新型冠状病毒信息简报

第 56 期（2020 年 5 月 23-29 日报）

上海科技大学免疫化学研究所

生物医学大数据平台和高通量筛选平台领衔编译制作

联系人：蒋立春 jianglch@shanghaitech.edu.cn

内容介绍

分类	标题名称
疫情播报	1. 2020 年 5 月 28 日疫情
流行病学	2. 西雅图 SARS-CoV-2 爆发初期,在医院看病儿童中的血清阳性率 3. 初级污水污泥中 SARS-CoV-2 RNA 浓度是 COVID-19 暴发动态的先行指标
疾病检测	4. 我国总共批准了 39 个新冠病毒检测试剂盒 5. FDA 于 2020 年 5 月 9 日 批准了第一个基于抗原检测的新冠试剂盒 6. 两种 SARS-CoV-2 快速指尖血抗体联合检测方法的性能评价 7. 用于 CRISPR 快速诊断的电场驱动微流控技术及其在 SARS-CoV-2 检测中的应用 8. 基于微球的抗 SARS-CoV-2 抗体检测方法的改进 9. 清华大学研制出了 30 分钟核酸检测的掌上仪器 10. 使用免疫层析检测口水中的 SARS-CoV-2 11. Ginkgo Bioworks 获得 7000 万美金投资，将建立基于二代测序的新冠病毒检测能力 12. 自 5 月初起 FDA 要求必须经过真实病人样品测试,用于 COVID-19 的分子诊断才可能获得紧急授权
疾病病理	13. 恢复期个体对 SARS-CoV-2 感染的趋同抗体反应 14. 一种系统的炎症研究方法确定 SARS-CoV-2 感染的治疗靶点 15. 对 COVID-19 患者的深层免疫分析揭示了患者的异质性和独特的免疫类型，及其对治疗干预的意义 16. 单细胞分析显示严重 COVID-19 感染时 T 细胞超活化和瘫痪 17. SARS-CoV-2 的病理生理学：靶向内皮细胞会导致血栓性微血管病变和异常免疫反应的复杂疾病,来自西奈山 COVID-19 尸检经验
药物研发	18. 细菌分泌的一种脂酶具备广谱地杀黄病毒、SARS-COV-2 以及其他披膜病毒的能力 19. 用于 SARS-CoV-2 研究的人支气管类器官的产生 20. 一种用于检测 SARS-CoV-2 中和抗体的高通量临床检测方法——IMMUNO-COV 的开发和验证 21. 通过分析 SARS-CoV-2 感染的细胞的形态学来确定治疗 COVID-19 的药物再利用候选药物
疫苗研发	22. 冠状病毒疫苗研制成功的三个关键参数
临床试验	23. 芦可替尼治疗重症冠状病毒疾病 2019(COVID-19)：一项多中心、单盲、随机对照试验 24. 瑞德西韦治疗 Covid-19 重症患者 5 天或 10 天的疗效比较 25. 一项目多国临床试验中氯喹或者羟氯喹对于治疗 COVID-19 无

	效 26. 纵向实验室检测结果表明 COVID-19 病人有非常独特的凝血症信号 27. 在 SARS 恢复病人和未感染人群中预存针对 SARS-CoV-2 的特异性 T 细胞免疫有差异 28. COVID-19 病人血清中的蛋白质组和代谢组学特征
基础研究	29. 肠道炎症调节 ACE2 和 TMPRSS2 的表达，并可能与 SARS-CoV-2 相关疾病发病机制的重叠 30. SARS-CoV-2 的反向遗传学揭示呼吸道里存在梯度感染 31. ACE2 诱导 SARS-CoV-2 Spike 蛋白构象变化的结构基础 32. SARS-CoV-2 Spike 三聚体与 ACE2 二聚体相互作用，并存在有限的 Spike 内亲和力 33. SARS-COV-2 病毒的 ORF8 基因编码的蛋白质下调 MHC-I 的表达帮助病毒进行逃逸免疫 34. SARS-CoV-2 中一个之前没有被研究过的重叠基因和 COVID-19 大流行的进化起源有关系 35. 系统预测 SARS-COV-2 刺突蛋白上的所有非同义突变对蛋白稳定性以及受体结合亲和力的影响 36. 通过生物物理模型预测感染 SARS-COV-2 的转录和翻译过程对抗病毒最有效 37. 对 COVID-19 病人的血液样品进行单细胞表达测序以及 VDJ 测序
疾病模型	38. 一个用 CRISPR/Cas9 构建的表达人 ACE2 的小鼠模型 39. 用腺病毒构建的表达人 ACE2 基因的用于 SARS-CoV-2 研究的小鼠模型

1. 2020 年 5 月 28 日疫情

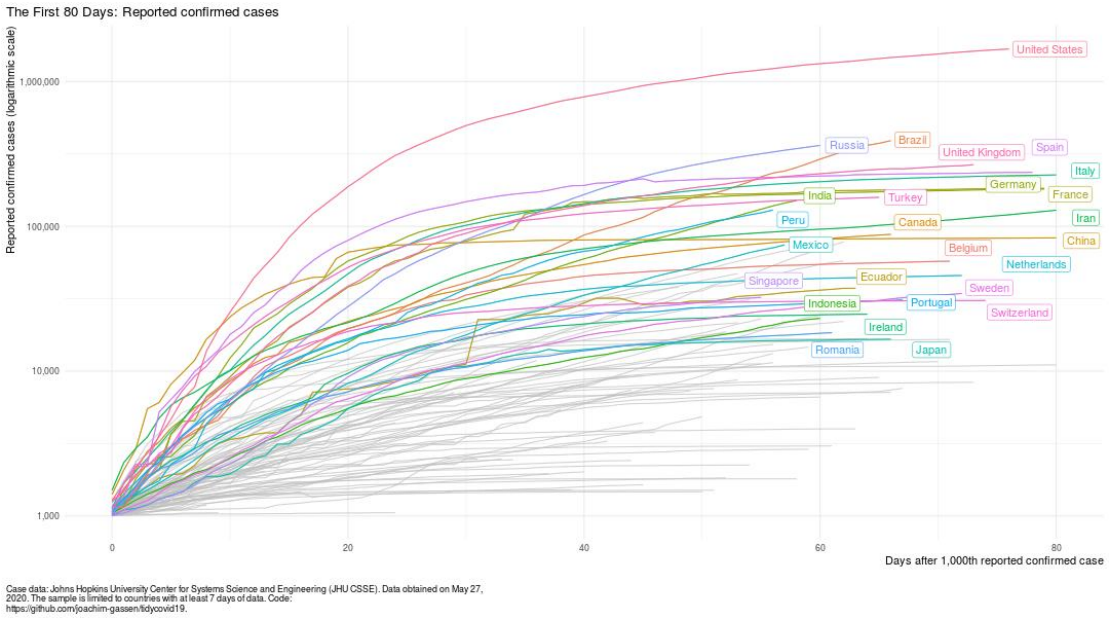
数据来源：WHO

发布时间：2020 年 5 月 28 日北京时间下午 4 点

链接：https://www.who.int/emergencies/diseases/novel-coronavirus-2019/situation-reports

根据 WHO 提供的数据，2020 年 5 月 28 日全球累计确诊新型冠状病毒病人 5593631 例，当日新增确诊 104505 例，累计死亡 353334 例，当日新增死亡 4221。

中国累计确诊 84547 例，累计死亡 4645 例，当日新增确诊 3 例，新增死亡 0 例。



重点国家确诊数量曲线（https://jgassen.shinyapps.io/tidycovid19/，数据截止 5 月 27 日北京时间下午 4 点）

全国新型冠状病毒肺炎新增确诊病例分布图（5 月 28 日，来源：http://2019ncov.chinacdc.cn/2019-nCoV/）

2. 西雅图 SARS-CoV-2 爆发初期,在医院看病儿童中的血清阳性率

Seroprevalence of SARS-CoV-2 among children visiting a hospital during the initial Seattle outbreak

来源：medrxiv

发布时间：2020-05-28

链接：https://www.medrxiv.org/content/10.1101/2020.05.26.20114124v1

第一作者：Adam S. Dingens

通讯作者：Janet A. Englund, Jesse D. Bloom

通讯作者单位：

Basic Sciences and Computational Biology, Fred Hutchinson Cancer Research Center, Seattle, WA 98109, USA

Department of Genome Sciences, University of Washington, Seattle, WA 98195, USA

Division of Infectious Disease, Seattle Children's Hospital, Seattle, WA 98105, USA

Department of Pediatrics, University of Washington, Seattle, WA 98195, USA

Howard Hughes Medical Institute, Seattle, WA 98103, USA

DOI 或 PUBMED ID：

编译者：王玮

中文摘要：

儿童在 COVID-19 案件中的代表性明显不足。在美国，儿童占人口的 22%，但仅占确诊的 SARS-CoV-2 病例的 1.7%。一种可能性是，基于症状的病毒检测不太可能识别受感染的儿童，因为他们通常比成人经历更温和的疾病发展过程。为了更好地评估小儿 SARS-CoV-2 感染的频率，该研究对在 2020 年 3 月和 4 月期间来西雅图儿童医院求医的 1076 名儿童的 1775 份残余样本进行了血清学筛查。三月份只有一名儿童血清阳性，但四月份有九名血清阳性，这一时期的血清流行率超过 1%。大多数血清阳性儿童（8/10）没有被怀疑患有 COVID-19。大多数血清阳性儿童的血清具有中和活性，其中一种在稀释度大于 1:18000 时依然有中和作用。因此，在寻求医疗的儿童中，尽管很少有病毒检测呈阳性，但在西雅图爆发早期，SARS-CoV-2 感染的频率明显增加。

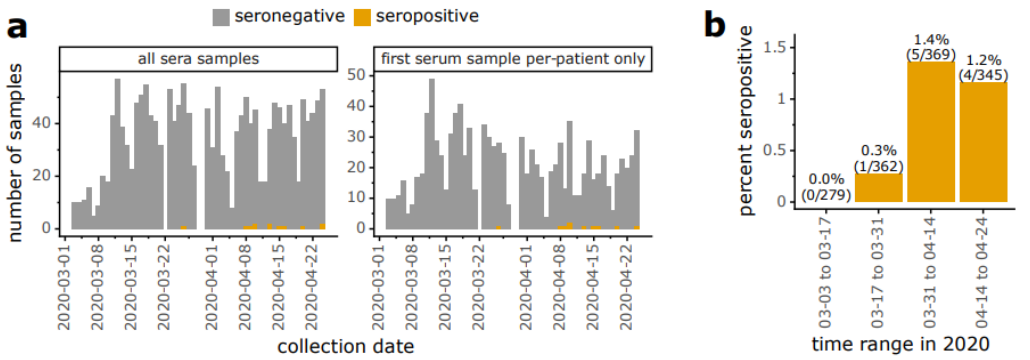


Figure 2: Seroprevalence over time. **(a)** Total and seropositive samples collected each day in the study period, with stacked bars showing seropositive samples in orange and seronegative ones in gray. The left panel shows all 1,775 residual samples, while the right panel shows only the first sample from each of the 1,075 patients. **(b)** Period seroprevalence in two-week intervals. Bars show percentage of tested patients with at least one seropositive sample during each period. Seroprevalence was significantly higher from March 31 to April 24 than March 3 to March 31 (P = 0.02, Fisher exact test, two-sided).

Abstract:
Children are strikingly underrepresented in COVID-19 case counts. In the United States, children represent 22% of the population but only 1.7% of confirmed SARS-CoV-2 cases. One possibility is that symptom-based viral testing is less likely to identify infected children, since they often experience milder disease than adults. To better assess the frequency of pediatric SARS-CoV-2 infection, we serologically screened 1,775 residual samples from Seattle Children's Hospital collected from 1,076 children seeking medical care during March and April of 2020. Only one child was seropositive in March, but nine were seropositive in April for a period seroprevalence of >1%. Most seropositive children (8/10) were not suspected of having had COVID-19. The sera of most seropositive children had neutralizing activity, including one that neutralized at a dilution >1:18,000. Therefore, among children seeking medical care, the frequency of SARS-CoV-2 infection increased markedly during the early Seattle outbreak despite few positive viral tests.

3. 初级污水污泥中 SARS-CoV-2 RNA 浓度是 COVID-19 暴发动态的先行指标

SARS-CoV-2 RNA concentrations in primary municipal sewage sludge as a leading indicator of COVID-19 outbreak dynamics

来源：medRxiv

发布时间：2020-05-22

链接：https://www.medrxiv.org/content/10.1101/2020.05.19.20105999v1

第一作者：Jordan Peccia, Alessandro Zulli, Doug E. Brackney

通讯作者：Jordan Peccia, Saad B. Omer

通讯作者单位：耶鲁大学

DOI 或 PUBMED ID：

编译者：宋张悦

中文摘要：

本研究报道了美国东北部城市地区春季 COVID-19 暴发期间，初级污水污泥中 SARS-CoV-2 RNA 浓度的时间进程。所有环境样本均检测到 SARS-CoV-2 RNA，调整滞后时间后，病毒 RNA 浓度与 COVID-19 流行病学曲线(R^2=0.99)和当地住院人数(R^2=0.99)高度相关。**SARS-CoV-2 RNA 浓度是一个先行指标，比统计的 COVID-19 检测数据提前 7 天（Figure 1），比当地医院的入院数据领先 3 天（Figure 2）。**实施或放宽公共卫生措施和限制的决定需要及时了解社区的疫情动态。

当城市废水排入污水处理厂后，固体物会沉淀并收集到一个称为（初级）污水污泥的基质中。本研究在 COVID-19 疫情期间，从 2020 年 3 月 19 日至 2020 年 5 月 1 日，在美国康涅狄格州纽黑文(CT)大都会地区，每天从为大约 20 万居民服务的污水处理厂收集初级污泥样本。SARS-CoV-2 病毒 RNA 可在所有检测样本中检测到，范围从 $1.7 \times 10^3 - 4.6 \times 10^5$ 病毒 RNA 拷贝 mL-1。

Abstract:
We report a time course of SARS-CoV-2 RNA concentrations in primary sewage sludge during the Spring COVID-19 outbreak in a northeastern U.S. metropolitan area. SARS-CoV-2 RNA was detected in all environmental samples and, when adjusted for

the time lag, the virus RNA concentrations were highly correlated with the COVID-19 epidemiological curve (R^2=0.99) and local hospital admissions (R^2=0.99). SARS-CoV-2 RNA concentrations were a seven-day leading indicator ahead of compiled COVID-19 testing data and led local hospital admissions data by three days. Decisions to implement or relax public health measures and restrictions require timely information on outbreak dynamics in a community.

