tendering Co., Ltd”, (China Government Procurement Network (2019a), shows that the Wuhan Institute of Virology installed a “scalable automated sample storage management system” in July 2019, featuring “High-throughput automated access to precious biological samples such as viruses, blood samples, and genetic resources at ultra-low temperature (-80°C)”. (China Government Procurement Network (2019b). This new system included:

- A 2D bar code management system for all sample tubes.
- Identification, Tracking and Recording of the whole process, including access to samples.
- A single-tube picking method to prevent unauthorised removal of samples.
- A management system automatically tracking records of sample entry and exit.
- Independent account and password for each user

It is thus clear that from the latter half of 2019, all samples at WIV were stored via this automated retrieval system based on two dimensional barcodes, which ensured consistent temperatures and electronic recording of movement of samples (China Government Procurement Network, 2019b). Again, investigators will need access to this data to compare with experimental data recorded in lab books and employee records.

15. LABORATORY DATA

Finally, Laboratory technicians who carry out genetic sequencing and animal imaging will generate significant amounts of data (McCall & Hardcastle, 2014). Thus, further mapping of physical and virtual assets will need to be done either remotely or physically:

- Computer machine addresses
- Internal viral pathogen databases
- Dedicated data centers
- Hardware firewalls
- Local area networks
- NGS generated data (interpreted variant call files, .bam, .fastq and .vcf files)
- PCR data
- Private cloud infrastructure
- Secure wireless networks
- Video monitoring systems
- Virtualized servers
- Virtualized desktops
- VoIP networks
- “WeChat” user groups
- WIV laboratory automation systems (McCall & Hardcastle, 2014).

16. TENDERS, EQUIPMENT PURCHASE AND MAINTENANCE CONTRACTS

Complete details of maintenance projects, tenders and contracts awarded in 2019 by the Wuhan Institute of Virology and other Wuhan BSL Laboratories of interest, will need to be made available to investigators, for example those during 2019 from WIV:

1. Maintenance Project of P3 Laboratory and Laboratory Animal Centre (March 01, 2019) awarded to Hubei Guohua Tendering Consulting Co., Ltd (CCGP, 2019a).
2. Renovation project of the hazardous waste treatment system (July 31, 2019), awarded to Hubei Guohua Tendering Consulting Co., Ltd (CCGP, 2019b).
3. Procurement project of the environmental air disinfection system and the scalable automated sample storage management system (August 14, 2019) (China Government Procurement Network, 2019b).
4. Procurement of air incineration devices and test service projects (December 03, 2019) (CCGP, 2019d)
5. Purchase of positive pressure protective clothing project (March 21, 2019) awarded to Hubei Guohua Tendering Consulting Co., Ltd. Purchased from Hong Kong Yi'an Huawei Co., Ltd (HK) (CCGP, 2019e)
6. Central Air Conditioning Renovation Project of Wuhan Institute of Virology. September 16, 2019 (CCGP, 2019f)
7. Procurement project of high-throughput cell clone screening system and upright multiphoton microscopy imaging system of Wuhan Institute of Virology awarded to Wuhan Ruikang Ogilvy Biotechnology Co., Ltd, Wuhan (CCGP, 2019g)

8. Procurement of Scientific Research Instruments and Equipment for Wuhan Institute of Virology (Probe/Optical Profiler, Piezoelectric micro inkjet dot matrix preparation system, Microcapillary cell analysis platform, Nuclear transfection system, Critical point dryer, High-throughput cell clone screening system, Pulsating vacuum autoclave) April 26, 2019 11:12 via Oriental International Tendering Co., Ltd. (CCGP, 2019h)

Some of the tender companies or service providers may hold laboratory documents which any investigation may need to analyse, for example: floorplans, schematics for plumbing and electricity can all give some idea of the extent of pathogen research and the location of animal facilities. These plans would also help investigators to locate network servers, video surveillance, intrusion alarms, water pipes and electricity supply lines, discharge pipes and even differences in pressure between laboratory areas as well as airflow (Reed & Dunaway, 2019). By comparing these plans from 2019 with current assets at WIV, differences or anomalies may be exposed.

17. WUHAN INSTITUTE OF VIROLOGY – VIRAL PATHOGENS DATABASES

Figure 15 below is from a now deleted website showing the full extent of the viral pathogen databases administered by the Wuhan Institute of Virology, all of which have been taken offline (Anon, Bostickson, & Demaneuf, 2021)..



