Mobile On Demand COVID-19 Vaccine Production Units for Developing Countries

Steffen Fahr[ag], Samantha Ayde Peña-Benavides[bd], Lukas Thiel[a], Carl Sengoba[a], Kaan Karacasulu[a], Nina Ihling[c], Juan Eduardo Sosa-Hernández[d], Gary Gilleskie[f], John M. Woodley[b], Roberto Parra-Saldivar[d*], Seyed Soheil Mansouri[b*], Kosan Roh[ae*]

[a] Process Systems Engineering (AVT.SVT), RWTH Aachen University, 52074 Aachen, Germany

[b] Department of Chemical and Biochemical Engineering, Technical University of Denmark, DK-2800 Kongens Lyngby, Denmark

[c] Biochemical Engineering (AVT.BioVT), RWTH Aachen University, 52074 Aachen, Germany

[d] Tecnologico de Monterrey, School of Engineering and Sciences, Monterrey, 64849, Mexico

[e] Department of Chemical Engineering and Applied Chemistry, Chungnam National University, 34141 Daejeon, Republic of Korea

[f] Golden LEAF Biomanufacturing Training and Education Center (BTEC), NC State University, 27606 Raleigh, NC, USA

[g] Institute of Plant and Process Technology, Technical University of Munich, 85748 Garching, Germany

Abstract: Almost two years into the COVID-19 pandemic and one year after the approval of the first vaccine, bottlenecks in production and supply chain infrastructure continue to delay vaccination campaigns in the Global South. Mobile on Demand (MOD) vaccine manufacture may help quickly ramp up production capacity while bypassing infrastructure bottlenecks. Such decentralised small-scale factories can help tip the scales in the battle against COVID-19 and future pandemics.

In this work, we designed two MOD vaccine manufacturing units based on a protein antigen expressed in yeast and *in vitro* transcription of mRNA. Each unit consists of two shipping containers and allows to produce on the order of 10,000 vaccine doses daily for competitive prices and in close proximity of their end users. Abandoning economies of scale may lead to a moderate increase in production costs that may be outweighed by reduced closed-vial dose wastage and an earlier protection of vulnerable populations.

1. Introduction

As of January 2022, the COVID-19 pandemic, caused by the coronavirus SARS-CoV-2, has caused more than 5.5 million confirmed deaths worldwide [1] and has left many more with long-term or even permanent effects [2, 3]. Since the early days of the pandemic, vaccines have been widely considered to be the only key to bringing down infection rates and keeping them permanently low. While highly effective vaccines have been developed [4] and their production has increased substantially, the race against the virus is far from being over. Firstly, while some developed countries are approaching the realm of herd

*Corresponding authors: R. Parra-Saldivar, E-mail: r.parra@tec.mx; S. S. Mansouri, E-mail: seso@kt.dtu.dk; K. Roh, E-mail: ksroh@cnu.ac.kr

immunity, the vaccination campaigns worldwide have just begun and will likely continue through this year and 2023 [5]. Secondly, new virus variants have emerged, such as the Delta variant (B.1.617.2) which is approximately 2.5 times as transmissible in non-immunised populations as the original strain [6]. More recently, Omicron (B.1.1.529) has been shown to increasingly escape the antibodies formed in response to vaccination against the original SARS-CoV-2 strain [7]. New and more transmissible virus variants are likely to emerge in the future and may require new and adapted vaccines. Thirdly, there is widespread agreement that after the global pandemic is tackled, the virus will not go extinct, but become endemic [8] and keep mutating.

In addition to the overall availability of vaccine doses, the lack of supply chain infrastructure in developing countries is a limitation to distributing vaccines to developing regions [5, 9, 10]. Deficiencies in supply chain infrastructure manifest themselves in lack of air mail capacity [5], lack of local cold chain infrastructure and unreliable power supply for cold storage [9, 10]. In the past (outside the context of COVID-19), the World Health Organization (WHO) estimated that 50 % of vaccine doses may be wasted worldwide due to problems related to storage and shipping [5]. Clearly, solutions are required that enable both a reliable global supply and rapid deployment of vaccines, while being flexible enough to respond to local outbreaks and new variants emerging in the future.

