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Research Article

Economic, Convenient, Ultra Rapid, Mass Diagnostic for Covid-19

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ABSTRACT

Two theoretical models are presented. Model 1 (turbid/gel clot); healthy human WBCs (8-10000 µl) are put into assay tubes (12 × 75 mm) in 200 µl solution of mono and divalent cations + urea + 0.0125 g of potassium iodate + Tris buffer (0.001M) + MgCl2 (0.05 M) at pH 7-8 range, exposed to throat swabs and incubated for 1-6 hrs at 27°C. In +ve cases the assay the solution turns turbid or gels (0.0125 g of potassium iodate may be added for select area pigmentation). $(CH_2COO)_2Ca$ 10% is used to reverse the turbidity as counter validation of a +ve result exclusively. Model ii (ground glass effect). WBCs + solution as above pour on plain glass slide air dry slowly in humid-moist incubator for 1-24 hrs. Convenient-Economic-Mass Ultra Rapid Covid Test.

Keywords: Ultra rapid Covid test; Turbidimetric test; Ground glass effect on plain glass slides

INTRODUCTION

The need for a diagnostic tool for the current dreaded pandemic contagion COVID-19 (SARS-CoV-2) is well known and is the focus of all nations [1]. A safe time vetted victorious vaccine is alike the proverbial World War-II "Bridge Too Far" [2]. Additionally, of the 7.8 billion pan global sub-population 80% i.e., 6.24 Billion as on 03-2020 (temperate-to-equatorial either hemisphere only) stand the risk of time-to-time self-mediated re-inoculation leading to active infection on an average twice a year. This because a gigantic market for diagnostics in continuum (few year scope). Peerless revenue driver. The moot point is that the overwhelming majority cannot afford should the diagnostic tools be expensive and or the process be time consuming. Respective national administration shall forever remain stressed pressed while the economies shall have to be down-turned and reset in-order to subsidies such lifesaving health care services. Eventually, the tax payer has to pay for the vaccine subsidies as well. No going away. Therefore, we herein lay bare applied theoretical models that are economic, ultra rapid, convenient and mass application oriented. Up-saleable. The outcome of our models is turbid and or gel clot (wet) and ground glass phenomena alike {dry filament}. Required time being 15 to 120 minutes (model specific). The sample is taken from the fossa (throat) via swab. In either, the unknown active bio-samples get irreversibly degenerated (leaving no trace toxins) and hence become bio-safe even for bare handling (post assay). We are guided by "Social Contract" (the foundation for altruistic public administration) and Mahatma Gandhi's dictum (the Father of the Largest Democracy), respectively [3,4].

Previous experience

Picogram Sensitive Indigenous LAL test invention, India, 1995; ORLYSATE [5].

Malaria Diagnostic-DiBDoT, 2016 [6].

MATERIALS

For model 1: Healthy human WBC washed packed cell banks (suspended in isotonic NaCL) containing at least 8000-10,000 cells per µl; Endotoxin free dilution tubes (13 × 100 mm) and Assay tubes (12 × 75 mm); incubator and heating block; 25 ml stock solution of Urea Solution i.e., Carbamide in isotonic normal saline also containing 0.0125 g of potassium iodate + Tris buffer (0.001 M) + $MgCl_2$ (0.05 M) at pH 7-8.

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For model 2: Plain Glass slides; Field microscope 4 x and 10x magnification.

Common: Pipette (range 10-100 µl); tips; Sample collection swabs; Isotonic normal saline; Tris buffer; $MgCl_2$; Calcium Acetate; Urea/Carbamide CH_4N_2O; Ph check devices; Silicone coat (Sigma coat); water for injection; other standard lab gadgets and glassware. Throat swab media and kit. Optional stand by: Lugol's liquid (iodine triodide-I3K); Calcium Acetate 10% $(CH_2COO)_2Ca$ solution in isotonic NaCl; transferin and immunoglobulin(s). Solutions and pH may vary within a range of 10%-20%.

All clean glassware and media contact implements; autoclaved; siliconised and again dry heat sterilized at 200°C for 2 hrs. Mini vortex.

METHODS

For model i: In endotoxin free assay tubes take 50-100 µl of 'urea solution' add to it 50 µl to 100 µl Healthy human WBC add to it 50-100 µl swab sample collection. Vortex lightly. This may also double up as virus culture media. Place in heating block and heat @27°C for 1hour or less or longer 12; 18; 24hrs. Total volume equals to 100-200 µl. RT normal; RH >85%.

Result: If the person even be a (asymptomatic-dormant) carrier of corona virus then the media shall turn turbid. This is +ve result. Non turbid means -ve result. (Mixed media may gel and or may not over next 3-6 hrs; additional indication).