Figure 15. Deleted and Missing WIV Databases (Anon, Bostickson, & Demaneuf, 2021).

An essential part of our proposed investigation would be to examine carefully any viral pathogen sequence databases (Anon, Bostickson, & Demaneuf, 2021) operated by laboratories in Wuhan (Figure 15). Disturbingly, it has been confirmed that WIV took all Chinese viral pathogen databases offline (i.e. The Static 61.5MB SQL version altered on December 30th, 2019 in Figure 16) and deleted it from Chinese public servers against Chinese database regulations. (Anon, 2020b, Segreto & Deigin, 2020, Devine, 2020).

| Introduction to the basic information of the database (set) [edit] | |
|--|---|
| Database (set) name | Database of pathogens of bat and mouse origin |
| Data author | Tang Yijie, Li Bei, Zhou Zijian, Zhu Yan, Zhao Kai, Ma Lili, Wu Yuewei, Shi Zhengli |
| Data Communication Author | Shi Zhengli (zlshi@wh.iov.cn) |
| The amount of data | 61.5 MB |
| Data Format | MYSQL |
| Data Service System URL | http://batvirus.whioi.ac.cn/ http://www.sciencedb.cn/dataSet/handle/768 |
| Fund project | Special project on information technology of Chinese Academy of Sciences (XXH13505-03-210), national major scientific research instrument development project (31727901). |
| Database (set) composition | This database is composed of four parts: bat collected sample data, bat virus pathogen data, mouse collected sample data and mouse virus pathogen data. The database covers a total of 22,257 samples and viral pathogen data accumulated by the research group for a long time, as well as relevant data published by foreign authorities. |

Figure 16: Description of SQL Pathogen Database (Anon, Bostickson, & Demaneuf, 2021)

There were at least 100 unpublished sequences of bat beta coronaviruses on the Chinese website hosting the SQL database. Professor Zhengli Shi publicly admitted that they were password protected and available by contacting her. (Anon, 2020b, Segreto & Deigin, 2020) These unpublished sequences have yet to be revealed, and it would certainly be essential to investigate and BLAST them to assess their similarity to SARS-CoV-2 (Segreto & Deigin, 2020).

Thus, another important question arises; were the databases deleted to avoid the identification of closely matching viral sequences which may have been involved in the development of genetic manipulation and experiments involving serial passage in animals such as ferrets (Sirotkin & Sirotkin, 2020).

To further address this question, it is essential to investigate fully why one of the WIV online pathogen databases was altered on December 30th and later deleted (Anon, Bostickson, & Demaneuf, 2021; Devine, 2020; Segreto & Deigin, 2020). In May 2020, several media outlets reported the renaming of the online database from “Wildlife borne viral pathogen database” to “Bat and rodent borne viral pathogen database” (Anon, 2020b, Devine, 2020).

The person responsible, most probably Zhengli Shi, as she was the database administrator, deliberately changed the description on December 30 after being informed of the market outbreak in Wuhan, by replacing “wild animal” by “bat and rodent”, and by deleting “arthropod vectors”. These alterations and deletions took place during her return train trip from Shanghai to Wuhan (Anon, 2020b, Devine, 2020). This seems to be a rather strange thing to do after hearing about a possible SARS related pandemic.

Originally, 61.5 Megabytes of SQL data was available for download as a zip file from the CNIC website, but in May 2020, the zip file finally became unavailable (Anon, 2020b, Segreto & Deigin, 2020). Sadly for independent researches, the whole database page was eventually deleted. Despite months of persistent requests to multiple institutions and individuals, including WIV and Peter Daszak, these polite requests have been ignored.

The authors of this report insist that this database be made available to independent researchers, especially the password protected section containing many unpublished viral sequences of interest, including bat betacoronaviruses.

From the database access metadata (CAS CNIC Big Data, 2020), Foreign and Chinese research institutions are known to have downloaded the SQL zip file in 2019 and the authors call on them to provide copies in the public interest.

WIV scientists have stonewalled international attempts to clarify the deleted databases issue. In email communications over 7 months ago and when questioned by the BBC more recently, Zhengli Shi claimed the deletion was due to “cybersecurity issues” and the databases would be back online when they felt “safe”. Another WIV colleague at the same time (July 2020) claimed, “They are being updated” and would be “available shortly” (Anon, Bostickson, & Demaneuf, 2021; Sudworth, BBC 2020).