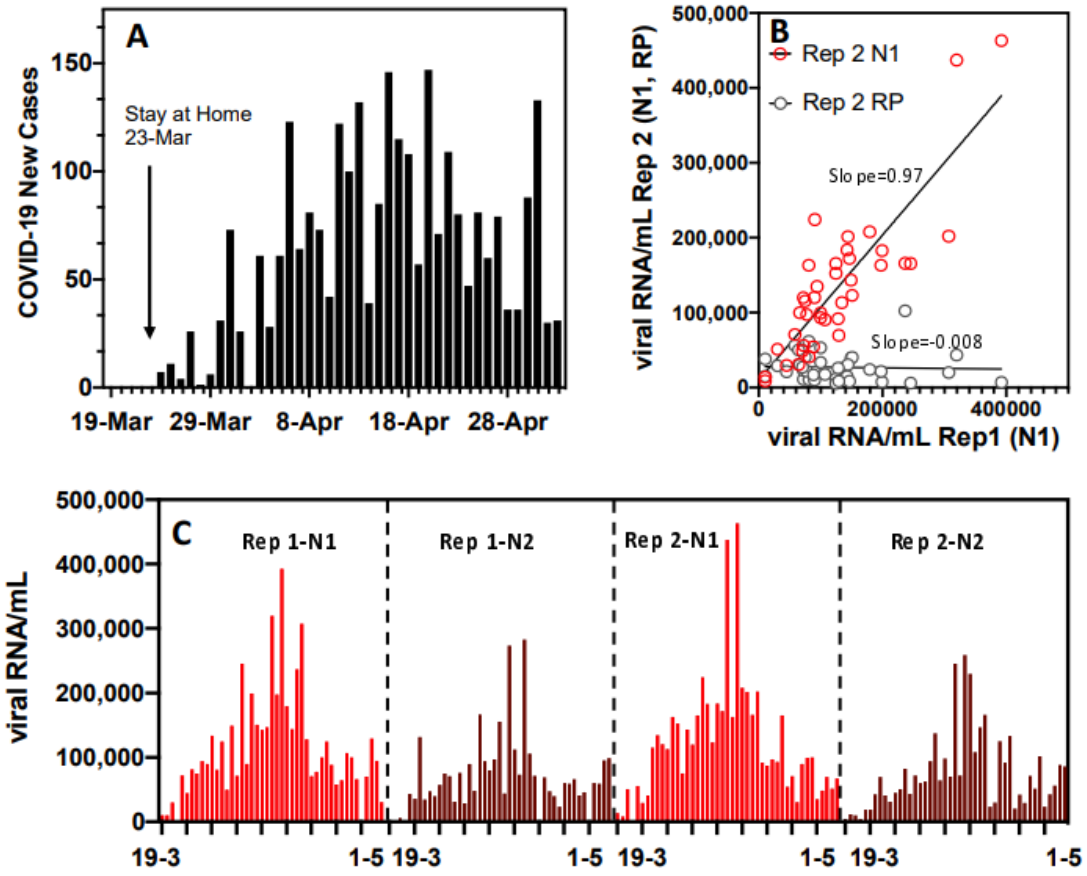


Figure 1. (A) Epidemiology curve for COVID-19 new cases (based on testing) from the cities in the wastewater catchment area (New Haven, East Haven, and Hamden, CT) served by the East Shore Water Pollution Abatement Facility (ESWPAF)1; **(B)** Example comparison of SARS-CoV-2 virus RNA replicates (N1 primer set) and comparison between N1 primer concentrations versus human RP concentrations; **(C)** SARS-CoV-2 virus RNA concentration time course for all replicates and primers considered.

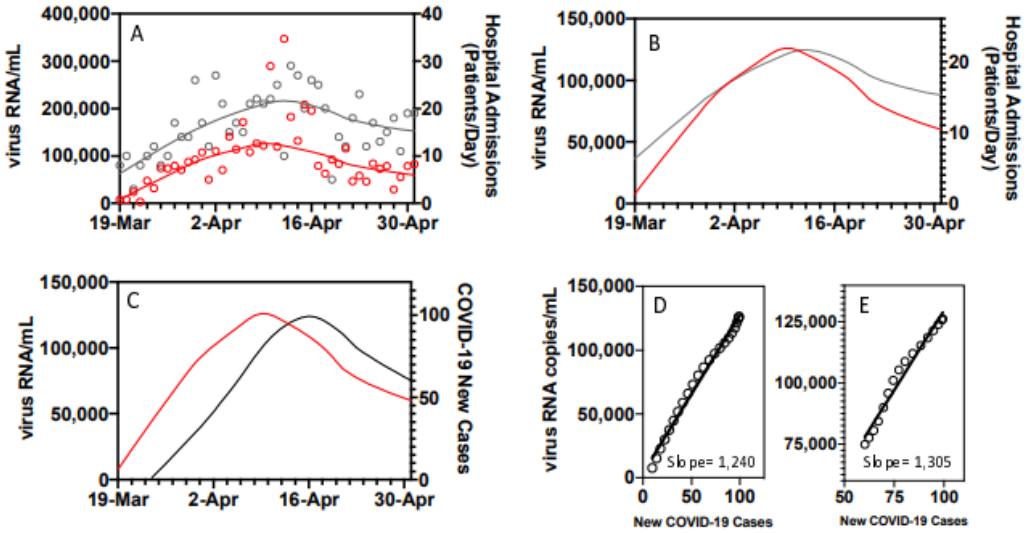


Figure 2. (A) Average sludge SARS-CoV-2 RNA concentration time course data (o) and average hospital admissions data (o) with LOWESS smoothing; **(B)** rescaled smoothed SARS-CoV-2 virus RNA concentrations (---) and hospital admissions (---); **(C)** smoothed sludge SARS-CoV-2 virus RNA concentration (---) with smoothed COVID19 epidemiology curve (---); **(D)** regression between smoothed virus RNA and new COVID-19 cases (ascending), slope=1,240 virus RNA copies/new case, R^2 =0.99; **(E)** regression between smoothed virus RNA and new COVID-19 cases (descending), slope=1,305 virus RNA copies/new case, R^2 =0.97.

4. 截止 5 月 29 日我国总共批准了 39 个新冠病毒检测试剂盒

截止 2020 年 5 月 29 日，我国总共批准了 39 个新冠病毒检测试剂盒，其中基于核酸检测的 20 个，基于抗体检测的 19 个。参考文件：国家药监局新型冠状病毒检测试剂注册信息 20200529.xlsl。

5. FDA 于 2020 年 5 月 9 日 批准了第一个基于抗原检测的新冠试剂盒

链接：https://www.fda.gov/news-events/press-announcements/coronavirus-covid-19-update-fda-authorizes-first-antigen-test-help-rapid-detection-virus-causes

6. 两种 SARS-CoV-2 快速指尖血抗体联合检测方法的性能评价

Evaluation of performance of two SARS-CoV-2 Rapid whole-blood finger-stick IgM-IgG Combined Antibody Tests

来源：medRxiv

发布时间：2020-05-27

链接：https://www.medrxiv.org/content/10.1101/2020.05.27.20112888v1

第一作者：Thierry Prazuck

通讯作者：Thierry Prazuck

通讯作者单位：Service des maladies infectieuses CHR Orléans, 14 avenue de l'hôpital, 45 067 Orléans, France

DOI 或 PUBMED ID：Preprint

编译者：孔娟

中文摘要：

SARS-CoV-2 病毒是被称为 COVID-19 的传染性呼吸道疾病的主要原因。为了应对日益增长的 COVID-19 大流行，已经开发了 29 种快速诊断检测方法(RDTs)，用于检测人全血中抗 SARS-CoV-2 的特异性抗体 IgG 和 IgM。研究中评估了两种快速诊断方法的性能，即 COVID-PRESTO 和 COVID-DUO，并与金标 RT-PCR 方法进行了比较。

方法：SARS-CoV-2 的 RT-PCR 检测样本来自医院传染病科成人患者的鼻咽拭子样本。在疾病发作后不同时间点采集的指尖全血样本用于 RDTs 进行检测。RDTs 检测的敏感性和特异性和 RT-PCR 检测结果进行了比较。

结果：381 名具有 COVID-19 症状的患者中，143 名 RT-PCR 检测阴性。RT-PCR 检测阴性的 143 名患者 RDTs 测试的结果均为阴性，结果表明两种 RDTs 的特异性为 100%。RT-PCR 阳性亚组(n=238)中，133 名患者接受了 COVID-PRESTO 检测，129 名患者接受了 COVID-DUO 检测(24 名患者同时接受了两种检测)。结果显示症状从采集之日起出现的时间越长，敏感度就越高。COVID-PRESTO 试验的灵敏度从 0 到 5 天前出现第一个症状的患者的 10.00%到在试验日期前 15 天以上出现症状的患者的 100%不等。对于 COVID-DUO 试验，灵敏度范围从 35.71% [0-5 天]到 100% (> 15 天)。

结论：COVID-PRESTO 和 DUO 的 RDTs 特异性较强(无假阳性)，并且在症状出现 15 天后具有较高的敏感性。这些易于使用 IgG/IgM 抗体组合检测试剂盒是第一个允许通过手指穿刺来筛查毛细血管血液样本的试剂盒。这些快速检测更有利于在低资源环境下 COVID-19 的检测。

Abstract

Background: The SARS-CoV-2 virus is responsible for the infectious respiratory disease called COVID-19(COronaVIrus Disease). In response to the growing COVID-19 pandemic, Rapid Diagnostic29 Tests (RDTs) have been developed to detect specific antibodies, IgG and IgM, to SARS-CoV- 2 virus in human whole blood. We conducted a real-life study to evaluate the performance of two RDTs, COVID-PRESTO® and COVID-DUO®, compared to the gold standard, RT-PCR.

Methods: RT-PCR testing of SARS-Cov-2 was performed from nasopharyngeal swab specimens collected in adult patients visiting the infectious disease department at the hospital (Orléans, France). Fingertip whole blood samples taken at different time points after onset of the disease were tested with RDTs. The specificity and sensitivity of the rapid test kits compared to test of reference (RT-PCR) were calculated.

Results: Among 381 patients with symptoms of COVID-19 who went to the hospital for a diagnostic, 143 patients were RT-PCR negative. Results of test with RDTs were all negative for these patients, indicating a specificity of 100% for both RDTs. In the RT-PCR positive subgroup (n=238), 133 patients were tested with COVID-PRESTO® and 129 patients were tested with COVID-DUO® (24 patients tested with both). The further the onset of symptoms was from the date of collection, the greater the sensitivity. The sensitivity of COVID-PRESTO® test ranged from 10.00% for patients having experienced their 1st symptoms from 0 to 5 days ago to 100% in patients where symptoms had occurred more than 15 days before the date of tests. For COVID-DUO® test, the sensitivity ranged from 35.71% [0-5 days] to 100% (> 15 days).

Conclusion: COVID-PRESTO® and DUO® RDTs turned out to be very specific (none false positive) and to be sensitive enough after 15 days from onset of symptom. These easy to use IgG/IgM combined test kits are the first ones allowing a

screening with capillary blood sample, by typing from a finger prick. These rapid tests are particularly interesting for screening in low resource settings.

