Catch me if you can: SARS-CoV-2 detection in brains of deceased patients with COVID-19

In the American biographical crime film *Catch Me If You Can*, FBI agent Carl Hanratty goes all out to catch the notorious impostor and cheque counterfeiter Frank Abagnale Jr. Hanratty's dogged pursuit of the culprit bears striking resemblance to current COVID-19 research efforts to find evidence of changes that severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection might leave in the brain. In *The Lancet Neurology*, Jakob Matschke and colleagues[1] give a detailed account of the histological alterations related to COVID-19 in the CNS. Through meticulous detective work, they mapped the brain's immunoinflammatory response to viral infection and detected SARS-CoV-2 protein expression in a substantial percentage of autoptic brains of patients with COVID-19.

Matschke and colleagues analysed 43 brains from a large cohort of patients who died with COVID-19,[2] focusing on inflammatory changes and detection of SARS-CoV-2. To find out which CNS cell types are prone to SARS-CoV-2 infection, the authors screened gene expression datasets for signatures related to viral entry and persistence. Their in-silico analysis showed high expression of angiotensin-converting enzyme 2 (*ACE2*) in oligodendrocytes and of transmembrane serine proteases 2 and 4 (*TMPRSS2* and *TMPRSS4*) in neurons—genes that code for proteins crucially implicated in SARS-CoV-2 host-cell entry (ACE2) and proteolytic priming of the virus-decorating spikes (TMPRSS2).[3] Interestingly, a study using human brain organoids, published in September, 2020,[4] showed that SARS-CoV-2 can readily infect and kill neurons. The neuronal cell death upon viral infection was preceded by aberrant intraneuronal localisation and hyperphosphorylation of Tau protein,[4] similar to the pathogenesis of Alzheimer's disease and other neurodegenerative diseases.

Matschke and colleagues used quantitative RT-PCR (qRT-PCR) and immunohistochemistry with antibodies against nucleocapsid and spike proteins to detect SARS-CoV-2. Whereas viral RNA was detected in 48% of cases, viral protein detection was positive in 40%. Overall, the authors found SARS-CoV-2, either viral RNA or viral protein (or both), in 51% of the brains investigated. Remarkably, SARS-CoV-2 presence did not correlate with the severity of neuropathological alterations. While the replicative and infective potential of the viral RNA remains unclear, the in-situ detection of SARS-CoV-2 proteins is an important finding, as it confirms the presence of the virus in the brain. In this context, concerns[5,6] that the comparably low viral genome levels detectable by qRT-PCR in brain tissue might be blood-derived deserve mention.

Of note, the authors found virus protein expression to be confined to the medulla oblongata and to cranial nerves originating from the lower brainstem (most likely glossopharyngeal or vagal nerve). Considering the capability of SARS-CoV-2 to infect human gut enterocytes as well as pneumocytes,[7,8] this finding is of particular interest, warranting future investigations of vagal nerve tissue as a potential viral CNS access route in COVID-19.

The study also identified pronounced, brainstem-accentuated microglia activation, confirming previous work.[5] As these brain-resident macrophage-like innate immune cells are highly heterogeneous with regard to gene expression, regional abundance, and perhaps functions,[9] it seems worth testing whether microglia activated in a COVID-19 context correspond to a specific subtype,[10] expressing sets of genes reflective of particular functional states.

Given the complex pathophysiology of COVID-19, any autopsy study is bound to have limitations (varying postmortem intervals, incomplete or lacking clinical data, etc) and the present study is no exception in that regard. Confounding factors, such as the multiple comorbidities present among older patients with COVID-19 and, equally important, common COVID-19 treatment modalities, such as invasive ventilation (which might promote cerebral microbleeds) or dexamethasone medication (known to modulate immune responses), have to be considered when interpreting neuropathological findings. In the context of dexamethasone, it is unfortunate that no data on steroid medication were used to investigate correlations between some of the findings. Likewise, in the absence of appropriate control cohorts, it remains unclear to what extent microglia activation and brain infiltration by cytotoxic T-lymphocytes represent COVID-19-specific findings. Both sparse lymphocytic infiltrates and microglia activation were recently documented in the brains of

individuals without COVID-19, and they appeared to be particularly pronounced in septic cases.[5]

At a time when a potential second wave of infections is increasingly becoming of global concern, the question of whether the neuropathological alterations in COVID-19 directly result from SARS-CoV-2 brain infection as opposed to reflecting sequelae of an overstimulated systemic immune response is of high clinical importance. Whereas the first scenario would support the use of remdesevir or other antivirals, anti-inflammatory modalities appear to be the treatment of choice once damaging immunoinflammatory mechanisms take over. Teasing apart these fundamentally different scenarios is an ongoing task for neuropathology experts. The work by Matschke and colleagues[1] represents an important step towards navigating the complex pathophysiology of COVID-19 in the brain. Just like agent Hanratty, the authors have done a superb job closing in on the culprit.