*“Prof Shi has also faced questions about why **the WIV's online public database of viruses** was suddenly taken offline. She told the BBC that the WIV's website and the staff's work emails and personal emails had been attacked and the database taken offline for security reasons”* (Sudworth, BBC 2020).

Both Peter Daszak and Edward Holmes presumably enjoyed privileged access or could have requested access to these databases due to their close collaboration with either Zhengli Shi, WIV (Hu et al., 2017, Li et al., 2019, Zhou et al, 2018, Yuan et al., 2014), George Gao (Huang et al., 2016) and the WDCDC (Shi et al, 2016). As part of an investigation, they should thus be asked to provide copies of the databases. Furthermore, a forensic database team should examine any changes in databases, especially the password protected section of the WIV viral pathogen database which contained unpublished viral sequences involving patents, upcoming papers, IP questions or classified military research (Anon, Bostickson, & Demaneuf, 2021).

18. CALL FOR A “FULL AND UNRESTRICTED INTERNATIONAL FORENSIC INVESTIGATION”

Recently, 26 scientists (virologists, microbiologists and zoologists) have now come forward to echo our concerns and proposals by signing an open letter which was published on the 4th of March 2021 calling for “a Full and Unrestricted International Forensic Investigation into the Origins of COVID-19” (WSJ, 2021). They consider that this new investigation should not “exclude the possibility of a leak or accident at or connected with a research facility such as the Wuhan Institute of Virology” (McKay et al, 2021).

These scientists call for a thorough independent international inquiry as they believe that the WHO was not given sufficient access “to adequately investigate possible sources of the new coronavirus” and that it was “all but impossible” for the WHO team to conduct a full investigation, concluding that “efforts to date do not constitute a thorough, credible, and transparent investigation” (McKay et al, 2021).

The scientists consider that the WHO investigation was hampered by faulty and limited terms of reference which rely on consensus involving 17 Chinese and 17 international members who were selected without regard to possible conflicts of interest and did not include the appropriate experts needed to carry out laboratory investigations (New York Times, 2021). They call for an independent international team including a wide range of experts, some of whom should have a deep knowledge of Chinese language and culture. This team should consider with due care and attention the possibility of (WSJ, 2021) a variety of factors which are listed below.

- “Infection at a sampling site of a lab employee or of non-lab personnel
- Infection during transport of collected animals and/or samples
- Lab Acquired Infection (LAI) in one of the laboratories in Wuhan

- Lab-escape without LAI, for instance via waste handling or animals that escaped or were disposed of inappropriately”

The 26 scientists also insist that a truly credible investigation will need to include full access to (WSJ, 2021):

- “Hospital records of Chinese coronavirus cases in late 2019
- Records including maintenance, personnel, animal breeding and experiment logs from all laboratories working with coronaviruses
- Early cases and their relatives and past and present personnel associated with the sites or institutions of interest such as markets, hospitals
- A secure reporting channel for people to confidentially contribute information, wherever they are based, without fear of punishment or retribution.
- Hospital records from fall 2019 of early or suspect patients, including interviews with patients or contacts
- Current and past personnel, such as employees of the labs in 2019 and people present at specific sampling sites
- All sites, records, samples, and personnel of interest
- Key Wuhan markets
- All laboratories and institutions known to have worked on coronaviruses or shared facilities or equipment with groups that worked on coronaviruses”
- Important pathogen sampling sites, such as the Mojiang mine
- All relevant records of the labs and institutions involved in coronavirus research
- Environmental reports
- Inspection reports
- Maintenance logs

- Lab experiment logs
- Raw sequence reads.
- Records of shipments of samples
- Specimen destruction records
- Personnel logs
- Incident reports
- Animal breeding records
- Sampling trip records, including the 2013 Mojiang sampling trip
- Key databases of pathogens, samples, and isolates, including those taken offline.
- Granular data, preferably directly from the source and in its raw form, not summarized data (which can be anonymized if necessary)
- Market samples
- Environmental samples
- Hospital samples
- Wastewater samples
- Blood banks samples
- Chinese CDC case records
- Primary hospital and/or clinic records.
- Other Chinese case databases describing pneumonia cases” (WSJ, 2021).

C. LIMITATIONS TO THIS REPORT

19. CHALLENGES

Kortepeter (2020) outlines four ways a pathogen like SARS-COV-2 could “leak” from a laboratory; by aerosol release, accidental exposure in the lab, via an animal or on an inanimate object/wastewater/sewage (fomites) and finally by the deliberate release of a pathogen.