7. 用于 CRISPR 快速诊断的电场驱动微流控技术及其在 SARS-CoV-2 检测中的应用

Electric-field-driven microfluidics for rapid CRISPR-based diagnostics and its application to detection of SARS-CoV-2

来源：bioRxiv

发布时间：2020-05-22

链接：https://www.biorxiv.org/content/10.1101/2020.05.21.109637v1

第一作者：Ashwin Ramachandran

通讯作者：Juan G. Santiago

通讯作者单位：美国斯坦福大学

DOI 或 PUBMED ID：https://doi.org/10.1101/2020.05.21.109637

编译者：刘焕珍

中文摘要：

COVID-19 在世界各地的迅速传播表明：人们在应对新型病原体的能力上存在重要差距。为了防止新疾病的全球传播，快速、准确和易于配置的分子诊断检测势在必行。基于 CRISPR 的诊断方法是可现场部署的解决方案。该诊断方法的基本形式是 CRISPR-Cas12 酶与合成的向导 RNA（gRNA）形成复合物。当该复合物高度特异性地与靶 DNA 结合时，该复合物被激活，并且该激活的复合物非特异性地切割标记有荧光猝灭剂的单链 DNA 报告探针。作者最近发现，通过共同聚焦 Cas12-gRNA、报告探针和靶标，电场梯度可用于控制和加速 CRISPR 分析。他们在微流控芯片上使用一种称为等速电泳（ITP）的选择性离子聚焦技术，可以获得合适的电场梯度。与之前的 CRISPR 诊断方法不同，他们还使用 ITP 从原始鼻咽拭子样本中自动纯化靶 RNA。他们将 ITP 纯化与环介导的等温扩增相结合，并通过 ITP 增强的 CRISPR 分析，在 30 分钟内实现对鼻咽拭子样品中 SARS-CoV-2 的 RNA（从原始样本到结果）检测。这种电场控制的方式使基于微流控技术的 CRISPR 的诊断方法成为新的诊断模式。

Abstract:

The rapid spread of COVID-19 across the world has revealed major gaps in our ability to respond to new virulent pathogens. Rapid, accurate, and easily configurable molecular diagnostic tests are imperative to prevent global spread of new diseases. CRISPR-based diagnostic approaches are proving to be useful as field-deployable solutions. In a basic form of this assay, the CRISPR-Cas12 enzyme complexes with a synthetic guide RNA (gRNA). This complex is activated when it highly specifically binds to target DNA, and the activated complex non-specifically cleaves single-stranded DNA reporter probes labeled with a fluorophore-quencher pair. We recently discovered that electric field gradients can be used to control and accelerate this CRISPR assay by co-focusing Cas12-gRNA, reporters, and target. We achieve an appropriate electric field gradient using a selective ionic focusing technique known as isotachophoresis (ITP) implemented on a microfluidic chip. Unlike previous CRISPR diagnostic assays, we also use ITP for automated purification of target RNA from raw nasopharyngeal swab sample. We here combine this ITP purification with loop-mediated isothermal amplification, and the ITP-enhanced CRISPR assay to achieve detection of SARS-CoV-2 RNA (from raw sample to result) in 30 min for both contrived and clinical

nasopharyngeal swab samples. This electric field control enables a new modality for a suite of microfluidic CRISPR-based diagnostic assays.

8. 基于微球的抗 SARS-CoV-2 抗体检测方法的改进

Improved detection of antibody against SARS-CoV-2 by microsphere-based antibody assay

来源：medRxiv
发布时间：2020-05-26
链接：https://www.medrxiv.org/content/10.1101/2020.05.26.20113191v1
第一作者：Carol Ho-Yan Fong
通讯作者：Kelvin Kai-Wang To
通讯作者单位：The University of Hong Kong
DOI 或 PUBMED ID：
编译者：张鹏伟
中文摘要：
目标：目前可用的 COVID-19 抗体检测方法有酶免疫法（EIA）和免疫层析法，其灵敏度和特异性各不相同。文中作者开发并评价了一种新的基于微球的抗体检测方法（MBA），用于检测 SARS-CoV-2 核蛋白（NP）和 spike 蛋白受体结合域（RBD）的免疫球蛋白 G（IgG）。
方法：开发了一种基于微球的检测方法（MBA），以测定抗 SARS-CoV-2 NP 和 spike RBD 的 IgG 水平。使用 2018 年收集的 294 份匿名血清样本，设定血清阳性截止平均荧光强度(MFI)。使用 2020 年前从器官捐献者或流感患者采集的血清样本评估特异性。在 COVID-19 患者中检测血清阳性率。比较 MBA 和 EIA 的血清阳性时间和信号截止比（S/CO）。
结果：MBA 对 NP-IgG 和 RBD-IgG 的特异性分别为 100%（93/93；95%可信区间，96-100%）和 98.9%（92/93；95%CI 94.2-100%）。抗 NP IgG 的 COVID-19 患者恢复期血清标本的 MBA 血清阳性率为 89.8%（35/39），抗 RBD IgG 为 79.5%（31/39）。MBA 组血清阳性时间较 EIA 组短。与 EIA 相比，MBA 能更好地区分 COVID-19 患者和阴性对照组，COVID-19 患者的 S/CO 比值显著高于阴性对照组，而阴性对照组的 S/CO 比值较低。MBA 的模棱两可的样本数（S/CO 0.9-1.1）也少于 EIA。
结论：MBA 方法简便、可靠，适用于临床微生物实验室进行抗 SARS-CoV-2 抗体的准确检测，用于回顾性诊断、血清监测和疫苗试验。
Abstract:
Objective: Currently available COVID-19 antibody tests using enzyme immunoassay (EIA) or immunochromatographic assay have variable sensitivity and specificity. Here, we developed and evaluated a novel microsphere-based antibody assay (MBA) for the detection of immunoglobulin G (IgG) against SARS-CoV-2 nucleoprotein (NP) and spike protein receptor binding domain (RBD).
Method: We developed a microsphere-based assay (MBA) to determine the levels of IgG against SARS-CoV-2 NP and spike RBD. The seropositive cut-off mean fluorescent intensity (MFI) was set using a cohort of 294 anonymous serum specimens collected in 2018. The specificity was assessed using serum specimens collected from organ donors or influenza patients before 2020. Seropositive rate was determined among patients with COVID-19. Time-to-seropositivity and signal-to-cutoff (S/CO) ratio were compared between MBA and EIA.

Results: MBA had a specificity of 100% (93/93; 95% confidence interval [CI], 96-100%) for anti-NP IgG and 98.9% (92/93; 95% CI 94.2-100%) for anti-RBD IgG. The MBA seropositive rate for convalescent serum specimens of COVID-19 patients were 89.8% (35/39) for anti-NP IgG and 79.5% (31/39) for anti-RBD IgG. The time-to-seropositivity was shorter with MBA than that of EIA. When compared with EIA, MBA could better differentiate between COVID-19 patients and negative controls with significantly higher S/CO ratio for COVID-19 patients and lower S/CO ratio with negative controls. MBA also had fewer specimens in the equivocal range (S/CO 0.9-1.1) than EIA.

Conclusion: MBA is robust and simple, and is suitable for clinical microbiology laboratory for the accurate determination of anti-SARS-CoV-2 antibody for retrospective diagnosis, serosurveillance, and vaccine trials.

9. 清华大学研制出了 30 分钟核酸检测的掌上仪器

链接：

https://mp.weixin.qq.com/s?__biz=MzA4OTIyMzgxMw==&mid=2659217316&idx=1&sn=9a10c90cf77b930033a93f858530a112&chksm=8b6ae84ebc1d61580df926c4ddd50adaa8c875ca5515cc084b1c40f040e30b452ce643c70880&mpshare=1&scene=1&srcid=0528yo3LRXP0SvS16mazpChd&sharer_sharetime=1590650202455&sharer_shareid=80f78c62f02832698f0a70d54f98b491&key=19653419b067e158d6ee744e9831a500cbf6d1b9ba77aab0bbc6984ccc90753086fa439a2c347ed2570255d15438a62d40668fb5ef1d7e7583b4cdea895a5eaf9d7bee608ab79f9821b5beea43136014&ascene=1&uin=MjgxMjY4NjgxNQ%3D%3D&devicetype=Windows+10&version=62080085&lang=zh_CN&exportkey=A0IaC03W%2FbkCEtQpaSmAgOI%3D&pass_ticket=v2oRRwKwiWlsS54mD6wC0lnh0Hcu%2FwABBWIdXZ8Wz%2Fb7wIA9Fu2KZaG9EFXi5kRh

根据清华大学微信公众号报道，清华大学研制出了 30 分钟核酸检测的掌上仪器。采用了巢式等温扩增加微流控的技术，最后采用胶体金显示方式实现结果展示。研究团队称该技术灵敏度接近 QPCR 检测方法， 大约为 400copy/ml。该技术在 19 个阳性样品中进行了测试，全部检出。

10. 使用免疫层析检测口水中的 SARS-CoV-2

Immunochromatographic test for the detection of SARS-CoV-2 in saliva

链接：https://www.medrxiv.org/content/10.1101/2020.05.20.20107631v1

5 月 25 日日本的研究团队报告了使用免疫层析的方法检测口水中 SARS-CoV-2 的实验结果。

11. Ginkgo Bioworks 获得 7000 万美金投资，将建立基于二代测序的新冠病毒检测能力

Ginkgo Bioworks Nabs $70M Investment from Illumina, Others to Build Coronavirus NGS Testing

链接：https://www.genomeweb.com/molecular-diagnostics/ginkgo-bioworks-nabs-70m-investment-illumina-others-build-coronavirus-ngs?utm_source=Sailthru&utm_medium=email&utm_campaign=GWDN%20Thurs%20AM%202020-05-28&utm_term=GW%20Daily%20News%20Bulletin#.XtB-JDozaUk

波士顿的合成生物学公司 Ginkgo Bioworks 获得了测序仪器试剂上游厂商 Illumina 以及其它方面的 7000 万美金投资，将建立以二代测序为基础的、价格低廉的测序能力。为能在美国能进行进行大规模人群 SARS-CoV-2 检测，进而为美国的复学，复工做准备。

12. 自 5 月初起 FDA 要求必须经过真实病人样品测试，用于 COVID-19 的分子诊断才能获得紧急授权

FDA Requiring Real Patient Samples for Emergency Use Authorization of COVID-19 MDx Tests

链接：https://www.genomeweb.com/regulatory-news-fda-approvals/fda-requiring-real-patient-samples-emergency-use-authorization-covid#.XtB-1zozaUk

5 月初 FDA 要求必需使用 30 个过往 COVID-19 阳性样品进行检测，获得和标准的 QPCR 接近的检出率的产品才能获得紧急授权用于 COVID-19 检测的分子诊断

13. 恢复期个体对 SARS-CoV-2 感染的趋同抗体反应

Convergent Antibody Responses to SARS-CoV-2 Infection in Convalescent Individuals

来源：bioRxiv
发布时间：2020-05-22
链接：https://www.biorxiv.org/content/10.1101/2020.05.13.092619v2
第一作者：Davide F. Robbiani
通讯作者：Davide F. Robbiani
通讯作者单位：The Rockefeller University, New York, USA
DOI 或 PUBMED ID：Preprint
编译者：张鹏伟
中文摘要：

在 COVID-19 大流行期间，SARS-CoV-2 感染了数百万人，夺去了数十万人的生命。病毒依赖于 SARS-CoV-2 刺突蛋白的受体结合域（RBD）进入细胞。虽然没有疫苗，但抗体很可能是保护的必要条件。然而，人们对 SARS-CoV-2 的抗体反应知之甚少。本文报告 149 例 COVID-19 恢复期患者。症状发作后平均 39 天收集的血浆半数最大中和滴度变化范围从 33％不可检测到 79％以下的 1：1000 以下，而只有 1％的滴度> 1：5000。抗体克隆显示 RBD 特异性记忆 B 细胞的扩增克隆在不同个体中表达密切相关的抗体。尽管血浆滴度较低，但针对 RBD 上三个不同表位的抗体却以低至一位数 ng/mL 的半数最大抑制浓度（IC_{50s}）中和。因此，从 COVID-19 恢复的个体获得的大多数恢复期血浆不含有高水平的中和活性。尽管如此，在所有被检测的个体中都发现了具有强大抗病毒活性的罕见但反复出现的 RBD 特异性抗体，这表明设计用于诱导此类抗体的疫苗可能是广泛有效的。

Abstract:

During the COVID-19 pandemic, SARS-CoV-2 infected millions of people and claimed hundreds of thousands of lives. Virus entry into cells depends on the receptor binding domain (RBD) of the SARS-CoV-2 spike protein (S). Although there is no vaccine, it is likely that antibodies will be essential for protection. However, little is known about the human antibody response to SARS-CoV-2. Here we report on 149 COVID-19 convalescent individuals. Plasmas collected an average of 39 days after the onset of symptoms had variable half-maximal neutralizing titers ranging from undetectable in 33% to below 1:1000 in 79%, while only 1% showed titers >1:5000. Antibody cloning revealed expanded clones of RBD-specific memory B cells expressing closely related antibodies in different individuals. Despite low plasma titers, antibodies to three distinct

epitopes on RBD neutralized at half-maximal inhibitory concentrations (IC_{50s}) as low as single digit ng/mL. Thus, most convalescent plasmas obtained from individuals who recover from COVID-19 do not contain high levels of neutralizing activity. Nevertheless, rare but recurring RBD-specific antibodies with potent antiviral activity were found in all individuals tested, suggesting that a vaccine designed to elicit such antibodies could be broadly effective.