Mobile on demand (MOD) vaccine production provides a solution to the aforementioned problems. Here, MOD production is defined as the local production of vaccines in small standardised facilities supplying their surrounding environment. Localised production and distribution stands in contrast to the conventional approach of a centralised production in few unique large-scale plants. MOD production enables local production of vaccines in the regions of most need and thereby avoids supply chain infrastructure limitations such as an insufficient capacity of planes and airports to ship and receive cooled vaccine doses via air freight. Moreover, the MOD plants may be moved between highly affected areas to quickly tackle local outbreaks.

MOD vaccine production can take place based on an established vaccine platform such as *in vitro* production of mRNA-based vaccines [4, 11, 12] or yeast-based production of virus proteins [13]. These very distinct platforms are chosen as examples in this work and will in the following be referred to as "yeast" and "mRNA", respectively. The production process for a given platform will be similar for different vaccines and enable use of the same production facility for all vaccines within the platform. The two chosen platforms belong to the category of recombinant vaccines that exhibit several advantages over the more established production of inactivated or live-attenuated viruses [14]. For example, in the production of recombinant vaccines, no human pathogen is deliberately introduced into the process, making them safer and easier to produce in small-scale plants [14, 15]. Viral vectors are also recombinant vaccines, but their production often requires mammalian cell lines, which are challenging to cultivate [16] and result in a time-consuming process [17].

Kis *et al.* [18] discuss and compare the economic feasibility of different vaccine types and the associated production platforms in a centralised and a distributed production scenario. However, the distributed scenario still assumes facilities producing one to several million doses per month, which places the distributed scenario in between fully centralised and fully MOD production as we define it. While Kis *et al.* [18] develop production schemes at the flowsheet level, a more detailed discussion of the design of the miniaturised plant for MOD production in developing countries is missing. Schmidt *et al.* [19] develop a digital twin of a state-of-the-art, centralised mRNA production process that, among others, facilitates training of new personnel. On the industry side, Curevac holds a patent referring to small-scale mRNA production [20]. The patent focuses on a reactor concept, without considering essential parts of the overall process, such as downstream processing [20].

Yeast-based fermentation for producing biopharmaceuticals has been suggested for MOD manufacturing. For instance, Crowell *et al.* [21] present a highly-automated fermentation process, capable of producing hundreds to thousands of doses of protein biologics, such as human growth hormone (hGH), interferon-alpha (IFN-alpha)-2b, and granulocyte colony stimulating factor (G-CSF), in perfusion mode. Other studies successfully demonstrate the production of these three proteins, from a decentralised perspective [22, 23]. In the context of COVID-19, Baylor College of Medicine and the Center for Genetic Engineering and Biotechnology (CIGB) each developed a protein subunit vaccine based on the spike protein receptor binding domain (RBD) of the SARS-CoV-2 virus using yeast as an expression host. The manufacture is based on production of a protein antigen by means of a classical large-scale fermentation process, making it suitable for technology transfer to developing country vaccine manufacturers [24, 25]. Nevertheless, a MOD plant design with techno-economic evaluation is missing for a COVID-19 yeast-based vaccine.

In this article, two small-scale, container-based vaccine production processes based on yeast and mRNA are designed. The aim of this study is to quantitatively compare the developed processes to each other and to a centralised production in terms of their potential for combating a virus pandemic in low and middle income countries. First, the designed small-scale processes and the container concept are described. Subsequently, the cost per vaccine dose of for both platforms are analysed and compared to a centralised production scenario. Finally, we conduct a facility location optimisation for a vaccination campaign in Nigeria as an application example.

2. Results

For both vaccine production processes, we create process models in SuperPro Designer [26], which we use to calculate mass and energy balances and to determine basic information on equipment sizing. In light of uncertain parameters, e.g., raw material prices and feasible batch volume, we specify a base scenario for each process (see Table 1), and then conduct a sensitivity analysis (see Section 2.3).

The mRNA process starts with *in vitro* transcription of a linearised DNA template with co-translational

capping [27], the raw materials for which are produced off-site. The mRNA is purified using a series of tangential flow filtration (TFF) steps and an Oligo-dT affinity chromatography column [28, 29]. Finally, with the addition of functional lipids, the product is formed into lipid nanoparticles (LNP) using a microfluidic device [30–33], and filled into vials after a final buffer exchange and sterile filtration [34]. The base case assumes a dose size of 100 μg mRNA, as is also present in the Moderna COVID-19 vaccine [35], and a production capacity of 10,000 doses per batch (see Table 1). We found that a batch can be finished within one day. Also, raw materials and single use equipment (SUE) for up to 100 batches as well as 210,000 finished vaccine doses can be stored in the containers. Therefore, a full vaccination campaign (see Table 1) can be conducted without resupply using the materials initially present in the container after delivery and is finished within 100 days. The mRNA process requires highly specific and difficult-to-ship raw materials, such as linearised DNA and a capping agent, so the frequency of resupply should be considered in the process analysis.

