GREYSCAN Detection of Viruses with the Fieldable GreyScan Trace Virus Detector (TVD-1)

Background

The GreyScan ETD100™ is the only technology in the world that can test and identify inorganic trace materials, and we believe that we can quickly repurpose our existing technology to identify trace amounts of COVID-19 living on surfaces worldwide, a capability which is now essential in stopping this global pandemic. Not only would our technology be effective in identification of COVID-19 on surfaces, but virtually for any future trace level of virus, as science has proven that there are many strains.

On Tuesday, March 17, 2020, the United States National Institutes of Health (NIH) announced that the coronavirus disease of 2019 (COVID 19) is stable on surfaces for several hours and even days, similar to the original SARS virus. This new information about the stability of SARS-CoV-2, which causes the COVID-19 disease, suggests that people may acquire the virus not only through the air, but by touching contaminated objects, adding another layer of prevention needed beyond simple social distancing.

NIH Scientists at the National Institute of Allergy and Infectious Diseases in Montana have now compared the SARS-CoV-2 and SARS-CoV-1 which causes SARS. SARS-CoV-2 is now terrorizing citizens worldwide, just as SARS COV-1 infected over 8000 people in 2002 and was not eradicated effectively until 2004 via intensive contact tracing and isolation measures.

The NIH also found that most secondary cases of the virus transmission of SARS-CoV-2 appear to be occurring in community settings that are not cleaned in the same way as health care settings, thus, the virus lives on surfaces and continues to wreak havoc on society by wrecking the economy and creating a sense of panic worldwide.

The GreyScan ETD100™ was created to provide a simple to use, sensitive portable detector to determine the presence of improvised explosive devices. To GreyScan, this virus is another threat that can be detected, addressed, and prevented to protect humanity during a major crisis.

Through our technology, GreyScan can enable the public to feel safe again someday and be able to return to their normal routines. It is critical to be able to demonstrate that cleaning or decontamination protocols have been followed and to encourage trust back into society. This approach to screening and the awareness of capability gap fulfilment will allow the general public to feel safe in environments not under their own control. This will be a critical step in creating confidence in the environment to enable people to assume societal norms, reducing the economic impact of the pandemic.

The Science Behind the Solution:

Traditional techniques for virus analysis, e.g. Quantitative real-time PCR (qPCR) and DNA microarrays with probes, usually require time consuming and expensive processes. Therefore, they are not suitable for high throughput analysis. In contrast, Capillary Zone Electrophoresis (CZE) offers quick, inexpensive, and automated analysis, making it highly suitable for automated, high throughput virus analysis. Capillary Zone Electrophoresis (CZE) is an established method for biomolecule analysis [6–8] and an emerging method for clinical applications [9,10]. Capillary electrophoresis of complex biological assemblies shows great potential in bioanalysis as it offers a unique opportunity for the analyte to move freely in the liquid solution and interact with ligands, allowing investigations without steric restrictions.

The presence of distinctive surface charges and zeta potential allows for the separation of native virus and subviral particles in an electric field, previously demonstrated for poliovirus that were separated by zone

electrophoresis in the glass tubes [1]. In 1987, Hjertenet al. for the very first time demonstrated capillary zone electrophoresis (CZE) of the tobacco mosaic virus, migrating as a single peak, using UV detection [2]. Since then, there have been numerous reports on CZE analysis of viruses including adenovirus [3] and Semliki Forest virus (SFV) [4] and rhinovirus.

GreyScan is already in the process of discussing and planning potential partnerships with the world's leading experts in the application of this technology in the laboratory setting. There are several articles by Institute of Analytical Chemistry, University of Vienna on employment of CZE for analysis and characterisation of viruses. In 1996, they published the first paper by on capillary isoelectric focusing CIEF of an animal virus – human rhinovirus type 2 (HRV2) – was published [3]. This was followed by CZE analyses of different serotypes of human rhinovirus (common cold virus) [5-11]. For example, different types of human rhinovirus, the main causative virus for common cold, was analysed by CE which allowed easy assessment of rhinovirus in less than 10 mins which is otherwise challenging to analyse requiring high purity, and large sample amount to perform sensitive analysis [5].

In addition to that there is another group Department of Chemistry, University of Ottawa, Canada, which is working on quantification of viruses using capillary electrophoresis [12, 13]. The quantification of a virus is of critical significance in clinical diagnostics, vaccine development, and environmental contamination assays. This group proposed a cost- effective, robust approach based on CZE for the characterization and sensitive quantification of intact viruses [13].