As this report proposes, an international scientific team should carry out a thorough investigation of Wuhan BSL Laboratories, but as Kortepeter (2020) points out, it is extremely difficult to objectively determine if a given outbreak was caused by intentional release of a pathogen. In a review of the topic, Dembek et al, (2006) use a scoring system previously developed to analyse four outbreaks definitely caused by bioterrorism and two natural outbreaks. Revealing the difficulty in making these assessments, they only managed to evaluate the 2001 anthrax letters as “highly likely to be caused by bioterrorism”, noting that “scoring systems for bioterrorism are imperfect” (Kortepeter, 2020).

This finding highlights the serious challenges that investigators have when trying to evaluate if any given outbreak was in fact “deliberate” Dembek et al, (2006). Some useful quotations from the article by Kortepeter (2020) clearly illustrate these challenges

A. Plausible Deniability

“One of the key attractions for bioterrorism is plausible deniability. The bar is high for proof, and an attack can be hidden under the cover of a natural outbreak. The only “smoking gun” for a biowarfare event is to find something like a spray device contaminated with the pathogen or something akin to the anthrax letters of 2001, which were undeniably put in the mail by a human” Kortepeter (2020)

B. Need for a Formal Investigation

“In the case of SARS-CoV-2, the only way to get closer to understanding its mysterious origin is to send an unbiased international team into China to conduct a formal investigation to determine the earliest humans infected in the outbreak, compare specimens in the Wuhan lab with human specimens, and analyze similar viruses in bat and other animal populations” Kortepeter (2020)

C. Finding Proof

“Finding specimens in the lab that existed prior to the current outbreak that genetically match human outbreak samples would be damning. That would only prove that the samples came from a lab, not whether the outbreak was started intentionally. Having someone who worked in the lab confess would certainly help, but I wouldn’t bet the farm on that happening, either” Kortepeter (2020)

Apart from the factors listed above, other factors may hamper any investigation into an outbreak, such as political issues. A clear example of the difficulties in identifying whether there was a laboratory leak of a pathogen would be the Sverdlovsk anthrax outbreak in 1979.

In 1979, in Sverdlovsk in the former Soviet Union, dozens of people died from inhalational anthrax from spores that leaked from a classified research laboratory when a filter was left unreplaced. At the time, the Soviet Union instituted a complex cover up plan which involved USSR scientists travelling to the USA to show that the disease was in fact merely a case of gastrointestinal anthrax caused by contaminated meat (Kortepeter, 2020).

However, when the USSR “dissolved”, US scientists determined “ from autopsy records, visits to graveyards, interviews with victims’ families and pathologists, and analysis of where the infections occurred” that the outbreak was most probably caused by a leak of anthrax spores from the classified bioweapons laboratory in Sverdlovsk. Indeed, this was broadly confirmed by the new President, Boris Yeltsin in due course (Kortepeter, 2020).

20. SPECIFIC LIMITATIONS

This report presents some hypotheses of laboratory negligence regarding the possible emergence of SARS-CoV-2 in Wuhan and its subsequent spread worldwide.

The authors of this report firmly believe that a full investigation needs to be conducted to clarify the origin of the SAR-CoV-2 pandemic. However, the passing of time is certainly an issue which needs to be taken into account, as it adversely affects any criminal investigation in terms of evidence which can be analyzed to prove or disprove the hypotheses presented.

An important question raised by this is FIRSTLY, how exactly investigators will gain full access to health and genomic databases related to this pandemic, in order to assess how and where this pandemic started. Secondly, at this time, entrance to China is restricted to investigators. Finally, some other limitations that can affect any investigation include:

- Language Barriers
- Distance and transport links between the US and China
- The political and economic environment of US- China relations
- The guarantee of security of investigators in China
- Lack of familiarity with Chinese culture and language by investigators
- Access to documents and electronic databases
- Access to blood samples and wastewater samples
- How to deal with strong local CCP influence and control over operations and policies at Chinese State laboratories (Zhang, 2020) in order to augment cooperation between Laboratory Staff and the Investigation Team
- Need for high level permission to obtain cooperation from local CCP units at Wuhan Laboratories, to ensure proactive and transparent communication with Laboratory Staff

- How to agree about the scope or SOP of different kinds of investigate tests
- What kind of testing is better (i.e. screening, diagnostic, pharmacogenomics, prognostic)?
- How to identify unknown diseases present in cell lines (e.g.: SARS-CoV-2)
- How to store, preserve and transfer biological genomic evidence from China to the US
- How to evaluate the safety of such evidence for scientists and investigators
- How to create criteria to recognize tainting in genomic samples
- How to evaluate the lab equipment used in China at Wuhan Laboratories
- How to ensure best practices doing such a forensic investigation, including insurance, variants of unknown significance, sensitivity of data and permanence of data.