The yeast process starts by thawing a vial from the working cell bank of the yeast *Komagataella phaffii*, also known as *Pichia pastoris* and inoculating the first bioreactor in the train. Glycerol is used as the carbon source for biomass growth. After reaching a biomass concentration of 30 g of cell dry weight (CDW) $L^{-}1$ in the production bioreactor, the production of the protein of interest, i.e. the SARS-CoV-2 spike protein, is induced by changing the carbon source to methanol [36–39]. The primary recovery of the secreted protein is carried out by filtration to separate the product from the cells. The subsequent purification sequence is based on hydrophobic interaction chromatography (HIC), TFF, and anion-exchange chromatography (AEC) [39, 40]. The base case for the yeast process assumes a production of 18,000 doses per batch each containing 50 μg of the antigen each and adjuvanted with 750 μg aluminium hydroxide (alum) and 500 μg of CpG 1018 [41]. For this process, 18 batches worth or raw materials can be stored in the containers. The time to produce a batch is found to be seven days without or four days with overlapping scheduling. More detailed process descriptions along with the flowsheets of the yeast and the mRNA processes are given in the supplementary information (SI).

Table 1: Overview of key assumptions and parameters for mRNA and yeast process.

Category	mRNA	Yeast
Dose size / μg (mRNA or spike protein)	100	50
Doses per batch	10,000	18,000
Batches per campaign	100	100
Campaigns per amortisation period	1	1
Min. campaign duration / days	100	400
Max. no. of batches per resupply	100	18

2.1. Container Design

Both small-scale processes were adapted to be run in two ISO-668 20-foot standard shipping containers each. SUE was determined to be the most appropriate for MOD production. First, use of SUE is widespread in pharmaceutical production and well known for reduced fixed costs and increased flexibility when setting up new production facilities [42, 43]. Second, SUE may be more sustainable than traditional biomanufacturing processes given the need for less purified water for cleaning in place and steaming in place [44, 45].

To produce pharmaceutical grade products, a cleanroom environment must be maintained inside the containers [46]. To this end, heating, ventilation, and air conditioning (HCAV) systems with high efficiency particulate air (HEPA) filtration should be installed. These systems ensure 20 air changes per hour [46], as well as a controlled room temperature in any climate. Highly purified water is necessary for both processes (e.g., RNase-free water for the mRNA process), which is produced on site by filtration of drinking water [47]. Due to the cooling requirements for the stored substrates and products, a continuous electricity supply is necessary, which cannot be guaranteed from the local grids [5, 48]. Therefore, a 13.5 kWh lithium-ion battery system is integrated into the floor of the containers, ensuring, e.g., for mRNA, 5.7 and 7.6 days of grid-independent operation during production downtime, respectively. Downtime electricity consumption is almost identical as it is dominated by refrigeration and the HVAC system. The batteries are used during road transport and in case of a power outage, which takes place frequently and lasts for around two hours on average in Nigeria, for example [48]. Finally, for both processes, filling, quality control, and storage of raw materials, SUE and products at cold and ambient temperature are included in the economic and spacial considerations. Figure 1 illustrates a possible configuration for a pair of mRNA production containers. More details on the spacial considerations and devices used can be found in the SI and in [49].