Stephan Frank
stephan.frank@usb.ch

Division of Neuropathology, Institute of Medical Genetics and Pathology, University Hospital Basel, University of Basel, CH-4031 Basel, Switzerland

1   Matschke J, Lütgehetmann M, Hagel C, et al. Neuropathology of patients with COVID-19 in Germany: a post-mortem case series. *Lancet Neurol* 2020; published online Oct 5. https://doi.org/10.1016/S1474-4422(20)30308-2.
2   Edler C, Schröder AS, Aepfelbacher M, et al. Dying with SARS-CoV-2 infection—an autopsy study of the first consecutive 80 cases in Hamburg, Germany. *Int J Legal Med* 2020; **134:** 1275–84.
3   Hoffmann M, Kleine-Weber H, Schroeder S, et al. SARS-CoV-2 cell entry depends on ACE2 and TMPRSS2 and is blocked by a clinically proven protease inhibitor. *Cell* 2020; **181:** 271–80.
4   Ramani A, Müller L, Ostermann PN, et al. SARS-CoV-2 targets neurons of 3D human brain organoids. *EMBO J* 2020; published online Sept 2. DOI:10.15252/embj.2020106230.
5   Deigendesch N, Sironi L, Kutza M, et al. Correlates of critical illness-related encephalopathy predominate postmortem COVID-19 neuropathology. *Acta Neuropathol* 2020; **140:** 583–86.
6   Solomon IH, Normandin E, Bhattacharyya S, et al. Neuropathological features of COVID-19. *N Engl J Med* 2020; **383:** 989–92.
7   Lamers MM, Beumer J, van der Vaart J, et al. SARS-CoV-2 productively infects human gut enterocytes. *Science* 2020; **369:** 50–54.
8   Schaefer IM, Padera RF, Solomon IH, et al. In situ detection of SARS-CoV-2 in lungs and airways of patients with COVID-19. *Mod Pathol* 2020; published online June 19. https://doi.org/10.1038/s41379-020-0595-z.
9   Masuda T, Sankowski R, Staszewski O, Prinz M. Microglia Heterogeneity in the Single-Cell Era. *Cell Rep* 2020; **30:** 1271–81.
10   Böttcher C, Schlickeiser S, Sneeboer MAM, et al. Human microglia regional heterogeneity and phenotypes determined by multiplexed single-cell mass cytometry. *Nat Neurosci* 2019; **22:** 78–90.

Biomarkers for dementia: too soon for routine clinical use

Dementia biomarkers are valuable research tools, providing glimpses of brain pathology and function previously only available from post-mortem investigations. Before large-scale adoption in clinical practice, however, these technologies should show that they contribute meaningfully, cost-effectively, and sustainably to patient care.

In *The Lancet Neurology*, Gaël Chételat and colleagues present a Personal View on the order in which use of PET-enabled dementia biomarkers should be considered in clinical practice.[1] The authors advocate the use of these biomarkers largely on the basis of diagnostic specificity established in controlled research conditions, rather than of added value for patient outcomes in real-life settings. Moreover, the authors promote the increased clinical use of PET-biomarkers in two ways. Firstly, by presenting the implementation of biomarkers beyond tertiary (research) centres as simply a matter of practical feasibility. Secondly, by suggesting that biomarkers might be indicated in populations beyond those covered by current so-called appropriate use criteria.[2] Consequently, the Personal View contributes to the further normalisation of

biomarkers as routine diagnostic tests, which in our view might be premature. It reinforces the shift towards a biological definition of dementia, a development that does not automatically benefit patients and carers and can introduce several hazards.[3]

Firstly, it is unclear to what extent patients with dementia benefit from a biomarker-based diagnosis. As Chételat and colleagues suggest, biomarkers facilitate an aetiological diagnosis, which is indicated when pathobiological information "is desired and considered meaningful for individual clinical reasons".[1] The crucial question is who decides what is desired and meaningful: the patient (and their carer), the clinician, or the clinician-scientist? Unfortunately, insights into whether patients value having access to biomarker knowledge are scarce and often weakened by framing bias (ie, influencing the responses obtained in patient surveys through the inclusion of non-neutral information and questions). Nonetheless, there are indications that patients, carers, and citizens (as potential future patients) consider aetiological biomarker information relevant if it helps address a clear need, both in the context of presymptomatic