Optional: the turbid media be filamentous (starch alike proteins) and reactive RNA materials. Addition of 100 µl of I3K (amber colour) shall slow turn the media towards a light blue\violate haze over next few hrs (spectrum shift). Total volume increases to 200-300 µl. The mixed media may gel and or may not at all even post 6hrs.

Counter validation: slow top loading of $(CH_2COO)_2Ca$ @1:1 vol/vol of the total in the assay tube + re-heating for 1 hr shall result in clarification action; the turbidity shall wane; translucence shall re-set in. This is double proof of a +Ve result.

In case of -ve result the WBCs and the test material shall all precipitate over time and form a slimy deposit at the bottom.

DISCUSSION

Test protocol model i

The near stable, quite shock resistant pathogenic SARS-CoV-2 virions react with the WBCs in the presence of nitrogen (Urea solution) the neo-adjuvant catalyst for such process in micro volumes at RT in the open. Limited proteolysis happens forming filamentous polymeric protinase i.e., pre/proto fibrions alias 'turbid' to the visual spectrum [8]. The starch-urea (carbamate) is hydrophilic and swells in water yet polymeric thus shall also indicate weak gelling tendency (if undisturbed during incubation) [7]. Diffuse fibrosis of the bilateral lung fields being the end point in the pathophysiology of Covid-19 patients at end of life stage being primarily due WBC degradation herein is aided, up-regulated and stabilized by mono and divalent cations (NaCl + $MgCl_2$); well buffered by Tris [hydroxymethyle] amino methane [9].

For model (ii-a): This is a higher speed work flow model. In Endotoxin free dilution tubes take 100 µl of 'urea solution' add to it 100 µl healthy human WBC add to it 100 µl swab sample collection. Vortex lightly. Pour on plain glass slide. Draw as a film end to end or 50% of the glass slides surface area. Air dry in humid moist condition 90+ relative humidity (incubator). Time duration may vary from 1 hr to 24 hrs.

Result: Ground glass type formation shall indicate +Ve. Else, -Ve. The 'ground glass phenomena can be viewed via reading glass and or 4x field microscope. Other actions as above may be considered.

Test protocol model (ii-a)

The largest real time clinical study in china COVID-19 have described the diffuse fibrosis of the bilateral lung fields as "Ground Glass" type phenomena [10,11]. We intend to mimic it.

For model (ii-b): Research grade sterile filter paper No.1 can also be used in place of glass slide. It shall additionally indicate bands as outer rims with distinctive mid fields. The semi dry and post dry spots can be evaluated optically (reading glass\4x field microscope). This may be aligned to 'Droplet Test' as alike DiBDoT.

WBCs and all cytokines being motile and RNA virions being charge particles experience efficient intra-complementing para-magnetic paths and hence get coupled. However, the CoV-2 spp. virion completely debacalises the phagocytizing character of the WBCs and the related cytokines. It multiplies on the dead WBC as the culture bed. The pulmonary paryncheme gradually gets loaded with N gas this (reducing oxidative thrust) further assists in rapid culture. Hence, we have incorporated ions; anti-oxidising carbamate; additional healthy WBC material; buffer; etc. Once the reactions are complete no virulence is left, not even any trace toxins. Everything gets converted irreversibly to inert state.

Note: (a) Addition of transferin and immunoglobulins shall rise the unit cost; slow down the fibrosis (ground glass/turbidity) formation and also open a small window for false –ve or falls +ve in in-vitro conditions. (b) addition of $(CH_2COO)_2Ca$ @5% will impart driver potential; uniform mid field; brighter ground glass effect; rims that shall appear as dark to the incident white light and as translucent when back lit (5-7000 Kelvin); non phagocytized cell debris being vectored to form rims; trap light; shift in colour spectrum with alteration in the incident light's colour temperature.

CONCLUSION

Our aim is to mimic that what Mother Nature does in status Covid-19 (condensing the time scale). Considerable Nano-tech is involved. Yet we have designed our reactions keeping pico-sensitivity and at room stability so that it (eventually due inputs by more able minds) gets fit for the remote of the rural. Our efforts in this transaction are nascent, ground breaking and recline quite a bit on our long years of slow flow (un-aided; non-patronized) works in related domains. The hidden agenda is to enable a role for the family physician with family welfare at heart with respective national resource conservation. Ruthless-marketing model has rendered the intently focused careful cum conservative first level of well-trained all weather clinician – redundant. All societies are re-discovering the loss wrought upon. This shall be the 1st preventive screening test en-mass. More sophisticated tests may logically follow

for the +ve cases (mass screening and effective tapering down of the referral numbers). Thence and thus, the competing stake holders of the health-care-pharma-research-marketing-logistics-assurance sector stake holders shall all be in win-win situations. We shall not apply for any patent nor for any copy right. Any can file same in own or assignee's name with or without any citations of these presents. No limitations.

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