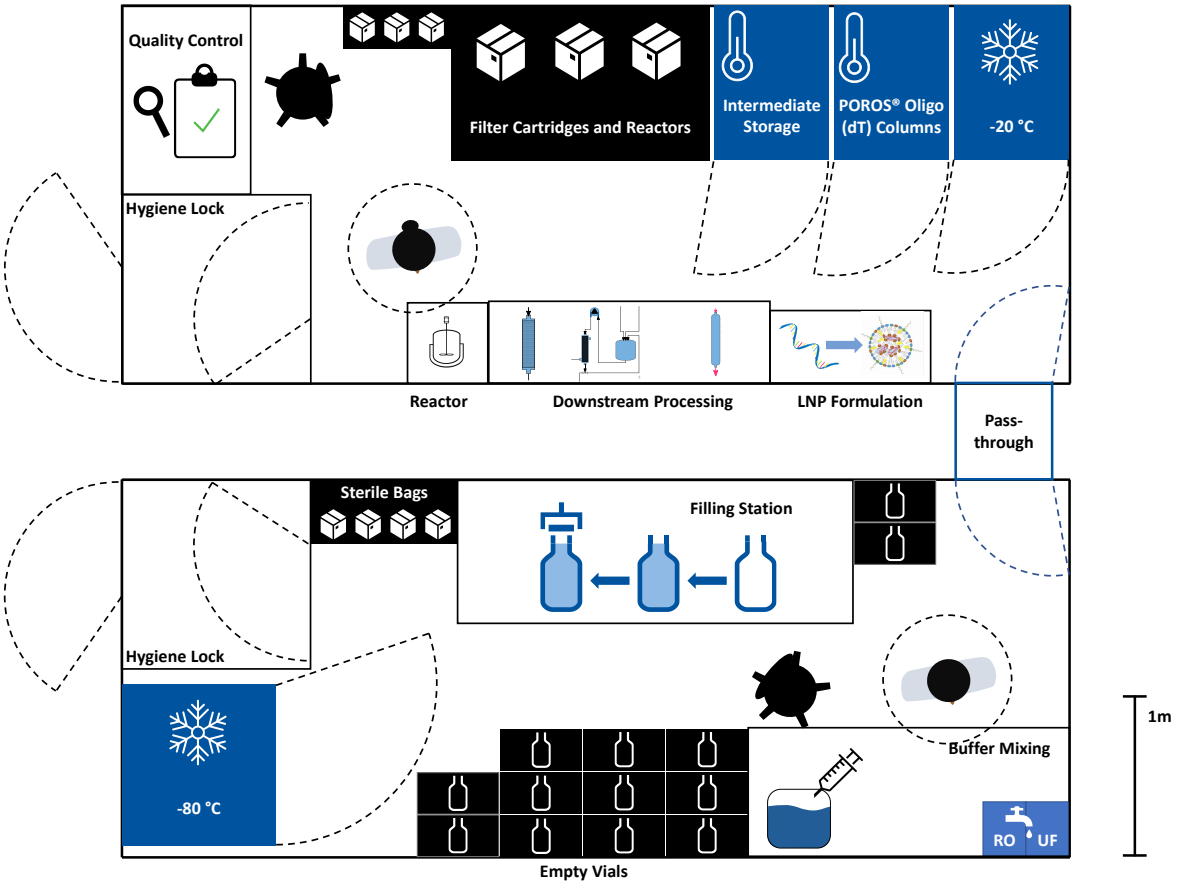


Figure 1: Schematic layout of the containerized mRNA production unit consisting of two shipping containers. Container dimensions shown are the inner dimensions of 2.35x5.90 m.

2.2. Cost Analysis

In the base scenario (see Table. 1), the total production costs per dose are 20.84 and 8.06 € for the mRNA and the yeast processes, respectively (see Figure 2). Zero residual value of the equipment after use is assumed, i.e. the investment costs are divided by the number of doses produced in the assumed plant lifetime. For both processes, the investment costs are relatively evenly distributed between process equipment and auxiliary materials such as shipping containers, freezers and air conditioning, including the building costs. The category "consumables" refers to items and materials consumed during the production process that do not enter the main process stream, such as single-use bags, filter cartridges, and chromatography resins. In the mRNA process, proprietary consumables and raw materials represent major cost contributors. These include the capping agent CleanCap® [11, 27], the Oligo-dT affinity resin used for purification [29], and the SUE used in microfluidic LNP formation [50, 51] accounting for 2.96, 2.97, and 5.95 € per dose, respectively.