14. 一种系统的炎症研究方法确定 SARS-CoV-2 感染的治疗靶点

A systems approach to inflammation identifies therapeutic targets in SARS-CoV-2 infection

来源：medRxiv
发布时间：2020-05-24
链接：https://www.medrxiv.org/content/10.1101/2020.05.23.20110916v1
第一作者：Frank L. van de Veerdonk, PhD、Nico A.F. Janssen, MD
通讯作者：Mihai G. Netea, MD PhD
通讯作者单位：荷兰拉德布德大学
DOI 或 PUBMED ID：Preprint
编译者：张丽双
中文摘要：

背景：需要对 COVID-19 中炎症过程的全面了解。方法：在这项前瞻性，多中心观察性研究中，纳入经 PCR 验证或临床推定为 COVID-19、进入重症监护病房（ICU）或临床病房的患者。获得了人口统计学和临床数据，并连续收集了血浆。通过 ELISA 测定血浆中 IL-6，TNF-α，补体成分 C3a，C3c 和末端补体复合物（TCC）的浓度。另外，使用靶向蛋白质组学评估了 269 种循环生物标志物。比较了 ICU 和非 ICU 患者的结果。结果：共纳入 119 例患者（38 例 ICU 和 91 例非 ICU）。发现 IL-6 血浆浓度在 COVID-19 中升高，而 TNF-α 浓度相对较低且 ICU 和非 ICU 患者之间没有差异。在采血的第一天，与非 ICU 患者相比，ICU 中的 C3a 和 TCC 浓度明显高于非 ICU 患者。靶向蛋白质组学表明，与非 ICU 患者相比，ICU 中 IL-6、几种趋化因子和肝细胞生长因子明显上调。相反，ICU 与非 ICU 患者相比，干细胞因子显著下调，DPP4 和蛋白 C 抑制剂也明显下调，后两个因子也参与了激肽-激肽释放酶途径的调控。无监督聚类表明大多数感染 SARS-CoV-2 的患者具有同质的致病机制，患者聚类主要基于疾病的严重程度。解读：研究者确定了重症 COVID-19 患者炎症反应异常的重要途径，包括 **IL-6，补体系统和激肽-激肽释放酶途径**。这个发现可能有助于开发针对宿主的新疗法。

Abstract:

Background Infection with SARS-CoV-2 manifests itself as a mild respiratory tract infection in the majority of individuals, which progresses to a severe pneumonia and acute respiratory distress syndrome (ARDS) in 10-15% of patients. Inflammation plays a crucial role in the pathogenesis of ARDS, with immune dysregulation in severe COVID-19 leading to a hyperinflammatory response. A comprehensive understanding of the inflammatory process in COVID-19 is lacking. Methods In this prospective, multicenter observational study, patients with PCR-proven or clinically presumed COVID-19 admitted to the intensive care unit (ICU) or clinical wards were included. Demographic and clinical data were obtained and plasma was serially collected. Concentrations of IL-6, TNF-α, complement

components C3a, C3c and the terminal complement complex (TCC) were determined in plasma by ELISA. Additionally, 269 circulating biomarkers were assessed using targeted proteomics. Results were compared between ICU and non ICU patients. Findings A total of 119 (38 ICU and 91 non ICU) patients were included. IL-6 plasma concentrations were elevated in COVID-19 (ICU vs. non ICU, median 174.5 pg/ml [IQR 94.5-376.3 vs. 40.0 pg/ml [16.5-81.0]), whereas TNF-α concentrations were relatively low and not different between ICU and non ICU patients (median 24.0 pg/ml [IQR 16.5-33.5] and 21.5 pg/ml [IQR 16.0-33.5], respectively). C3a and terminal complement complex (TCC) concentrations were significantly higher in ICU vs. non ICU patients (median 556.0 ng/ml [IQR 333.3-712.5]) vs. 266.5 ng/ml [IQR 191.5-384.0 for C3a and 4506 mAU/ml [IQR 3661-6595 vs. 3582 mAU/ml [IQR 2947-4300] for TCC) on the first day of blood sampling. Targeted proteomics demonstrated that IL-6 (logFC 2.2), several chemokines and hepatocyte growth factor (logFC 1.4) were significantly upregulated in ICU vs. non ICU patients. In contrast, stem cell factor was significantly downregulated (logFC -1.3) in ICU vs. non ICU patients, as were DPP4 (logFC -0.4) and protein C inhibitor (log FC -1.0), the latter two factors also being involved in the regulation of the kinin-kallikrein pathway. Unsupervised clustering pointed towards a homogeneous pathogenetic mechanism in the majority of patients infected with SARS-CoV-2, with patient clustering mainly based on disease severity. Interpretation We identified important pathways involved in dysregulation of inflammation in patients with severe COVID-19, including the IL-6, complement system and kinin-kallikrein pathways. Our findings may aid the development of new approaches to host-directed therapy.

15. 对 COVID-19 患者的深层免疫分析揭示了患者的异质性和独特的免疫类型，及其对治疗干预的意义

Deep immune profiling of COVID-19 patients reveals patient heterogeneity and distinct immunotypes with implications for therapeutic interventions

来源：bioRxiv
发布时间：2020-05-23
链接：https://www.biorxiv.org/content/10.1101/2020.05.20.106401v1
第一作者：Divij Mathew
通讯作者：Michael R. Betts1, Nuala J.Meyer, E. John Wherry
通讯作者单位：美国宾夕法尼亚大学佩雷尔曼医学院,
DOI 或 PUBMED ID：Preprint
编译者：刘焕珍
中文摘要：
能引起免疫失调的 COVID-19 已成为全球流行病，但对 COVID-19 引起的免疫反应仍然知之甚少。作者通过高维度流式细胞仪对 71 例 COVID-19 患者与康复和健康受试者进行比较分析，对～200 种免疫功能和>30 种临床特征的综合分析显示，仅在某些患者中发现 T 细胞和 B 细胞亚群的激活。一小群患者具有急性病毒感染的 T 细胞活化特性，成浆细胞反应可达到循环 B 细胞的 30％以上。但是，另一小群患者的淋巴细胞活化程度与未感染的受试者相当。

通过分析确定了患者稳定的和动态的免疫学特征，并将其与疾病严重性变化的轨迹相关联。这些分析确定了三种“免疫型”与不良的临床轨迹和改善健康状况有关。这些免疫型可能对治疗和疫苗有影响。

Abstract:
COVID-19 has become a global pandemic. Immune dysregulation has been implicated, but immune responses remain poorly understood. We analyzed 71 COVID-19 patients compared to recovered and healthy subjects using high dimensional cytometry. Integrated analysis of ～200 immune and >30 clinical features revealed activation of T cell and B cell subsets, but only in some patients. A subgroup of patients had T cell activation characteristic of acute viral infection and plasmablast responses could reach >30% of circulating B cells. However, another subgroup had lymphocyte activation comparable to uninfected subjects. Stable versus dynamic immunological signatures were identified and linked to trajectories of disease severity change. These analyses identified three “immunotypes” associated with poor clinical trajectories versus improving health. These immunotypes may have implications for therapeutics and vaccines.

16. 单细胞分析显示严重 COVID-19 感染时 T 细胞超活化和瘫痪

T-cell hyperactivation and paralysis in severe COVID-19 infection revealed by single-cell analysis

来源：biorxiv
发布时间：2020.05.26
文章链接：https://www.biorxiv.org/content/10.1101/2020.05.26.115923v1
第一作者：Bahire Kalfaoglu
通讯作者：Masahiro Ono
通讯作者单位：伦敦帝国理工学院生命科学系，熊本大学国际医学研究中心
DOI: https://doi.org/10.1101/2020.05.26.115923
编译者：张怡

中文摘要：
重症 COVID-19 患者可出现呼吸衰竭、T 细胞减少和细胞因子释放综合征(CRS)，这对年轻和老年患者都是致命的，是大流行的一个主要问题。然而，对 COVID-19 中 CRS 的发病机制了解甚少。在这里，作者展示了严重 SARS-CoV-2 感染中 T 细胞失调的单细胞水平的机制，从而论证了严重 COVID-19 患者 T 细胞过度激活和瘫痪的机制。通过对单个细胞 RNA 序列数据库中 CD4+ T 细胞进行虚拟分选，CD4+T 细胞在严重 COVID-19 患者的肺中被高度激活，并显示出独特的分化途径。值得注意的是，在严重 COVID-19 患者中，T 细胞高表达免疫调节受体和 CD25，同时抑制转录因子 FOXP3 的表达，有趣的是，调节性 T 细胞(Treg)和 Th17 的分化均受到抑制。同时，在重症患者中，高度活化的 CD4+ T 细胞与表达 PD-1 配体的巨噬细胞一起表达 PD-1，说明 PD-1 介导的免疫调节部分起作用。此外，作者发现 CD25+超活化 T 细胞分化为多个辅助 T 细胞系，显示具有 Th1 和 Th2 特征的多面效应 T 细胞。最后，作者发现 CD4+ T 细胞，尤其是表达 CD25 的高活化 T 细胞，会产生弗林蛋白酶，促使 SARS-CoV-2 病毒的进入。总体而言，来自重症 COVID-19 患者的 CD4+ T 细胞处于高活化状态，肺中 FOXP3 介导的负反馈机制受损，而活化的 CD4+ T 细胞通过产生弗林蛋白酶继续促进进一步的病毒感染。因此，研究提出了一种新的 T 细胞过度活化和瘫痪模型，导致重症 COVID-19 患者肺

损伤、CRS 和器官衰竭。

Abstract

Severe COVID-19 patients can show respiratory failure, T-cell reduction, and cytokine release syndrome (CRS), which can be fatal in both young and aged patients and is a major concern of the pandemic. However, the pathogenetic mechanisms of CRS in COVID-19 are poorly understood. Here we show single cell-level mechanisms for T-cell dysregulation in severe SARS-CoV-2 infection, and thereby demonstrate the mechanisms underlying T-cell hyperactivation and paralysis in severe COVID-19 patients. By in silico sorting CD4+ T-cells from a single cell RNA-seq dataset, we found that CD4+ T-cells were highly activated and showed unique differentiation pathways in the lung of severe COVID-19 patients. Notably, those T-cells in severe COVID-19 patients highly expressed immunoregulatory receptors and CD25, whilst repressing the expression of the transcription factor FOXP3 and interestingly, both the differentiation of regulatory T-cells (Treg) and Th17 was inhibited. Meanwhile, highly activated CD4+ T-cells express PD-1 alongside macrophages that express PD-1 ligands in severe patients, suggesting that PD-1-mediated immunoregulation was partially operating. Furthermore, we show that CD25+ hyperactivated T-cells differentiate into multiple helper T-cell lineages, showing multifaceted effector T-cells with Th1 and Th2 characteristics. Lastly, we show that CD4+ T-cells, particularly CD25-expressing hyperactivated T-cells, produce the protease Furin, which facilitates the viral entry of SARS CoV-2. Collectively, CD4+ T-cells from severe COVID-19 patients are hyperactivated and FOXP3-mediated negative feedback mechanisms are impaired in the lung, while activated CD4+ T-cells continue to promote further viral infection through the production of Furin. Therefore, our study proposes a new model of T-cell hyperactivation and paralysis that drives pulmonary damage, systemic CRS and organ failure in severe COVID-19 patients.

17. SARS-CoV-2 的病理生理学：靶向内皮细胞会导致血栓性微血管病变和异常免疫反应的复杂疾病,来自西奈山 COVID-19 尸检经验

Pathophysiology of SARS-CoV-2: targeting of endothelial cells renders a complex disease with thrombotic microangiopathy and aberrant immune response. The Mount Sinai COVID-19 autopsy experience

来源： medrxiv

发布时间：2020-05-22

链接：https://www.medrxiv.org/content/10.1101/2020.05.18.20099960v1

第一作者：Clare Bryce, MD; Zachary Grimes, DO; Elisabet Pujadas, MD, PhD

通讯作者：Mary Fowkes, MD, PhD

通讯作者单位：Icahn School of Medicine at Mount Sinai, New York, NY

DOI 或 PUBMED ID：

编译者：王玮

中文摘要：

背景：SARS-CoV-2 及其相关临床综合征 COVID-19 在全球范围内引起了巨大的发病率和死亡

率，纽约市的发病率和死亡率都很高。全面的，综合性的尸检依然缺乏，尸检能够帮助推进疾病机理研究。

方法：在西奈山医院对 67 例 COVID-19 阳性患者进行尸检，并从西奈山数据库中获取临床资料。实验设计包括由一组专业病理学家进行的全面显微镜检查，以及透射电镜、免疫组织化学、RNA 原位杂交以及免疫学和血清学分析。

结果：COVID-19 患者的队列实验室结果显示炎症标志物升高，凝血功能异常，细胞因子 IL-6、IL-8 和 TNFα 升高。尸检发现 4 例出现大肺栓塞。该研究报告了包括大脑在内的多种脏器中存在微血栓，以及在许多患者中明显的噬血细胞综合征和继发性噬血细胞性淋巴组织细胞增生症（hemophagocytosis and a secondary hemophagocytic lymphohistiocytosis-like syndrome）。该研究提供了利用电子显微镜，免疫荧光和免疫组化证据表明了病毒和 ACE2 受体在样品中的存在。

结论：该研究报道了 67 例 COVID-19 阳性患者的综合尸检结果，发现尽管该病目前被认为主要表现为呼吸道病毒性疾病，但也会导致内皮功能障碍、高凝状态以及先天性和适应性免疫反应的失衡。该研究的新发现包括血管内皮细胞 ACE2 在选定器官（肺，脑和心脏）的表型，它与凝血异常和血栓微血管病相关，解释了显著的凝血病和神经精神症状。另一个观察是巨噬细胞激活综合征，伴随着噬血细胞和噬血细胞淋巴组织细胞增生样疾病，是微血管病变和过度释放细胞因子的基础。该研究还讨论了关键调控途径。

Abstract:

BACKGROUND: Severe Acute Respiratory Syndrome Coronavirus-2 (SARS-CoV-2) and its associated clinical syndrome COVID-19 are causing overwhelming morbidity and mortality around the globe, disproportionately affecting New York City. A comprehensive, integrative autopsy series that advances the mechanistic discussion surrounding this disease process is still lacking.

METHODS: Autopsies were performed at the Mount Sinai Hospital on 67 COVID-19 positive patients and data from the clinical records were obtained from the Mount Sinai Data Warehouse. The experimental design included a comprehensive microscopic examination carried out by a team of expert pathologists, along with transmission electron microscopy, immunohistochemistry, RNA in situ hybridization, as well as immunology and serology assays.

RESULTS: Laboratory results of our COVID-19 cohort show elevated inflammatory markers, abnormal coagulation values, and elevated cytokines IL-6, IL-8 and TNFα. Autopsies revealed large pulmonary emboli in four cases. We report microthrombi in multiple organ systems including the brain, as well as conspicuous hemophagocytosis and a secondary hemophagocytic lymphohistiocytosis-like syndrome in many of our patients. We provide electron microscopic, immunofluorescent and immunohistochemical evidence of the presence of the virus and the ACE2 receptor in our samples.

