Supplementary table 1: List of RCTs on off-target effect of BCG vaccine against COVID-19.

References	Sample size	Age (years)	BCG strains/dosage	Control/dosage	Location	Clinical trial registration No.	Findings
Tsilika *et al*[82]	301	≥50	Moscow/ 0.1 mL	Saline/0.1 mL	Greece	NCT04414267	BCG vaccination may provide potential protection against COVID-19 in patients aged over 50 years with comorbidities.
Moorlag *et al*[83]	2014	≥60	Danish 1331/0.1 mL	Saline/0.1 mL	Netherlands	NCT04417335	BCG vaccination boosted cytokine responses generated by influenza and SARS-CoV-2 and elicited greater antibody titers following COVID-19 infection, rather than affecting the incidence of RTIs, including SARS-CoV-2 infection, in the elderly.
Santos *et al*[84]	510	≥18	Moreau or Moscow /0.1 mL	Saline/0.1 mL	Brazil	NCT04659941	(1) No protective HR for COVID-19 infection rates has been shown by BCG (HR: 0.65, 95% CI: 0.31–1.39). (2) BCG has higher immunoglobulin G levels against COVID-19 than placebo, but no statistical significance.
Claus *et al*[85]	1511	≥18	Danish 1331/0.1 mL	Saline/0.1 mL	Netherlands	NCT04328441	(1) Vaccination with BCG vaccine did not reduce SARS-CoV-2 infection, infection duration, or severity. (2) BCG vaccination may facilitate SARS-CoV-2 antibodies during SARS-CoV-2 infection.
Upton *et al*[86]	1000	≥18	Danish 1331/0.1 mL	Saline/0.1 mL	South Africa	NCT04379336	Health care workers were not protected by BCG against SARS-CoV-2 infection or severe disease and hospitalization due to COVID-19.
Pittet *et al*[87]	3988	≥18	Danish/ 0.1 mL	Saline/0.1 mL	Australia, Netherlands,	NCT04327206	BCG vaccination had no effect on the risk of COVID-19 infection among health care workers (HR:

	Spain, England, and Brazil	1.23, 95% CI: 0.96–1.59).

BCG: Bacille Calmette-Guérin; COVID-19: Coronavirus disease 2019; HR: Hazard ratio; RCT: Randomized controlled trial; RTI: Respiratory tract infection; SARS-CoV-2: Severe acute respiratory syndrome coronavirus 2; TB: Tuberculosis; –: Not available.

Supplementary table 2: Trials of using BCG vaccine as part of COVID-19 vaccine regimen or treatment.

References	Sample size	Age (years)	Location	Study design	Objectives/findings
Ramos-Martinez *et al*[88]	60	41 (30–50)*	Mexico	Subjects were randomly assigned BCG or placebo, followed by two doses of Pfizer-BioNTech (21 days apart), and blood samples were collected 30 days after each vaccination to determine serum concentrations of Th1/Th2 cytokines. Neutralizing antibody detection and HLA-DRB loci genotyping were performed after the full immunization.	(1) Serum cytokine concentrations (i.e., IL-1β, IL-4, IL-6, IL-12p70, IL-13, IL-18, GM-CSF, INF-γ, and TNF-α) and neutralizing antibody titers were higher in the BCG vaccine-BioNTech group compared to the placebo-BioNTech group. (2) In the placebo-BioNTech group and the BCG vaccine-BioNTech group, 12 and 9 HLA-DRB1 alleles were identified, respectively. The DRB1*04 allele showed higher frequency in the placebo-BioNTech group; however, no confounding effect of this allele was found.
Counoupas *et al*[89]	–	–	Australia	Mice were vaccinated subcutaneously in the footpad with 5×10^5 CFU of BCG alone, 5 μg of SpK† combined with either BCG (BCG^{SpK}) or 100 μg of Alhyhdrogel (Invivogen, California, USA), or BCG:CoVac‡. Three weeks after the first vaccination, some mice were boosted with 5 μg SpK and 100 μg Alhydrogel. Blood samples were collected every 2 weeks after the first immunization. Experimental methods included flow cytometry	(1) BCG:CoVac induced the production of not only virus-specific IgG antibodies but also high titers of specific anti-SARS-CoV-2 neutralizing antibodies and Th1-biased cytokines. (2) Single-dose BCG:CoVac was sufficient to completely protect mice from COVID-19 infection with no detectable virus and minimal inflammatory manifestations in the mice's lungs. (3) Heterologous boosting vaccine (SpK formulated in alum) of BCG:CoVac-primed mice enhanced the SARS-CoV-2

				analysis to assess T cell responses, ELISA antibody detection and high-content live SARS-CoV-2 neutralization assay, SARS-CoV-2 challenge assay, and *Mycobacterium tuberculosis* aerosol challenge.	specific antibody response and effectively neutralized the mutant strains of B.1.1.7 and B.1.351.
Xu[90]	20	24–74	America	Screened for eligibility after informed consent, 20 patients with lighter than mild COVID-19 patients were inoculated with one-dose AD26-BCG§ via the percutaneous route of a multiple-puncture device at week 3 day 3. Patients should take testing COVID-19 by standard RT-PCR assay at week 1 day 1 and week 3 day 4, and take testing for COVID-19 spike protein derivative at week 4 day 3. And patients should report online on COVID-19 symptoms every day for up to 30 days (NCT02403505).	To assess AD26-BCG to treat infection of multiple gene mutation COVID-19 virus strains that suggests the potential for clinical benefit of COVID-19 patients.

BCG: Bacille Calmette-Guérin; COVID-19: Coronavirus disease 2019; SARS-CoV-2: Severe acute respiratory syndrome coronavirus 2; HLA-DRB: human leukocyte antigen DR beta; Th1: T help 1; Th2: T help 1; IL-1β: interleukin-1 beta; IL-4: interleukin-4; IL-6: interleukin-6; IL-12p70, IL-13: interleukin-13; IL-18: interleukin-18; GM-CSF: Granulocyte-macrophage colony-stimulating factor, INF-γ: interferon-gamma; and TNF-α: tumor necrosis factor-alpha; CFU: colony-forming unit; SpK: SARS-CoV-2 full-length spike stabilised, trimeric protein; ELISA: the enzyme-linked immunosorbent assay; AD26: replication-incompetent human adenovirus type 26 vector; –: Not available. *The data were medians (interquartile ranges) of age. †SpK was the full-length spike-stabilized trimeric protein of SARS-CoV-2, expressed and purified in EXPI293F™ cells. ‡BCG:CoVac was the combination of BCG (5×10^5 CFU), SpK (5 μg), and Alhydrogel (100 μg), and developed jointly by the University of Sydney and the Centenary Institute. §AD26-BCG was the combination of therapeutic biologics mix that was obtained with the use of Janssen Ad26 COVID-19 spike organism plus BCG organism