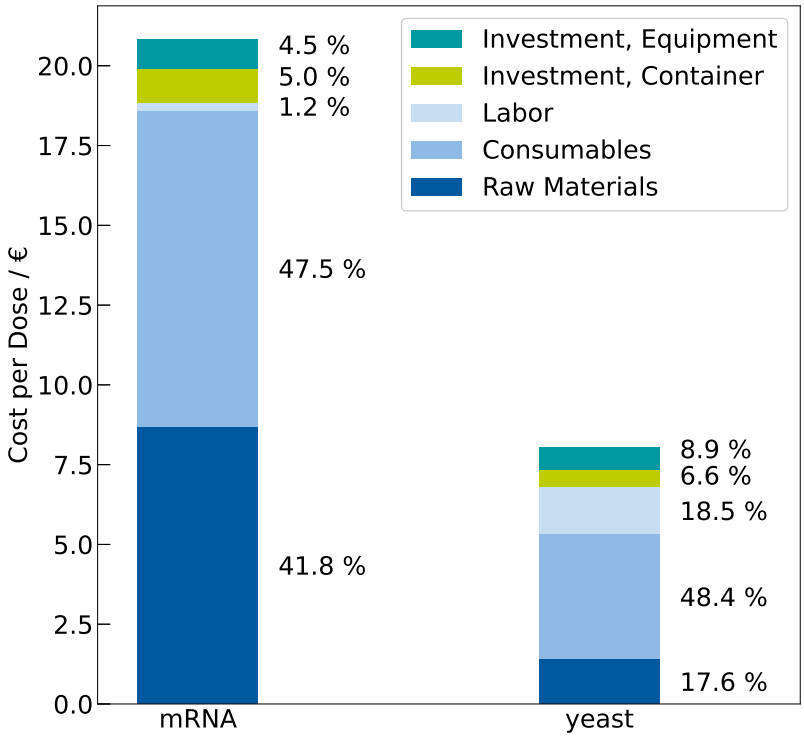


Figure 2: Breakdown of the production cost per dose for the mRNA and the yeast processes.

For the yeast process, consumables are the major cost contributors (48.4 %) due to the use of single-use bioreactors and the single-use packed chromatography columns that are replaced after every batch. Due to the longer batch duration associated with the yeast process compared to mRNA, labor cost accounts for a more significant proportion. The raw materials representing 17.6 % of the final vaccine cost include those materials used for cultivating the yeast and the adjuvants added in the formulation step. The former are cheap and accessible, representing only 1.13 % of the total costs. However, the adjuvants that are added later in the process alone account for 16.5 % of the total dose cost. Protein subunit vaccines are formulated with adjuvants to elicit a stronger immune response [52]. Alum is the most commonly used adjuvant in vaccines globally, and is inexpensive [52]. Nevertheless, most COVID-19 protein-based vaccines contain a second adjuvant, namely CpG 1018, to improve the immune response [52]. The cost of this adjuvant was not provided by the manufacturer, but was estimated by the cost of finishing operations in vaccine production, as it includes formulation with adjuvants or stabilisers [53], accounting for 1.33 € per dose (see the SI). For both processes, the cost of quality control equipment and its operation is included with the equipment and consumable costs. A description of quality control measures is included in the SI. The operators will be free to perform such tasks during most of the batch duration, as action is mostly required between process steps not during them.

2.3. Sensitivity Analysis

We carried out a local sensitivity analysis, which varies individual parameters within a realistic range to quantify the effect of certain changes to the process or the pricing of raw materials on the final production cost.

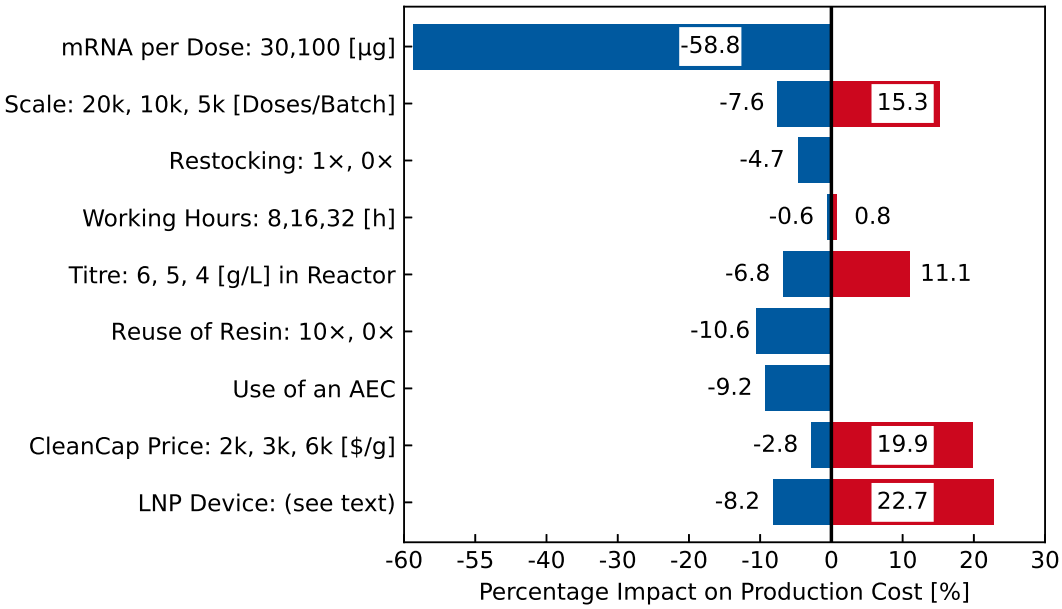
The amount of active ingredient per dose ("dose size") varies within each platform. The currently approved mRNA vaccines contain 30 - 100 μg of mRNA per dose [35] with dose sizes down to 3 μg intended for children [54]. For yeast-based protein vaccines, 15 - 50 μg of protein per dose are currently investigated in clinical trials [41]. Due to the expensive raw materials required for producing mRNA, the effect of dose size on the production cost per dose is much bigger for the mRNA case than it is is for yeast (see Figure 3). In addition to directly decreasing the cost per dose, a smaller dose size allows production of more doses per batch. For example, when producing 33,333 doses of 30 μg mRNA instead of 10,000 doses of 100 μg mRNA, the production cost per dose drops by 72 % to 5.82 €. For the yeast process, the scale had the biggest influence on the final cost per dose. For instance, producing 36,000 doses of 50 μg protein decreases the cost per dose to 5.23 €.