21. LESSONS TO BE LEARNED

Time based constraints in terms of the time passed (over a year) since the outbreak began in Wuhan in 2019 may have a significant effect on the ability to collect any evidence in China and may render it inconclusive. Despite these serious challenges to an effective investigation, the alternative would be to willfully ignore the national security implications involved in modern gene-sequencing technology in foreign countries, which may give rise to bioterror attacks (Needham, 2020). In view of the threat of future pandemics resulting from BSL laboratory leaks, it is absolutely crucial that the Intelligence agencies prepare for such worst case scenarios by investigating the local environment of BSL Laboratories in different nations before outbreaks occur, and specifically conducting an in-depth investigation at Wuhan Laboratories to confirm or disprove the origin of SARS-CoV-2.

D. CONCLUSION

Due largely to an overwhelming lack of transparency by the Chinese State and obfuscation and stonewalling by Chinese Health Officials and Laboratories in Wuhan (the WIV and WCDC in particular), it is impossible to confirm whether or not SARS-CoV-2 emerged due to natural zoonotic spillover or as the result of a laboratory leak, intentional or otherwise and even if this was due to deficient biosafety regulations in China.

With the currently, available sequences, analysis based on phylogenies of complete virus genomes are not sufficient to draw firm conclusions of the evolutionary origin of SARS-CoV-2, as pointed out in recent papers by Colin Butler (2020) and Etienne Decroly (2020).

It is clear from the above, that the possible origins of the Coronavirus pandemic, SAR-CoV-2 have been seriously underestimated as a target for a formal microbial forensics investigation. Despite the fact that the WHO was mandated by 194 countries on November 12, 2020 to carry out a thorough investigation into the origins of SARS-CoV-2, recent revelations by members of the early WHO team in the NYT and National Geographic and recently published WHO documents reveal that the WHO is unable to pursue a transparent and independent investigation (Drury, 2020).

We all have witnessed just how many people have lost their lives to this virus, and how many have lost everything they have ever worked for in their lives. We therefore feel that there is an urgent need to investigate how this pandemic has originated not only for moral and ethical reasons but also for practical and scientific ones. It can only be through tracing the virus to its source that we can ever hope to prevent or predict future similar pandemics arising from zoonotic transmission via hidden reservoirs in the animal world.

Indeed, this is the current situation with mink, which have been culled in their millions in the Netherlands, Spain and Denmark, due to the fear of mink to human reverse transmission of a mutated form of the virus which may not be stopped by proposed vaccines or antibodies developed so far by humans (Briggs, 2020; Dyer, 2020; Dyer, O., 2020; ECDC, 2020; Kiro et al., 2020; Oreshkova, 2020, WHO, 2020h).

To conclude, there is no definitive evidence to date to support a “natural origin” hypothesis via an intermediate animal host as none have been found yet. Neither is there concrete evidence for a laboratory leak, or a laboratory acquired infection, however compelling the circumstantial evidence is (Mallapaty, Maxmen, & Callaway, 2021).

The authors of this paper propose nothing more nor less than what would have taken place if SARS-CoV-2 had originated in Europe or the USA or another location, that is, to reconsider the need for an open, transparent and thorough investigation into the origin of SARS-COV-2 without regard to politics or fear of embarrassment to any government.

This pandemic may or may not have been caused by a natural event, and that remains to be determined through an impartial forensic investigation. This investigation is essential for the future security of the United States and indeed the World, as the pandemic origin must be explained using evidence-based data, and as Professor Ebright pointed out: “Understanding the origin of the outbreak is a crucial step to reduce the risk of future outbreaks” (Field, 2020).

Although many months have passed since the outbreak in Wuhan and there are many barriers to an effective forensic investigation in a foreign country (Mallapaty, Maxmen, & Callaway, 2021) by a US government agency, we reiterate that such an investigation into the origins of the pandemic is essential for the security of the United States, the World, for the relatives of the almost three million deceased and above all to prevent future pandemics from destroying more human lives.

REFERENCE LIST

(Quite lengthy as it includes references from all sections of our report)

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