RCloud Tasking Form – Part B: Statement of Requirement (SoR)

Title of Requirement	In vitro model of viral encephalitis: The contribution of cytokines to multi-cell death and in priming naïve cells for infection.
Requisition No.	1000160801
SoR Version	**0.1**

1.	**Statement of Requirements**
1.1	**Summary and Background Information**
	Viral infection of the brain is often accompanied by inflammation, with symptoms ranging from mild headache to encephalitis with long-term neurological complications or death (Zacks MA, Paessler S. 2010). During acute inflammation, astrocytes and glia express cytokines to suppress viral replication and chemokines to attract peripheral immune cells across the blood brain barrier (BBB) to resolve the infection. However, chronic inflammation can develop into encephalitis with a 'cytokine storm' killing neurones, astrocytes and damaging the BBB, which has long-term neurological consequences and can be fatal. Cytokine storm is partly due to unique synergy between brain microglia, astrocytes and BBB endothelial cells, in close proximity to each other in the enclosed skull. Inflammatory mediators secreted by an infected cell can initiate cytokine secretion by naïve 'bystander' cells, leading to a positive feedback increasing inflammation and cell death. Our pilot work has shown that an infected glia secretome results in death of naïve bystander glia at the same rate as death caused by direct viral infection. These data point to a much more important role of inflammatory cell death in early stages of infection than was previously thought. Virus exposed neurones and BBB endothelial cells also produce inflammatory mediators (e.g. apoptotic TNFα), but we do not know how important their secretomes are for naïve cell death, compared to direct infection of cells. An unexpected consequence of an inflammatory secretome, is that it primes naïve glia to be more permissive to subsequent viral infection, so prolonging the infection. Factors implicated in this are priming by cytokines to elicit mitochondrial damage and increasing oxidative stress making cells more vulnerable to attack. This is primarily through action of TNFα and interleukin 1β. It is not known whether neurones and BBB endothelial cells might also be primed for infection in this way, and form part of the longer-term damage to the central nervous system. Anti-inflammatory drugs can successfully treat the cytokine storm of viral infections e.g. in Herpes Simplex encephalitis and Influenza encephalopathy. But for alphavirus infections in particular,

	important questions remain about the type of anti-inflammatory drug to use for example NSAIDs are detrimental to recovery from influenza encephalopathy. The timing of treatment is also a key factor, to allow beneficial acute inflammation suppressing virus, but then treat chronic inflammation-mediated cell death and naïve cell infection. While this is complicated by the range of cell types involved and the infectivity of different viruses, an in vitro cell approach will allow us to begin dissecting the important factors and timelines. In the absence of approved vaccines or anti-viral drugs for many alphaviruses, elucidating the contribution of cytokines to death of neuro-vascular cells, and their effect on viral replication will help identify additional targets for cytoprotection**.** **Such strategies are needed to both help speed recovery from infection, but also protect against long-term neurological consequences which cannot be treated by anti-viral therapies alone.**
1.2	**Requirement**
	The overall aims are for KCL to investigate the effect of the virus-induced inflammatory environment on bystander cell viability and subsequent viral infection. The study will use the alpha-virus model Semliki-Forest Virus (SFV) with GFP reporter and *in vitro* cultures of three primary cell types, neurones, astrocytes and endothelial cells. The study will focus on 3 main areas; 1) Effect of inflammatory environment on bystander cell viability • What is the contribution of an infected-glia secretome on cell death in bystander neurones and astrocytes, compared to direct infection? Which cell type is more vulnerable? • Do virus-exposed BBB endothelia produce inflammatory factors that affect bystander astrocyte and neurone viability? • Are virus-exposed endothelial cells a source of persistent (long-term) inflammatory mediators? • Do infected neurones contribute to early inflammation? What is effect of their secretome on bystander neurones and astrocytes? 2) Enhanced viral infection of bystander cells: • Does an inflammatory environment (especially IL-1β), prime naïve astrocytes and neurones to sustain a higher viral load, particularly during the early stages of infection? • Can this also enable endothelial cell infection, which are usually more resistant to viral replication? • Can bystander cell infection be modified with antioxidants or anti-IL-1β treatments? 3) Anti-inflammatory therapy: • Can bystander cell death or viral replication be attenuated by anti-inflammatory drugs? • What is the optimum treatment window to allow acute beneficial response to infection, but reduce chronic detrimental effects?