Doubling the plant lifetime of the mRNA plant from one to two campaigns by restocking it with raw materials after producing 100 batches reduces the cost per dose by approximately 5 %. A similar moderate effect of plant lifetime on production cost per dose can be observed for the yeast-based process, with a cost reduction by 7.74 %. Note that producing 100 batches takes 100 and 400 days and results in 1 and 1.8 million doses in total for the mRNA and yeast process, respectively (compare Table 1).

Having to increase the number of employees per shift or being able to decrease labor cost, e.g., through automation, could reduce the production costs significantly for the more labor-intensive yeast process, as labor represents almost 20 % of the costs. For the mRNA process, the product titre after the reaction step for a fixed raw material usage (i.e., the conversion efficiency) has a significant effect on the production cost. This is also the case for yeast, as here, the bioreactor volume limits the process scale, and therefore the titre affects the feasible scale. Reusing chromatography resins can lead to significant cost savings in both processes and should therefore be considered in practice.

In the mRNA process, the cost of certain raw materials and process equipment has a large effect on the production cost per dose. These materials include the affinity resin, the capping agent CleanCap® and the operation cost of the microfluidic device for LNP formation. As an alternative to resin reuse, the cost of resin may be decreased by replacement of affinity chromatography by AEC [19, 55]. For the LNP formation device, we choose to include a 13 % discount in the sensitivity analysis as well as another device that is GMP certified, but does not exactly fit our desired scale and is therefore less cost-effective. The cost of CleanCap® and the microfluidic cartridges may vary substantially and depend on negotiations with the manufacturers including potential discounts for products intended for low and middle income countries [56]. With all sensitivity factors taken into account, the mRNA production cost per dose ranges

(a)



(b)