CONCLUSIONS: We report a comprehensive autopsy series of 67 COVID-19 positive patients revealing that this disease, so far conceptualized as a primarily respiratory viral illness, also causes endothelial dysfunction, a hypercoagulable state, and an imbalance of both the innate and adaptive immune responses. Novel findings reported here include an endothelial phenotype of ACE2 in selected organs, which correlates with clotting abnormalities and thrombotic

microangiopathy, addressing the prominent coagulopathy and neuropsychiatric symptoms. Another original observation is that of macrophage activation syndrome, with hemophagocytosis and a hemophagocytic lymphohistiocytosis-like disorder, underlying the microangiopathy and excessive cytokine release. We discuss the involvement of critical regulatory pathways.

18. 细菌分泌的一种脂酶具备广谱地杀黄病毒、SARS-COV-2 以及其他披膜病毒的能力

Broad-spectrum virucidal activity of bacterial secreted lipases against flaviviruses, SARS-CoV-2 and other enveloped viruses

链接：https://www.biorxiv.org/content/10.1101/2020.05.22.109900v2

清华大学医学院程功团队报告了一个细菌分泌的脂酶可以广谱地杀黄病毒、SARS-COV-2 以及其他披膜病毒。该团队的研究主题是虫媒病毒性传染病。最近该团队发表过虫媒肠道为微生物对病毒感染的调控机制的综述。

19. 用于 SARS-CoV-2 研究的人支气管类器官的产生

Generation of human bronchial organoids for SARS-CoV-2 research

来源：bioRxiv
发布时间：2020-05-26
链接：https://www.biorxiv.org/content/10.1101/2020.05.25.115600v1
第一作者：Tatsuya Suzuki
通讯作者：Toru Okamoto, Kazuo Takayama
通讯作者单位：日本大阪大学，日本京都大学
DOI 或 PUBMED ID：https://doi.org/10.1101/2020.05.25.115600
编译者：刘焕珍
中文摘要：

COVID-19 是一种能够引起包括严重肺炎在内的致命疾病。为了开发治疗 COVID-19 的药物，需要建立一种能够复制病毒生命周期和评价抗病毒药物疗效的模型。在这项研究中，作者建立了一种从市售的冷冻人支气管上皮细胞生成人支气管类器官（hBO）的方法，并研究 hBO 是否可作为 SARS-CoV-2 研究的模型。他们的 hBO 包含基底细胞、棒状细胞、纤毛细胞和杯状细胞，并高表达 SARS-CoV-2 的受体血管紧张素转换酶 2（ACE2）和 S 蛋白必须的跨膜丝氨酸蛋白酶 2（TMPRSS2）。SARS-CoV-2 感染 hBO 后，不仅可以观察到细胞内病毒基因组，而且还可以观察到子代病毒、细胞毒性、固缩细胞和 I 型干扰素信号的适度增加。用 TMPRSS2 抑制剂 camostat 治疗，可以将病毒拷贝数减少到对照组的 2%。此外，通过 RNA 序列分析，获得了 SARS-CoV-2 感染 hBO 的基因表达谱。综上所述，作者成功地产生了可用于 SARS-CoV-2 研究和 COVID-19 药物研发的 hBO。

Abstract:

Coronavirus disease 2019 (COVID-19) is a disease that causes fatal disorders including severe pneumonia. To develop a therapeutic drug for COVID-19, a model that can reproduce the viral life cycle and evaluate the drug efficacy of anti-viral drugs is essential. In this study, we established a method to generate human bronchial organoids (hBO) from commercially available cryopreserved human bronchial epithelial cells and examined whether they could be used as a model for severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) research. Our

hBO contain basal, club, ciliated, and goblet cells. Angiotensin-converting enzyme 2 (ACE2), which is a receptor for SARS-CoV-2, and transmembrane serine proteinase 2 (TMPRSS2), which is an essential serine protease for priming spike (S) protein of SARS-CoV-2, were highly expressed. After SARS-CoV-2 infection, not only the intracellular viral genome, but also progeny virus, cytotoxicity, pyknotic cells, and moderate increases of the type I interferon signal could be observed. Treatment with camostat, an inhibitor of TMPRSS2, reduced the viral copy number to 2% of the control group. Furthermore, the gene expression profile in SARS-CoV-2-infected hBO was obtained by performing RNA-seq analysis. In conclusion, we succeeded in generating hBO that can be used for SARS-CoV-2 research and COVID-19 drug discovery.

20. 一种用于检测 SARS-CoV-2 中和抗体的高通量临床检测方法——IMMUNO-COV 的开发和验证

Development and validation of IMMUNO-COV a high-throughput clinical assay for detecting antibodies that neutralize SARS-CoV-2

来源：bioRxiv
发布时间：2020-05-27
链接：https://www.biorxiv.org/content/10.1101/2020.05.26.117549v1
第一作者：Rianna Vandergaast
通讯作者：Rianna Vandergaast、Stephen J. Russell
通讯作者单位：美国明尼苏达州罗切斯特 Imanis 生命科学
DOI 或 PUBMED ID：Preprint
编译者：张丽双
中文摘要：
这里描述 IMMUNO-COV 的开发和验证，IMMUNO-COV 是一种高通量临床检测方法，用于定量测量可以阻断病毒感染的 SARS-CoV-2 中和抗体。该方法构建了一种编码 SARS-CoV-2 spike 糖蛋白的重组水泡性口腔炎病毒（VSV）(VSV-SARS-CoV-2-S-Δ19CT)，测量血清或纯化抗体中和该重组病毒的能力。这种重组病毒感染可诱导 Vero 细胞单层融合，并检测到荧光素酶信号。重组病毒感染可被单克隆抗 SARS-CoV-2-spike 抗体以及来自 SARS-CoV-2 康复者的血浆或血清阻断。该试验在验证试验中表现出 100％的特异性，并且在所有试验中检测假阳性为零。在 230 份血清样品的盲法分析中，根据可用的临床数据仅观察到两个意外结果。观察到检测结果与 80 份样品之间的完美关联，这些样品也同时使用市售 ELISA 进行了检测。为了量化抗病毒反应的强度，研究者通过向合并的 SARS-CoV-2 血清阴性血清中加入梯度浓度的抗 SARS-CoV-2-spike 单克隆抗体来生成校正曲线。使用校正曲线和单一的最佳 1：100 血清测试稀释液，研究者可靠地测量了每个测试样品中的中和抗体水平。从测定中计算出的病毒中和单位（VNU）与针对 SARS-CoV-2 临床分离株的噬斑减少中和测试确定的 PRNT（EC50）值密切相关（$p < 0.0001$）。综上所述，这些结果表明，IMMUNO-COV 测定法可准确定量人血清中的 SARS-CoV-2 中和抗体，因此是当前可用血清学检测的潜在有价值的补充。该测定法可提供重要信息，以比较对目前正在开发的各种 SARS-CoV-2 疫苗的免疫反应，或评估在恢复性血浆疗法研究中的供体资格。
Abstract:
We here describe the development and validation of IMMUNO-COV, a high-throughput

clinical test to quantitatively measure SARS-CoV-2-neutralizing antibodies, the specific subset of anti-SARS-CoV-2 antibodies that block viral infection. The test measures the capacity of serum or purified antibodies to neutralize a recombinant Vesicular Stomatitis Virus (VSV) encoding the SARS-CoV-2 spike glycoprotein. This recombinant virus (VSV-SARS-CoV-2-S-Δ19CT) induces fusion in Vero cell monolayers, which is detected as luciferase signal using a dual split protein (DSP) reporter system. VSV-SARS-CoV-2-S-Δ19CT infection was blocked by monoclonal anti-SARS-CoV-2-spike antibodies and by plasma or serum from SARS-CoV-2 convalescing individuals. The assay exhibited 100% specificity in validation tests, and across all tests zero false positives were detected. In blinded analyses of 230 serum samples, only two unexpected results were observed based on available clinical data. We observed a perfect correlation between results from our assay and 80 samples that were also assayed using a commercially available ELISA. To quantify the magnitude of the anti-viral response, we generated a calibration curve by adding stepped concentrations of anti-SARS-CoV-2-spike monoclonal antibody to pooled SARS-CoV-2 seronegative serum. Using the calibration curve and a single optimal 1:100 serum test dilution, we reliably measured neutralizing antibody levels in each test sample. Virus neutralization units (VNUs) calculated from the assay correlated closely ($p<0.0001$) with PRNT(EC50) values determined by plaque reduction neutralization test against a clinical isolate of SARS-CoV-2. Taken together, these results demonstrate that the IMMUNO-COV assay accurately quantitates SARS-CoV-2 neutralizing antibodies in human sera and therefore is a potentially valuable addition to the currently available serological tests. The assay can provide vital information for comparing immune responses to the various SARS-CoV-2 vaccines that are currently in development, or for evaluating donor eligibility in convalescent plasma therapy studies.

21. 通过分析 SARS-CoV-2 感染的细胞的形态学来确定治疗 COVID-19 的药物再利用候选药物

Morphological Cell Profiling of SARS-CoV-2 Infection Identifies Drug Repurposing Candidates for COVID-19

来源：bioRxiv
发布时间：2020-05-27
链接：https://www.biorxiv.org/content/10.1101/2020.05.27.117184v1
第一作者：Carmen Mirabelli
通讯作者：Jonathan Z. Sexton
通讯作者单位：美国密歇根大学
DOI 或 PUBMED ID：https://doi.org/10.1101/2020.05.27.117184
编译者：刘焕珍
中文摘要：
SARS-CoV-2 及其相关疾病 COVID-19 的全球传播，需要能够迅速转化为临床护理的治疗干预措施。不幸的是，传统的药物研发的失败率超过 90%，从靶点识别到临床应用可能需要 10-

15 年的时间。相反，药物再利用可以显著加快治疗 COVID-19 药物的转化。作者开发了一种高通量的定量筛选方法，以确定抗 SARS-CoV-2 的有效单药和联合疗法。将定量的高内涵形态学分析与基于人工智能的机器学习策略相结合，对感染和应激的细胞特征进行分类。本实验检测了多种抗病毒作用机制，包括抑制病毒进入、繁殖和调节宿主细胞应答。从 1441 种 FDA 批准的化合物和临床候选化合物库中,确定了 15 个具有抗病毒作用的剂量反应化合物。特别是，他们发现乳铁蛋白是 SARS-CoV-2 感染的有效抑制剂，其 IC_{50} 为 308nM，并且在联合用药疗法中增强了瑞德西韦和羟氯喹的疗效。乳铁蛋白还刺激抗病毒宿主细胞应答，并在 iPSC 衍生的肺泡上皮细胞中保留抑制活性。鉴于乳铁蛋白在人体中的安全性，这些数据表明乳铁蛋白是 COVID-19 的易于转化的治疗辅助剂。此外，还发现几种常用处方药会加重病毒的感染，值得临床调查研究。作者的结论是，对于 COVID-19 流行病和其他新兴传染病，药物再利用的形态学分析是选择和优化药物和药物组合的有效策略。

Abstract:

The global spread of the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), and the associated disease COVID-19, requires therapeutic interventions that can be rapidly translated to clinical care. Unfortunately, traditional drug discovery methods have a >90% failure rate and can take 10-15 years from target identification to clinical use. In contrast, drug repurposing can significantly accelerate translation. We developed a quantitative high-throughput screen to identify efficacious single agents and combination therapies against SARS-CoV-2. Quantitative high-content morphological profiling was coupled with an AI-based machine learning strategy to classify features of cells for infection and stress. This assay detected multiple antiviral mechanisms of action (MOA), including inhibition of viral entry, propagation, and modulation of host cellular responses. From a library of 1,441 FDA-approved compounds and clinical candidates, we identified 15 dose-responsive compounds with antiviral effects. In particular, we discovered that lactoferrin is an effective inhibitor of SARS-CoV-2 infection with an IC_{50} of 308 nM and that it potentiates the efficacy of both remdesivir and hydroxychloroquine. Lactoferrin also stimulates an antiviral host cell response and retains inhibitory activity in iPSC-derived alveolar epithelial cells. Given its safety profile in humans, these data suggest that lactoferrin is a readily translatable therapeutic adjunct for COVID-19. Additionally, several commonly prescribed drugs were found to exacerbate viral infection and warrant clinical investigation. We conclude that morphological profiling for drug repurposing is an effective strategy for the selection and optimization of drugs and drug combinations as viable therapeutic options for COVID-19 pandemic and other emerging infectious diseases.