The above-mentioned work on utilisation of capillary electrophoresis demonstrate the suitability of a converted ETD-100 device for applications in virus identification and quantification. Our ETD-100™ system could be used with simple modifications such as capillary changes and addition of UV or Laser induced fluorescence detection, two most common detectors for virus analysis. Furthermore, the commonly used reagents for virus analysis are also compatible with ETD-100™.

The GreyScan ETD-100™, which was created for explosive trace detection, is based on technology developed by the University of Tasmania detect inorganic materials, differentiating the device from traditional ETD technology. This technology was taken and commercialized into GreyScan and is now available as a product for use in counterterrorism and deployed worldwide. The ETD-100™ has a great potential to be adopted and repurposed into another device for medical diagnostic testing, and environmental screening of COVID-19 and other future bio threats.

These are the following points that build our confidence on the capability of ETD-100™ to be converted to a virus detector and allow quick, sensitive and inexpensive diagnosis of the presence of COVID-19.

- The ETD-100 is based on Capillary Zone Electrophoresis (CZE) – a technique previously used for the analysis and characterization of human viruses such as common cold virus (human rhinovirus serotype 2 (HRV2)) and adenovirus (cause cold like symptoms).

- Sensitive virus analysis requires staining the virus to make it fluorescent. Kremser et al (Analytical Chemistry, 2004, 76 [14] 4175-4181) have demonstrated this using RiboGreen to stain the RNA inside the capsid with a 5 to 6 hour incubation time.

- Professor Breadmore's group at the University of Tasmania have been working on cell analysis by capillary electrophoresis since 2014.

- They have been able to detect 7 cells in 0.1 mL (Analytical and bioanalytical chemistry, 2015, 407 (23) 6995-7002), used capillary electrophoresis to reduce the time to stain intact cells to 2 min (Analytical Chemistry 2017, 89(12) 6513-6520), and demonstrated a 90% recovery of bacteria from a swab in conditions compatible with their rapid staining method (unpublished).

The current ETD-100™ system is designed in such a way that it could be used for virus detection after simple modifications such as capillary fluid changes, Laser induced fluorescence detection, and different extraction swabs. The GreyScan ETD-100™ took a manual inorganic analysis from over 30 min, to completely integrated in under 2 minutes. The analysis is conducted as follows; the operator takes a swab from a surface of interest, whereupon it is inserted into the inlet of the system. The swab is then washed through a fluid solution and all trapped materials are removed and passed through a separation system where materials are separated under a high voltage.

The work to convert the explosives detection capability into virus detection would include a program with collaboration between University groups and GreyScan to repurpose the ETD™ to be able to analyze viruses and replicate the extensive work conducted in the laboratory using CZE for this application.

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Examples of Species Detection [14]:

Species	BGE	Capillary treatment	Detection
Virus			
Tobacco mosaic	20 mmol/L Tris-HCl buffer, pH 7.5	Coated with methyl-cellulose	UV
Tobacco mosaic	2.0 mmol/L potassium borate buffer, pH 8.35	Uncoated	UV
Adenovirus	25 mmol/L sodium phosphate buffer, pH 7.0	Coated with polyvinyl alcohol (PVA)	UV
Semliki forest	100 mmol/L Tris-150 mmol/L borate buffer, pH 8.2	Microdevice, coated with linear polyacrylamide	UV
Human rhinovirus	100 mmol/L borate buffer, pH 8.3, 10 mmol/L SDS as additive	Uncoated	UV, FL
	100 mmol/L borate buffer, pH 8.3, 0.5% sodium deoxycholate, 0.05% SDS, 0.5% Triton X-100R		UV

Proposed Use

The ETD-100™ is commercialized for the detection of the presence of components of explosives mixtures on swabs taken from surfaces.

GreyScan Pty Ltd proposes to repurpose an existing commercialized technology designed for detection of certain explosives to detect viruses in order to fill this critical capability gap. The GreyScan ETD 100™ was introduced this year: it is a breakthrough technology designed to detect certain categories of common explosives not readily detectable by other systems. It is based on an underlying analytical laboratory technology that has been demonstrated to be effective in the detection of viruses. The GreyScan ETD 100™ has engineered an existing analytical detection technology, Capillary Zone Electrophoresis (CZE) into a fieldable system contained within a pelican case that can be set up within ten minutes and used to screen for improvised explosive devices. This allowed for trace amounts of inorganic explosive residue to be rapidly analyzed in the field environment rather than returning samples to a laboratory for analysis.