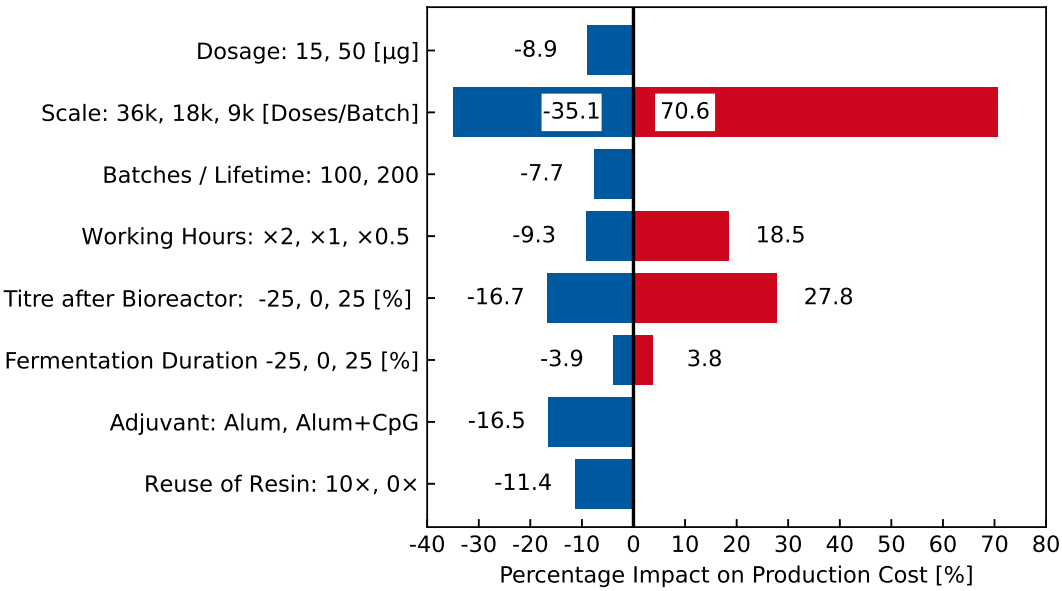


Figure 3: Sensitivity analysis results of (a) mRNA and (b) yeast process. mRNA production is highly sensitive to dose size, proprietary equipment and raw materials. Yeast-based vaccine production strongly depends on dose size, batch size and product titre after the bioreactor with constant raw material input.

from 4.24 to 46.82 €.

The yeast-based vaccine is expected to use an expensive proprietary adjuvant [53], (see Section 2.2). The sensitivity of the adjuvant covers the effect of having alum exclusively in the formulation, as it is the case for some yeast- and protein- based COVID-19 vaccines, such as the highly effective Abdala vaccine (CIGB-66) [57] and one vaccine candidate by Chen *et al.* [58]. With all sensitivity factors taken into account, the yeast-based vaccine production cost per dose ranges from 2.61 to 18.19 €.

Recently, Kis *et al.* [59] performed a global sensitivity analysis on centralised mRNA, self-amplifying RNA (saRNA) and yeast-based viral vector vaccine production and identified the same main sensitivity factors for the yeast- and mRNA-based vaccines.

Investigating the effect of the process scale in terms of doses per batch and batches per lifetime on the production cost per dose is of high interest in a MOD scenario. Figure 4 shows the effect of these factors on vaccine production costs for a fixed dose size. The batch size of both MOD processes is limited due to spacial limitations visualised in Figure 4 as hatched areas. The costs achieved in these areas would therefore only be feasible when relaxing the spacial constraints of MOD production as assumed in this work. Note that, when producing a vaccine using a smaller dose size, the limitation is shifted right to a larger number of doses per batch (see the SI). A special feature of the mRNA production plant is its ability to run without resupply for up to 100 batches with 10,000 doses per batch. If the number of doses per batch is increased over the base case (white point), raw material storage becomes limiting. In contrast, if the number of doses per batch is decreased, the storage space available for SUE limits the number of batches produced in between resupplies. Resupply for the yeast-based process is expected to be necessary more frequently than for mRNA, but at the same time to be less problematic. While many materials for the mRNA process are delivered frozen and are unlikely to be available locally, the raw materials for the yeast process are dominated by the carbon sources, glycerol and methanol, which may be available locally and shipped more easily.

Overall, the vaccine production costs are highly sensitive to dose size, batch size, and purchase prices of consumables and raw materials. The charts in Figure 4 show that further scale-up of the processes or a longer operational lifetime beyond the chosen operation point comes with only a moderate decrease