22. 冠状病毒疫苗研制成功的三个关键参数

Certainty of success: three critical parameters in coronavirus vaccine development

来源：npj Vaccines

发布时间：2020.05.25

文章链接：https://www.nature.com/articles/s41541-020-0193-6

作者：David C. Kaslow

作者单位：美国帕斯适宜卫生科技组织
DOI: https://doi.org/10.1038/s41541-020-0193-6
编译者：张怡
中文摘要：
针对 17 种病毒病原体的疫苗已获准用于人类。此前，提出了病原体和宿主-病原体相互作用的两个关键生物学参数，潜伏期和广泛的保护性的相对免疫原性，这被认为是过去疫苗开发成功的主要原因，并且针对目前尚无疫苗的病毒性病原体，有助于评估其有效疫苗开发的“成功确定性”。在考虑到人类冠状病毒疫苗，特别是 SARS-CoV-2 在研发中的“成功的确定性”时，提出了第三个相关的关键参数——个体层面的感染接种强度和人群层面的感染力。预计减少传染性接种量的强度(以及在群体水平上的感染力)将延长潜伏期，进而预计将减少疾病的严重程度，并增加接触流动的病毒后记忆反应的机率。同样，在试验和部署 COVID-19 疫苗的同时，成功地实施个体行为和群体的行为，分别降低感染的接种强度和力度，预计将增加展示疫苗“成功确定性”，控制 SARS-CoV-2 感染、疾病、死亡和大流行。

Abstract:
Vaccines for 17 viral pathogens have been licensed for use in humans. Previously, two critical biological parameters of the pathogen and the host-pathogen interaction—incubation period and broadly protective, relative immunogenicity—were proposed to account for much of the past successes in vaccine development, and to be useful in estimating the “certainty of success” of developing an effective vaccine for viral pathogens for which a vaccine currently does not exist. In considering the “certainty of success” in development of human coronavirus vaccines, particularly SARS-CoV-2, a third, related critical parameter is proposed—infectious inoculum intensity, at an individual-level, and force of infection, at a population-level. Reducing the infectious inoculum intensity (and force of infection, at a population-level) is predicted to lengthen the incubation period, which in turn is predicted to reduce the severity of illness, and increase the opportunity for an anamnestic response upon exposure to the circulating virus. Similarly, successfully implementing individual- and population-based behaviors that reduce the infectious inoculum intensity and force of infection, respectively, while testing and deploying COVID-19 vaccines is predicted to increase the “certainty of success” of demonstrating vaccine efficacy and controlling SARS-CoV-2 infection, disease, death, and the pandemic itself.

23. 芦可替尼治疗重症冠状病毒疾病 2019(COVID-19)：一项多中心、单盲、随机对照试验

Ruxolitinib in treatment of severe coronavirus disease 2019 (COVID-19): A multicenter, single-blind, randomized controlled trial

来源：Journal of Allergy and Clinical Immunology
发布时间：2020-05-26
链接：https://www.sciencedirect.com/science/article/pii/S0091674920307387
第一作者：Yang Cao，Jia Wei，Liang Zou，Tiebin Jiang
通讯作者：黄刚，王伟，周剑峰

通讯作者单位：美国辛辛那提儿童医院医学中心，华中科技大学同济医学院
DOI 或 PUBMED ID：https://doi.org/10.1016/j.jaci.2020.05.019
编译者：雷颖
中文摘要：
国际医学杂志《过敏及临床免疫学杂志》发表了一份在中国进行的临床研究结果：在使用一个叫做芦可替尼（Ruxolitinib）的药物进行治疗后，重症新冠肺炎患者出现“零死亡”，而使用正常标准治疗的对照组，死亡率为 14.3%。该研究的缺陷是入组的患者太少：治疗组 20 人，对照组 21 人。基于目前的数据，诺华已经开始提供芦可替尼，作为“同情用药”使用。

Abstract
Background: Accumulating evidence proposed JAK inhibitors as therapeutic targets warranting rapid investigation.
Objective: This study evaluated the efficacy and safety of ruxolitinib, a Janus-associated kinase (JAK1/2) inhibitor, for COVID-19.
Methods: We conducted a prospective, multicenter, single-blind, randomized controlled phase II trial involving patients with severe COVID-19.
Results: Forty-three patients were randomly assigned (1:1) to receive ruxolitinib plus SoC treatment (22 patients) or placebo based on SoC treatment (21 patients). After exclusion of 2 patients (1 ineligible, 1 consent withdrawn) from the ruxolitinib group, 20 patients in intervention group and 21 patients in control group were included in the study. Treatment with ruxolitinib plus SoC was not associated with significantly accelerated clinical improvement in severe patients with COVID-19, although ruxolitinib recipients had a numerically faster clinical improvement. Eighteen (90%) patients from the ruxolitinib group showed CT improvement at D14 compared with 13 (61.9%) patients from the control group (P = 0.0495). Three patients in the control group died of respiratory failure, with 14.3% overall mortality at D28; no patients died in the ruxolitinib group. Ruxolitinib was well tolerated with low toxicities and no new safety signals. Levels of 7 cytokines were significantly decreased in the ruxolitinib group in comparison to the control group.
Conclusions: Although no statistical difference was observed, ruxolitinib recipients had a numerically faster clinical improvement. Significant chest CT improvement, a faster recovery from lymphopenia and favorable side-effect profile in ruxolitinib group were encouraging and informative to future trials to test efficacy of ruxolitinib in a larger population.
This trial is registered at www.chictr.org.cn as ChiCTR-OPN-2000029580.

24. 瑞德西韦治疗 Covid-19 重症患者 5 天或 10 天的疗效比较

Remdesivir for 5 or 10 Days in Patients with Severe Covid-19

来源：The New England Journal of Medicine
发布时间：2020-05-27
链接：https://www.nejm.org/doi/full/10.1056/NEJMoa2015301?query=featured_home
第一作者：Jason D. Goldman

通讯作者：Diana M. Brainard
通讯作者单位：吉利德科学公司
DOI 或 PUBMED ID：10.1056/NEJMoa2015301
编译者：宋张悦
中文摘要：

背景：瑞德西韦是一种 RNA 聚合酶抑制剂，在体外具有强大的抗病毒活性，在 Covid-19 动物模型中疗效较好。

方法：研究团队进行了一项随机、开放标签的 3 期临床试验，研究对象为已确诊的 SARS-CoV-2 感染的住院患者，这些患者呼吸周围空气时血氧饱和度小于等于 94%，且伴有影像学确诊的肺炎。本试验中，患者招募自 2020 年 3 月 6 日至 3 月 26 日，美国、意大利、西班牙、德国、香港、新加坡、韩国和台湾的 55 家医院。按 1:1 的比例随机分配患者接受 5 天或 10 天的静脉注射瑞德西韦。所有患者第 1 天给药 200mg 瑞德西韦，随后每天给药 100mg。主要终点为第 14 天的临床状况，采用 7 分制序量表进行评估。

结果：本研究总共 397 名患者接受了随机分组并开始治疗（200 名患者治疗 5 天，197 名患者治疗 10 天）。5 天组的治疗时间中位数为 5 天（四分位数范围，5-5），10 天组为 9 天（四分位数范围，5-10）。在基线时，随机分配到 10 天组的患者的临床状况明显比分配到 5 天组的患者差（P=0.02）。到第 14 天，5 天组 64%的患者和 10 天组 54%的患者在 7 分制序量表上出现 2 分或更多的临床改善。校正基线临床状态后，10 天组患者在 14 天的临床状态分布与 5 天组患者的分布相似(P=0.14)。最常见的不良反应是恶心(9%的患者)、呼吸衰竭恶化(8%)、丙氨酸转氨酶水平升高（7%）和便秘（7%）。

结论：在不需要机械通气的重症 Covid-19 患者中，本研究的试验没有显示瑞德西韦 5 天疗程和 10 天疗程之间疗效的显著差异。然而，在没有安慰剂对照的情况下，疗效的大小无法确定。（由吉利德科学公司资助，GS-US-540-5773，临床试验注册号：NCT04292899，链接：https://www.clinicaltrials.gov/ct2/show/NCT04292899。）

编者注：该临床试验的结果早些时间被 NIAID 的所长福奇博士口头通报过。

Abstract:

BACKGROUND: Remdesivir is an RNA polymerase inhibitor with potent antiviral activity in vitro and efficacy in animal models of coronavirus disease 2019 (Covid-19).

METHODS: We conducted a randomized, open-label, phase 3 trial involving hospitalized patients with confirmed SARS-CoV-2 infection, oxygen saturation of 94% or less while they were breathing ambient air, and radiologic evidence of pneumonia. Patients were randomly assigned in a 1:1 ratio to receive intravenous remdesivir for either 5 days or 10 days. All patients received 200 mg of remdesivir on day 1 and 100 mg once daily on subsequent days. The primary end point was clinical status on day 14, assessed on a 7-point ordinal scale.

RESULTS: In total, 397 patients underwent randomization and began treatment (200 patients for 5 days and 197 for 10 days). The median duration of treatment was 5 days (interquartile range, 5 to 5) in the 5-day group and 9 days (interquartile range, 5 to 10) in the 10-day group. At baseline, patients randomly assigned to the 10-day group had significantly worse clinical status than those assigned to the 5-day group (P=0.02). By day 14, a clinical improvement of 2 points or more on the ordinal scale occurred in 64% of patients in the 5-day group and in 54%

in the 10-day group. After adjustment for baseline clinical status, patients in the 10-day group had a distribution in clinical status at day 14 that was similar to that among patients in the 5-day group (P=0.14). The most common adverse events were nausea (9% of patients), worsening respiratory failure (8%), elevated alanine aminotransferase level (7%), and constipation (7%).

CONCLUSIONS: In patients with severe Covid-19 not requiring mechanical ventilation, our trial did not show a significant difference between a 5-day course and a 10-day course of remdesivir. With no placebo control, however, the magnitude of benefit cannot be determined. (Funded by Gilead Sciences; GS-US-540-5773 ClinicalTrials.gov number, NCT04292899.)

25. 一项目多国临床试验中氯喹或者羟氯喹对于治疗 COVID-19 无效

Hydroxychloroquine or chloroquine with or without a macrolide for treatment of COVID-19: a multinational registry analysis 22 May 2020

链接：

https://www.sciencedirect.com/science/article/pii/S0140673620311806?via%3Dihub

5 月 22 日，美国和瑞士的研究团队报告了在 6 大洲 671 个医院进行的氯喹治疗 COVID-19 的临床试验结果。该研究包括了 96032 病人。研究者们没有发现氯喹或者羟氯喹的疗效。

26. 纵向实验室检测结果表明 COVID-19 病人有非常独特的凝血症信号

Longitudinal laboratory testing tied to PCR diagnostics in COVID-19 patients reveals temporal evolution of distinctive coagulopathy signatures

链接：https://www.medrxiv.org/content/10.1101/2020.05.21.20109439v1

5 月 23 日，来自于强生和 nference 的研究者们对电子病历里对 COVID-19 病人和非 COVID-19 病人的纵向实验检测结果进行比较分析，发现随着感染的进展，COVID-19 病人表现初非常不一样的凝血症信号。

27. 在 SARS 恢复病人和未感染人群中预存针对 SARS-CoV-2 的特异性 T 细胞免疫有差异

Different pattern of pre-existing SARS-COV-2 specific T cell immunity in SARS-recovered and uninfected individuals

链接：https://www.biorxiv.org/content/10.1101/2020.05.26.115832v1

杜克-新加坡国立大学的团队研究了具有不同 SARS-CoV-2 和 SARS-Cov 感染历史的恢复病人的 T 细胞。其中包括 24 个从 SARS-CoV-2 感染中恢复的病人，23 个从 SARS-CoV 感染中恢复的病人，以及 18 个两种病毒都没有感染/接触的人。他们发现冠状病毒感染后，T 细胞对 SARS-CoV-2 的核心蛋白有持久的免疫力，而在既没有接触/感染过 SARS-CoV 或者 SARS-CoV-2 里的人的样本中发现了针对 SARS-CoV-2 NSP7 和 NSP13 的 T 细胞。作者指出研究人群中预存的针对 ORF-1 的 NSP7 和 NSP13 特异性的 T 细胞将会怎么影响 COVID-19 的易感性以及病理过程有非常重要的意义。

28. COVID-19 病人血清中的蛋白质组和代谢组学特征

Proteomic and Metabolomic Characterization of COVID-19 Patient Sera

链接：https://www.cell.com/cell/fulltext/S0092-8674(20)30627-9

西湖大学的团队在 CELL 上发表了 COVID-19 病人血清中的蛋白质组和代谢组学特征，4 月 8 日的简报第 8 条报道了该文章的预印版本。

29. 肠道炎症调节 ACE2 和 TMPRSS2 的表达，并可能与 SARS-CoV-2 相关疾病发病机制的重叠

Intestinal inflammation modulates the expression of ACE2 and TMPRSS2 and potentially overlaps with the pathogenesis of SARS-CoV-2 related disease

来源：biorxiv
发布时间：2020-05-23
链接：https://www.biorxiv.org/content/10.1101/2020.05.21.109124v1
第一作者：Mayte Suárez-Fariñas
通讯作者：Saurabh Mehandru
通讯作者单位：The Dr. Henry D. Janowitz Division of Gastroenterology, Department of Medicine, Icahn School of Medicine at Mount Sinai, New York, NY, USA
DOI 或 PUBMED ID：Preprint
编译者：孔娟
中文摘要：
免疫调节障碍和细胞因子释放综合征已成为 COVID-19 的病理特征，细胞因子拮抗剂已被作为治疗剂进行评估。许多针对 COVID-19 患者的免疫导向疗法已经在临床上用于慢性炎症，如炎症性肠病(IBD)。基于这些发现，作者系统地评估了 COVID-19 和健康及肠道炎症之间可能交叉特征。研究中发现 IBD 药物，无论是生物的还是非生物来源，都不会显著影响非炎症性肠中 ACE2 和 TMPRSS2 的表达。此外，通过比较 SARS-CoV2 诱导的上皮基因特征和 IBD 相关基因，他们确定了 COVID-19 和 IBD 之间共有的分子亚网络。这些发现对 COVID-19 和 IBD 相关炎症的新认识，并为进一步研究治疗 COVID-19 的特定 IBD 药物的机制提供了新的理论支持。
Abstract：
Immune dysregulation and cytokine release syndrome have emerged as pathological hallmarks of severe Coronavirus Disease 2019 (COVID-19), leading to the evaluation of cytokine antagonists as therapeutic agents. A number of immune-directed therapies being considered for COVID-19 patients are already in clinical use in chronic inflammatory conditions like inflammatory bowel disease (IBD). These considerations led us to systematically examine the intersections between COVID-19 and the GI tract during health and intestinal inflammation. We have observed that IBD medications, both biologic and nonbiologic, do not significantly impact ACE2 and TMPRSS2 expression in the uninflamed intestines. Additionally, by comparing SARS CoV2-induced epithelial gene signatures with IBD-associated genes, we have identified a shared molecular subnetwork between COVID-19 and IBD. These data generate a novel appreciation of the confluence of COVID-19- and IBD-associated inflammation and provide mechanistic insights supporting further investigation of specific IBD drugs in the treatment of COVID-19.