1.3	**Options or follow on work** *(if none, write 'Not applicable')*
	Depending on data generated from this period of performance, DSTL may request additional work to be conducted around enhanced characterisation of BBB infection models.
1.4	**Health & Safety, Environmental, Social, Ethical, Regulatory or Legislative aspects of the requirement**
	It is expected that KCL Laboratories will have extant Risk Assessments, SOPs or processes in place for the study to follow. These will be made available to Dstl if requested.

1.5	Deliverables & Intellectual Property Rights (IPR)					
Ref.	**Title**	**Due by**	**Format**	**Expected classification (subject to change)**	**What information is required in the deliverable**	**IPR Condition**
D1	Quarterly Progress and Technical Review	T0+3 Months	Presentation (.pptx)	Redacted under FOIA Section 23 – National S	Quarterly written report and PowerPoint Presentation pack to include but not limited to: • Update on technical progress • Progress report against project schedule.	DEFCON 705
D2	Final Progress Report	T0+12 Months	Written Report (pdf/word)	Redacted under FOIA Section 23 – National Se	Summary of findings from the 12 month study, conclusions and recommendations for further work if appropriate.	DEFCON 705

1.6	**Deliverable Acceptance Criteria**
	All Reports included as Deliverables under the Contract e.g. Progress and/or Final Reports etc. must comply with the Defence Research Reports Specification (DRRS) which defines the requirements for the presentation, format and production of scientific and technical reports prepared for MoD. • Interim or Progress Reports: The report should detail, document, and summarise the results of work done during the period covered and shall be in sufficient detail to comprehensively explain the results achieved; substantive performance; a description of current substantive performance and any problems encountered and/or which may exist along with proposed corrective action. An explanation of any difference between planned progress and actual progress, why the differences have occurred, and if behind planned progress what corrective steps are planned. • Final Reports: shall describe the entire work performed under the Contract in sufficient detail to explain comprehensively the work undertaken and results achieved including all relevant technical details of any hardware, software, process or system developed there under. The technical detail shall be sufficient to permit independent reproduction of any such process or system. All Reports shall be free from spelling and grammatical errors and shall be set out in accordance with the Statement Of Requirement (1) herein. Failure to comply with the above may result in the Authority rejecting the deliverables and requesting re-work before final acceptance.

2	**Evaluation Criteria**
2.1	Method Explanation
	The suppliers proposal shall be assessed on the following basis: • Technical – assurance that the supplier has the technical capability to meet this single source requirement • Commercial – assurance that the supplier can meet the requested commercial requirements as detailed below The placing of any contract will depend upon consideration of the proposal received and the Authority reserves the right, at its sole discretion, not to proceed to contract for any part or all of a contractors proposal. And if necessary, not to place any contract as a result.
2.2	Technical Evaluation Criteria

	The supplier shall provide evidence to demonstrate that they can meet the full requirement as outlined in section 1.2 of this SOR. This should be presented in a technical proposal redacting any commercially sensitive information.
2.3	Commercial Evaluation Criteria
	The supplier shall provide evidence to demonstrate that they can meet the following commercial requirements; • A completed 'Tasking Order Form' confirming a resulting contract will be in accordance with the R-cloud Version 4 Terms and Conditions • The supplier must provide their full FIRM price breakdown for all costs to be incurred to fulfil this requirement, including: What rates are being used for what Grade (using their respective R-Cloud Grades), Quantity of manpower hours per Grade, Materials costs Facility costs, Profit rate applied, Any sub-contractor costs and the level of sub-contracting required, Any other costs applicable to this requirement. The Authority will assess the proposal to ensure that all costs are fully detailed, in line with the R-Cloud rates and price shall be commensurate with the work to be undertaken. When placing any contract the Authority is required to satisfy itself that the agreed price represents Value for Money (VFM). In single source contracting you must provide to the Authority sufficient information in support of your price proposal and during subsequent price negotiation, to enable the Authority to fulfil its obligation to assure VFM. The Authority approaches all contract pricing on the basis of the NAPNOC principle (No Acceptable Price, No Contract). The Authority reserves the right to not enter into any contract that is unacceptably priced or unaffordable.