The GreyScan ETD 100™ technology has the performance capability of a laboratory CZE system. It has been designed to be field operational, with the analysis time compressed from 20-30 minutes to less than a minute. The GreyScan has been designed for continuous throughput of samples, with low cost consumables, and a high degree of maintainability and robustness being key features of the system. The system is an environmentally and health conscious solution for trace explosive detection with no radioactive components or toxic reagents. We believe that we can design a virus detector that incorporates all these features with appropriate methods development and its transition to the current detector platform.

The GreyScan system, a $25 Million investment to date, has been able to maintain the sensitivity and separation capability of the laboratory system in a small field deployable system that can perform analysis on a time scale compatible for security screening operations. Before the GreyScan ETD 100™, a single analysis of this type of explosive using capillary zone electrophoresis took at least 30 minutes. The major engineering designs that that were necessary to design a compact, fast, fieldable screening instrument that can be run with a minimally trained operator is the baseline technology that facilitates the repurposing of this technology into a fieldable virus detector. Samples are collected from surfaces using commercially available swabs that are used to screen passengers through aviation checkpoints and by agents employed in counter terrorism screening. These swabs are currently used to collect residues of explosives but can be used to swab viruses.

The same concept of operations can be applied to the environmental screening for the presence of Corona and other viruses. The fast, mobile, cheap and accurate technique can be used by a non-expert to screen an area in hospitals, hotels, buildings, airplanes, homes, schools, factories, all workplaces etc and check that the areas are free of contamination. This technology can then be used to continuously assess the environment, and if a contaminant is found then appropriate actions can be taken. This would ease the burden of the current environment where no one knows where the virus may be. By having an environmental testing and screening procedure the government can rebuild confidence and individual businesses can ensure they’re creating a safe space for staff and customers.

Surfaces to be targeted for swabbing include high touchpoint areas in all areas, including but not limited to - hotels, shops, mailrooms, planes, vehicles, places of work, restaurants, public areas. The system chemistry including the sample removal from swab, the reagent used to carry the sample and the detector would all have to be reoptimized to look for viruses instead of explosives, but the technological concept of CZE would remain the same.

Timeline

The GreyScan technology for explosive trace detection in its current format is market ready and can be produced at volumes. The company is amenable to producing in the USA, creating much-needed new American jobs.

By implementing small changes in the chemistry of the analysis; the technology remains in the same form factor but with revised reagent and capillary chemistry. This means that the GreyScan system can provide fast, cheap, accurate and mobile detection with red screen/green screen alarm capability. The system effectively provides what a laboratory system can while offering a product that's ~ 25% of the price of a lab system, up to 30 times faster, can be carried with one hand fully mobile, and is already proven in the explosives detection counter terrorism world. The capabilities of this device would empower first responders to detect and prevent the virus while keeping them out of harm's way.

A concentrated program to alter the chemistry and reflect the work done by laboratory experts globally would enable a product to be ready within 12 months end to end depending on the resources and partnership that the U.S. Government is able to provide. However, breaking the process down into two phases as explained below, the product could be ready as early as six months.

The project will be demonstrated in two phases. Phase I will develop the CZE technology in a laboratory necessary to separate and identify the virus. During this phase, the detection method will be developed, and the sensitivity and selectivity of the method will be analyzed. At the end of this phase, a new method of virus detection will be demonstrated that is evolutionary to viral detection in that it will be faster and less complex than existing laboratory methods. The successful completion of phase I produces a cheaper, faster, simpler method for SARS-CoV-2 detection, a major win for surveillance in its own right.

Phase II will repurpose of the existing GreyScan detector for virus detection, based upon the detection method developed in Phase I. The current technology readiness level of the GreyScan ETD-100™ is TRL-9. Once the laboratory SARS-CoV-2 CZE method is developed in Phase I, the current device can be rapidly repurposed to detect this virus. The transition of the laboratory detection method to the detector will require some additional methods development. But because of the modular nature of the system we believe that major modifications of the system will be minimal. The development chemistry, detector, and software command control parameters will not be difficult but must addressed. The subsystem development will be at TRL-7 or TRL-8 and the detector type will probably be changed. The virus detector will utilize a similar aqueous phase buffer solutions and capillary tubes to accommodate the new chemistry, resulting in minimal changes to the overall system design. The product from phase two is the prototype TVD-1 that will be evaluated for field detection suitability, that would need to follow CDC protocols for handling the epidemic.

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