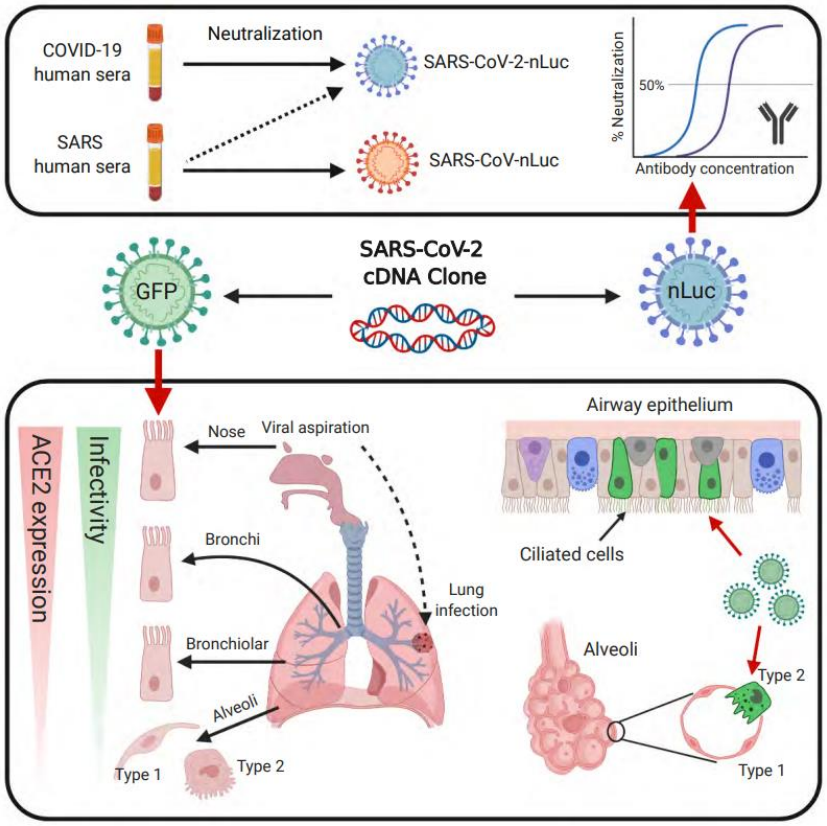
30. SARS-CoV-2 的反向遗传学揭示呼吸道里存在梯度感染

SARS-CoV-2 Reverse Genetics Reveals a Variable Infection Gradient in the Respiratoty Tract

来源：Cell
发布时间：2020-05-27
链接：https://www.cell.com/cell/fulltext/S0092-8674(20)30675-9
第一作者：Yixuan J Hou
通讯作者：Ralph S.Baric
通讯作者单位：University of North Carolina at Chapel Hill
DOI 或 PUBMED ID：
编译者：蒋立春
中文摘要：

COVID-19 病人怎么感染病毒，为什么病人会表现出不同的临床症状，目前了解的还很少。研究者们采用反向遗传学的方法将 SARS-CoV-2 的 ORF7 的一个片段进行替换，构建出包含 GFP 荧光蛋白报告系统以及荧光素酶报告系统的病毒（编者注 1）。然后用带 GFP 报告系统的病毒研究了 SARS-CoV-2 的病理。用带荧光素酶报告系统的病毒研究 SARS 和 COVID-19 病人的血清学反应发现这两种病毒感染的病人存在有限的中和反应。

高灵敏度的 RNA 原位 mapping 揭示 ACE2 在鼻子有最高的表达，在下呼吸道逐渐降低。这和 SARS-CoV-2 的在肺部上皮培养中近端和远端的显著梯度一致。COVID-19 尸检的研究在肺部发现集中的感染，和培养的数据一致，SARS-CoV-2 感染的有纤毛的细胞以及在气道和肺泡区域的 2 型肺细胞。这些发现强调了鼻子对 SARS-CoV-2 的易感性，以及后续呼吸介导的病毒向肺部的扩散。作者们通过反向遗传学构建的病毒为研究保护性免疫、宿主易感性以及病毒的致病机理中的病毒-宿主相互作用提供了一个基础。



编者注 1:
因为 SARS-CoV-2 的病毒基因组在病毒基因组里算很大的，达到了约 3 万个碱基。其反向遗传学操作难度高。

Summary:

The mode of acquisition and causes for the variable clinical spectrum of COVID-19 remain unknown. We utilized a reverse genetics system to generate a GFP reporter virus to explore SARS-CoV-2 pathogenesis and a luciferase reporter virus to demonstrate sera collected from SARS and COVID-19 patients exhibited limited cross-CoV neutralization. High-sensitivity RNA *in situ* mapping revealed the highest ACE2 expression in the nose with decreasing expression throughout the lower respiratory tract, paralleled by a striking gradient of SARS-CoV-2 infection in proximal (high) vs distal (low) pulmonary epithelial cultures. COVID-19 autopsied lung studies identified focal disease and, congruent with culture data, SARS-CoV-2-infected ciliated and type 2 pneumocyte cells in airway and alveolar regions, respectively. These findings highlight the nasal susceptibility to SARS-CoV-2 with likely subsequent aspiration-mediated virus seeding to the lung in SARS-CoV-2 pathogenesis. These reagents provide a foundation for investigations into virus-host interactions in protective immunity, host susceptibility, and virus pathogenesis.

31. ACE2 诱导 SARS-CoV-2 Spike 蛋白构象变化的结构基础

Structural basis of SARS-CoV-2 spike protein induced by ACE2

来源：bioRxiv

发布时间：2020-05-24

链接：https://www.biorxiv.org/content/10.1101/2020.05.24.113175v1

第一作者：Tomer Meirson

通讯作者：Tomer Meirson1, 2, Gal Markel2, 3

通讯作者单位：

1 Azrieli Faculty of Medicine, Bar-Ilan University, Israel

2 Ella Lemelbaum Institute for Immuno-oncology, Sheba Medical Center, Ramat-Gan 526260, Israel

3 Department of Clinical Microbiology and Immunology, Sackler Faculty of Medicine, Tel Aviv University, Tel-Aviv, Israel

DOI 或 PUBMED ID：

编译者：宋珂

中文摘要：

动机：SARS-CoV-2 在世界范围的传播造成了全球性的卫生紧急状况。病毒利用 Spike 糖蛋白（S）与受体（血管紧张素转换酶 2）ACE2 结合，并促使 SARS-CoV-2 病毒侵入宿主细胞。S 蛋白三聚体通过末端的受体结合结构域（RBD）与受体结合，结合后的 S 蛋白会发生构象变化，从而可以被宿主细胞的蛋白酶激活。理解 SARS-CoV-2 病毒侵入细胞的动态结构特征，可能会对研究病毒的传播机制提供启发，并发现新的治疗靶标。作者收集整理了使用 X 射线衍射和 cryo-EM 解析的 S 蛋白的结构，并对 SARS-CoV-2-RBD 的不同构象进行了结构分析和原子精度的比较。

结果：本文中，作者确定了 S 蛋白中被受体诱导的关键结构区域，并表征了其分子内的相互作用。作者发现，κ-螺旋（也称为聚脯氨酸 II）是与 ACE2 结合的界面上的主要结构，并在 S 蛋白转变为活性构象时起到促进作用。作者还在类似开关功能的 κ-螺旋和 β-折叠中发现了一系列的构象转变，以及影响近端连接区域的一组短 α-螺旋的构象变化。这种构象

变化导致位于连接区域的保守的二硫键组成发生了交替变化，表明可能存在二硫键交换。而这正是与 HIV 和鼠冠状病毒等多种病毒侵入宿主过程相关的重要变构开关。本文展示的结构信息，使我们能够发现和了解 SARS-CoV-2-RBD 的重要动态特征，并提出了一种新颖的潜在治疗策略来阻断病毒的进入。总体而言，本研究为针对 SARS-CoV-2 病毒而进行的基于结构的干预策略的设计和优化提供了指导。

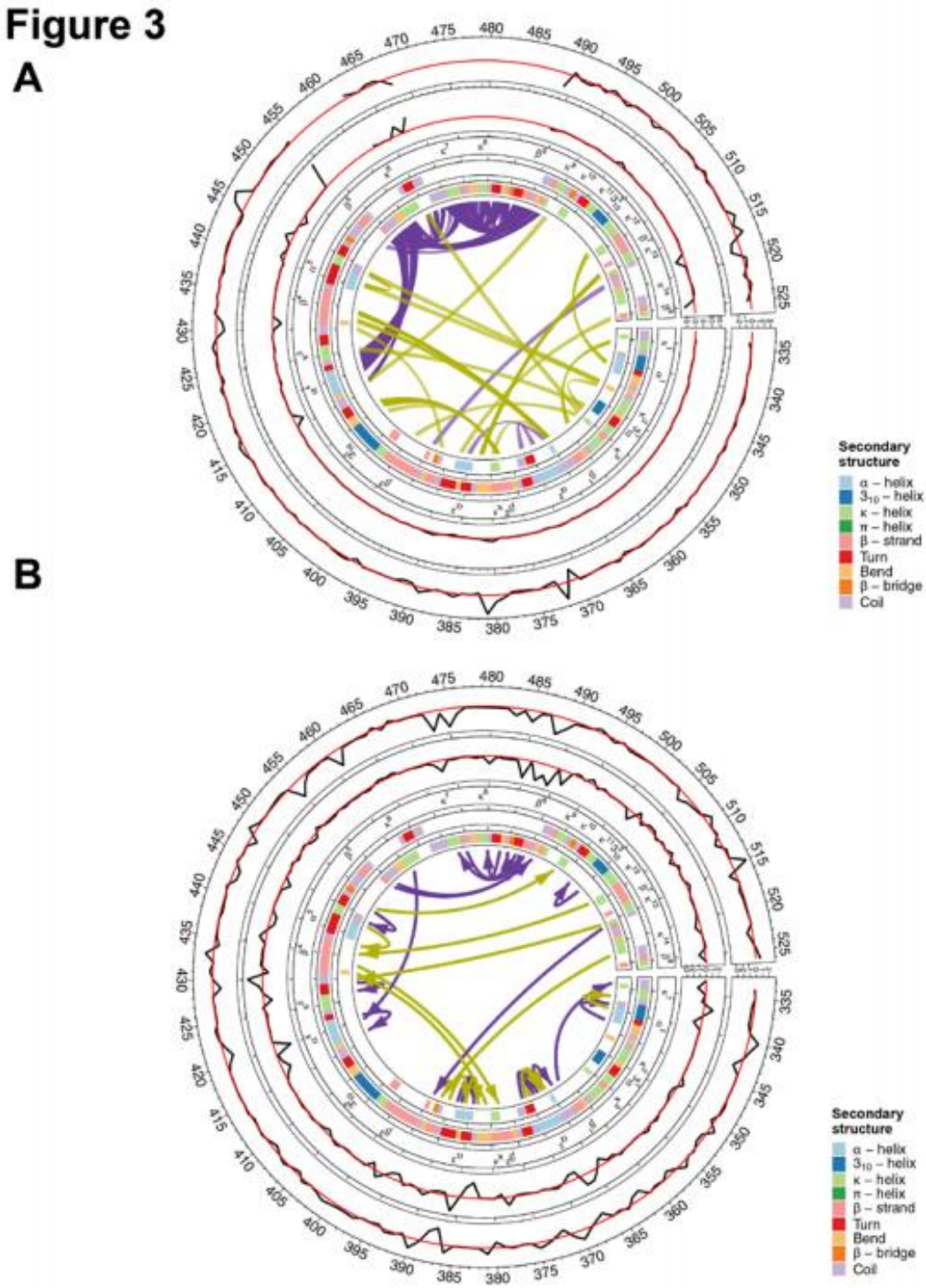


Figure 3. Intramolecular interactions induced by SARS-CoV-2-RBD and ACE2 complex. (A) Circos plot depicting the main differences in intramolecular interactions (< 3.5Å) between the unbound (6VXX:B and 6VYB:B) and bound (6M0J:E) conformations of the SARS-CoV-2-RBD.

Track order: shift in the angular step (ϑ) per residue (i); shift in the rise (d) per residue (ii); labels of secondary structural elements (iii); secondary structure assignment of the unbound-open conformation (iv); transformed secondary structure of the bound conformation, representing the induced conformational changes (v). Gained and lost interactions are shown as magenta and yellow lines, respectively. (B) Circos plot depicting the main differences in main-chain H-bond profile between the unbound and bound conformations of the SARSCoV-2-RBD. Track order: mean difference in the electrostatic interaction energy of the donor (i) and acceptor (ii); labels of secondary structural elements (iii); secondary structure assignment of the unboundopen conformation (iv); transformed secondary structure of the bound conformation, representing the induced conformational changes (v). Gained and lost H-bonds (|ΔE| > 1kcal/mol) illustrating the direction of donor-acceptor are shown as magenta and yellow arrows, respectively

Abstract:

Motivation: The recent emergence of the novel SARS-coronavirus 2 (SARS-CoV-2) and its international spread pose a global health emergency. The viral spike (S) glycoprotein binds the receptor (angiotensin-converting enzyme 2) ACE2 and promotes SARS-CoV-2 entry into host cells. The trimeric S protein binds the receptor using the distal receptor-binding domain (RBD) causing conformational changes in S protein that allow priming by host cell proteases. Unravelling the dynamic structural features used by SARS-CoV-2 for entry might provide insights into viral transmission and reveal novel therapeutic targets. Using structures determined by X-ray crystallography and cryo-EM, we performed structural analysis and atomic comparisons of the different conformational states adopted by the SARS-CoV-2-RBD.

Results: Here, we determined the key structural components induced by the receptor and characterized their intramolecular interactions. We show that κ-helix (also known as polyproline II) is a predominant structure in the binding interface and in facilitating the conversion to the active form of the S protein. We demonstrate a series of conversions between switch-like κ-helix and β-strand, and conformational variations in a set of short α-helices which affect the proximal hinge region. This conformational changes lead to an alternating pattern in conserved disulfide bond configurations positioned at the hinge, indicating a possible disulfide exchange, an important allosteric switch implicated in viral entry of various viruses, including HIV and murine coronavirus. The structural information presented herein enables us to inspect and understand the important dynamic features of SARS-CoV-2-RBD and propose a novel potential therapeutic strategy to block viral entry. Overall, this study provides guidance for the design and optimization of structure-based intervention strategies that target SARS-CoV-2.

32. SARS-CoV-2 Spike 三聚体与 ACE2 二聚体相互作用，并存在有限的 Spike 内亲和力

Trimeric SARS-CoV-2 Spike interacts with dimeric ACE2 with limited intra-Spike avidity

来源：bioRxiv
发布时间：2020-05-21
链接：https://www.biorxiv.org/content/10.1101/2020.05.21.109157v1
第一作者：Irene Lui, Xin X. Zhou, Shion A. Lim, Susanna K. Elledge (Contributed equally)
通讯作者：James A. Wells
通讯作者单位：Department of Pharmaceutical Chemistry, University of California San Francisco, San Francisco, California, 94158, USA
DOI 或 PUBMED ID：
编译者：宋珂

中文摘要：

由 SARS-CoV-2 引起的疫情造成了严重的公共卫生危机。SARS-CoV-2 病毒借助 Spike 蛋白三聚体的受体结合结构域（Spike-RBD）与人类血管紧张素转化酶 2（ACE2）二聚体受体之间的相互作用侵入宿主。显然，阻断 Spike 与 ACE2 间的相互作用，是较有希望的抵抗 SARS-CoV-2 的治疗策略。然而，以目前我们对 Spike/ACE2 相互作用的了解，还不足以理性地设计出效果最好的药物分子。本文中，作者利用重组的 ACE2 和 Spike-RBD 结构域的不同多聚体形式，通过表征 Spike/ACE2 间的亲和力和动力学特性，研究了 Spike/ACE2 间相互作用的机理。同时，还将 ACE2 工程改造设计出融合纳米荧光素酶的报告系统，用来探测 Spike 三聚体中 Spike-RBD 结构域的构象分布情况。有趣的是，相比于 ACE2 单体，二聚体形式的 ACE2 与 Spike 的亲和力更高，并可以在病毒中和试验中阻止假病毒和 SARS-CoV-2 活病毒侵入宿主。作者发现，ACE2 二聚体与 Spike 三聚体中的一个 RBD 相互作用时，存在有限的 Spike 内亲合力，但其仍对增加相互作用的亲和力有贡献。此外，作者还发现部分 Spike 可以同时与多个 ACE2 二聚体相互作用，表明 Spike 三聚体中可以允许多个 RBD 结构域同时处于 ACE2 可接触的“up”构象。本文的发现对以阻断 Spike/ACE2 相互作用为目标的药物分子的设计策略具有重要影响。作者描述的方法可以作为分子工具免费提供给研究团体，以进一步加深我们对 SARS-CoV-2 的生物学了解。

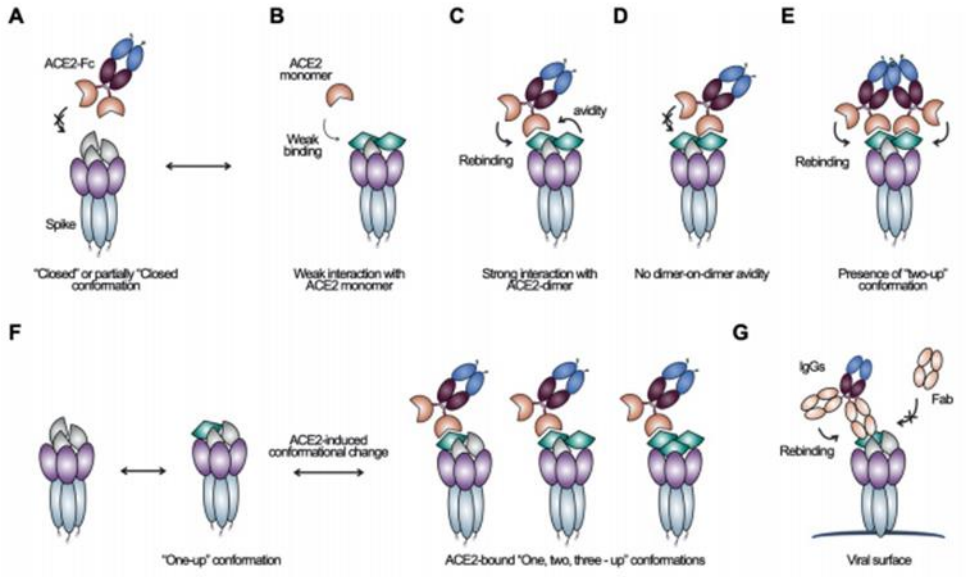


Figure 6. Model of the ACE2/Spike interaction and therapeutic strategies. (A) The majority of FL-Spike proteins are in the "closed" or partially "closed" conformation and "opening up" of RBD on Spike is necessary for ACE2 binding. **(B)** Monomeric ACE2 binds poorly to trimeric Spike, **(C)** ACE2-Fc binds much stronger to Spike than monomer, indicating that rebinding and intra-Spike avidity contribute significantly to the high-affinity nature of this interaction. **(D)** Dimeric ACE2-Fc does not interact with FL-Spike with a full two-on-two intramolecular avidity. **(E)** More than one RBD can be in the "up" conformation, enabling the engagement of separate ACE2 molecules. For **(B-E)**, only the "two-up" conformation is shown but other RBD conformations are also possible. **(F)** A proposed model where ACE2 binding may induce a conformational change in Spike, resulting in "two-up" or "three-up" RBD conformations. **(H)** Dimeric molecules such as ACE2-Fc or IgG will be more potent than a monomeric inhibitor for neutralizing SARS-CoV-2 virus. For all panels, RBD in the "open" conformation is colored in green and RBD in the "closed" formation colored in grey.

Abstract:
A serious public health crisis is currently unfolding due to the SARS-CoV-2 pandemic. SARS-CoV-2 viral entry depends on an interaction between the receptor binding domain of the trimeric viral Spike protein (Spike-RBD) and the dimeric human angiotensin converting enzyme 2 (ACE2) receptor. While it is clear that strategies to block the Spike/ACE2 interaction are promising as anti-SARS-CoV-2 therapeutics, our current understanding is insufficient for the rational design of maximally effective therapeutic molecules. Here, we investigated the mechanism of Spike/ACE2 interaction by characterizing the binding affinity and kinetics of different multimeric forms of recombinant ACE2 and Spike-RBD domain. We also engineered ACE2 into a split Nanoluciferase-based reporter system to probe the conformational landscape of Spike-RBDs in the context of the Spike trimer. Interestingly, a dimeric form of ACE2, but not monomeric ACE2, binds with high affinity to Spike and blocks viral entry in pseudotyped virus and live SARS-CoV-2 virus neutralization assays. We show that dimeric ACE2 interacts with an RBD on Spike with limited intra-Spike avidity, which nonetheless contributes to the affinity of this interaction. Additionally, we demonstrate that a proportion of Spike can simultaneously interact with multiple ACE2 dimers, indicating that more than one RBD domain in a Spike trimer can adopt an ACE2-accessible "up" conformation. Our findings have significant implications on the design strategies of therapeutic molecules that block the Spike/ACE2 interaction. The constructs we describe are freely available to the research community as molecular tools to further our understanding of SARS-CoV-2 biology.

33. SARS-COV-2 病毒的 ORF8 基因编码的蛋白质下调 MHC-I 的表达帮助病毒进行逃逸免疫

The ORF8 Protein of SARS-CoV-2 Mediates Immune Evasion through Potently Downregulating MHC-I

链接：https://www.biorxiv.org/content/10.1101/2020.05.24.111823v1

5 月 24 日，中国广东的研究团队报告了 SARS-COV-2 病毒的 ORF8 基因编码的蛋白质可以在多种细胞中和 MHC-I 直接接触并且下调 MHC-I 的表达，从而帮助病毒逃逸免疫攻击。

34. SARS-CoV-2 中一个之前没有被研究过的重叠基因和 COVID-19 大流行的进化起源有关系

A previously uncharacterized gene in SARS-CoV-2 illuminates the functional dynamics and evolutionary origins of the COVID-19 pandemic

链接：https://www.biorxiv.org/content/10.1101/2020.05.21.109280v1

加州伯克利分校的研究者研究了 SARS-CoV-2 中一个之前没有被研究过的重叠基因。重叠基因（OLGs）是指一个核酸序列可以通过不同的读码框翻译出不同的两个蛋白的基因。这类基因在病毒中很常见，往往和大流行的起源有关系，但是常常被研究人员忽视。研究者们在 SARS-CoV-2 的基因组中鉴定到一个之前没有被注释，也没有命名的 ORF3c 基因。然后作者们对 ORF3c 在三个水平进行了进化分析：不同种之间，不同宿主之间，以及同宿主之内。结果显示在 21 个有代表性的 sarbecovirus 亚属的基因组中，ORF3c 存在于一些穿山甲冠状病毒中，却不存在于更相近的蝙蝠来源冠状病毒。3978 个 SARS-CoV-2 的基因组显示 ORF3c 在 COVID-19 大流行中获得了一个新的中止密码子（G25563U），该突变在大流行中的比例突然增加。401 个对 SARS-CoV-2 的深度测序数据显示这个突变重复出现在不同的宿主（病人）中。让人感到吃惊的是，新产生的 ORF3c 中止密码子早期搭了 241U/3037U/14408U/23403G 单倍型的便车。而该单倍型有可能是驱动早期欧洲大流行扩散的单倍型。

35. 系统预测 SARS-COV-2 刺突蛋白上的所有非同义突变对蛋白稳定性以及受体结合亲和力的影响

Systemic Effects of Missense Mutations on SARS-CoV-2 Spike Glycoprotein Stability and Receptor Binding Affinity

链接：https://www.biorxiv.org/content/10.1101/2020.05.21.109835v2

5 月 23 日，美国的研究团队系统地预测了 SARS-COV-2 刺突蛋白上的所有非同义突变对蛋白稳定性以及受体结合亲和力的影响。

36. 通过生物物理模型预测感染 SARS-COV-2 的转录和翻译过程对抗病毒最有效

Biophysical modeling of the SARS-CoV-2 viral cycle reveals ideal antiviral targets

链接：https://www.biorxiv.org/content/10.1101/2020.05.22.111237v1.full.pdf

5 月 23 日，来自美国的研究团队通过生物物理模型预测 SARS-COV-2 病毒生命周期中哪些环节可能对干扰最敏感。他们的模型显示病毒的产生对病毒进入、组织以及释放的相关参数不敏感，但是对病毒转录和翻译的参数高度敏感。另外，同时靶向病毒转录和翻译可以有叠加效应。

37. 对 COVID-19 病人的血液样品进行单细胞表达测序以及 VDJ 测序

Single-cell RNA-seq and V(D)J profiling of immune cells in COVID-19 patients

链接：https://www.medrxiv.org/content/10.1101/2020.05.24.20101238v1

中国广东再生医学与健康实验室和军事医学院的团队对 13 个不同症状严重层度、病程不同的 COVID-19 病人的血液样品进行了单细胞表达测序以及免疫系统 BCR 和 TCR 的 VDJ 的测序。

38. 一个用 CRISPR/Cas9 构建的表达人 ACE2 的小鼠模型

A mouse model of SARS-CoV-2 infection and pathogenesis

链接：https://www.cell.com/action/showPdf?pii=S1931-3128%2820%2930302-4

中国食品药品检定研究院和军事医学院于 5 月 22 日在 Cell Host and Microbe 发表了采用 CRISPR/Cas9 技术构建了表达人 ACE2 基因的基因敲入小鼠（在小鼠的 ACE2 基因的二号

外显子中插入）。和野生型 C57BL/6 小鼠相比，经鼻感染的年轻和年老的 hACE2 小鼠的肺部、支气管以及脑部都有持久的高滴度病毒载量。

作者们发现通过灌胃的方式让该小鼠感染 SARS-CoV-2 可以引起病毒复制性感染，也会导致肺部病变。

39. 用腺病毒构建的表达人 ACE2 基因的用于 SARS-CoV-2 研究的小鼠模型

Mouse model of SARS-CoV-2 reveals inflammatory role of type I interferon signaling

链接：https://www.biorxiv.org/content/10.1101/2020.05.27.118893v1

耶鲁大学的作者用　腺病毒构建了一个表达人 ACE2 基因的小鼠模型，用来研究 SARS-CoV-2 的感染。病毒可以感染这些小鼠，并且可以复制，产生和病人类似的病理。I 型干扰素不能控制小鼠中 SARS-CoV-2 的复制。