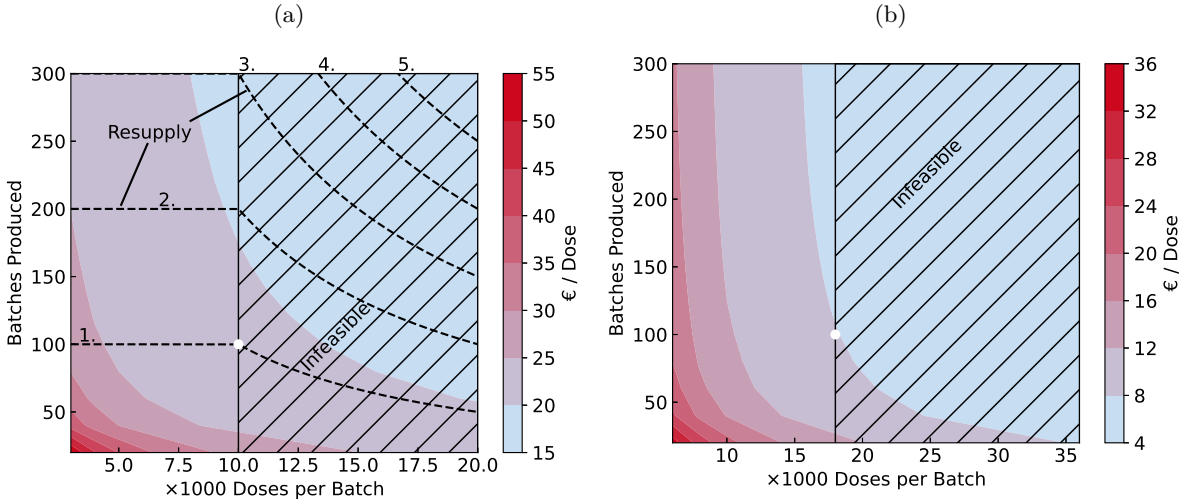


Figure 4: Sensitivity of dose size and batch scale on the production cost per dose of mRNA (a) and yeast-based (b) vaccine under otherwise base assumptions (see Table 1). The hatched area marks a possible feasibility limit that depends on the dose size. Dashed lines mark resupplies of raw materials and SUE needed for mRNA production after a certain number of produced batches. White points mark the base scenario operation point. Dose sizes of 100 μg mRNA and 50 μg protein per are assumed

in costs. However, for less than 50 batches of less than 5,000 and 10,000 doses of mRNA and yeast-based vaccine, respectively, the vaccines become exceedingly expensive to produce, i.e, the plants should produce more than approximately 250,000 - 500,000 doses during their lifetime.

2.4. Comparison with Centralised Production

We showed that MOD vaccines based on an mRNA and yeast platform are technically feasible at the production costs of 20.84 and 8.06 € per dose, respectively. For comparison, current selling prices per dose range from 8.62 to 31.98 € for the Moderna mRNA vaccine and from 5.82 to 20 € for the Pfizer/BioNTech vaccine [60]. In contrast to our base assumption of 100 μg of mRNA per dose, the Pfizer/BioNTech vaccine requires only 30 μg mRNA per dose, for which we calculate a cost per dose of 12.24 € with all other parameters held at their base value. The Novavax vaccine Covovax is a protein subunit vaccine like the one proposed in this work, although not produced by yeast, and is sold for 2.59 - 18.01 € per dose. The cheapest vaccine currently listed by UNICEF is the AstraZeneca viral vector vaccine Vaxzevria, sold for 1.89 € per dose [60] to the European Commission.

As shown in Figure 2, a significant portion of the vaccine cost is associated with raw materials and therefore, on a per-dose basis, stays constant when scaling the process. However, previous works studying centralised vaccine production assumed much cheaper purchase prices for some raw materials, such as CleanCap® for mRNA [43] and the adjuvant CpG 1018 for the yeast-based vaccine [61]. If these prices are achieved through volume discounts, we would expect the same discounts to also apply to a distributed concept such as ours with a central raw material supply. The mentioned works on centralised vaccine production estimate the production cost to be around 1.72 and 1.30 € per dose for mRNA and yeast-based vaccine, respectively [43, 61]. The lower production costs can be partly attributed to cheaper raw material purchase prices assumed, and partly to economies of scale.

The reduced need for shipping of MOD-produced vaccines reduces the cost to produce and distribute a dose by two mechanisms. Firstly, the cost of shipping and intermediate storage is directly reduced. The shipping costs in Vietnam [62] and various African countries [63–65] have been found by observational studies [62, 65] and simulations [63, 64] to lie on the order of 0.2 - 0.7 € per dose. The second effect is the reduced loss of doses. Eliminating interregional transport and regional intermediate storage hubs from the distribution network reduces the occurrence of storage temperature excursions [9]. Considering that previous works report a 50 % shipment-related waste of vaccine doses worldwide [5], reducing dose spoilage represents a strong economic incentive.

Another point to consider are investment costs. Centralised production requires large on-time investments and construction projects usually taking on the order of seven years [18], although having been significantly accelerated during the COVID-19 pandemic. Meanwhile, achieving the same manufacturing capacity as large centralised facilities, MOD manufacture requires scale-out of production units, which

lacks the classic economies of scale as found in centralised plants. A comparison of investment costs reported in previous literature with our results is shown in Table 2. For mRNA, the results clearly show reduced investment costs for a single very large-scale plant as compared to building many smaller facilities, whereas, for yeast, our concept compares quite favorably to more large-scale concepts. It should be noted that the container-based plants, while potentially being cheaper, may have a shorter lifetime than a conventional facility. While investment costs are important to consider, only a small proportion of the production cost per dose is associated with them (see Figure 2), limiting their effect on the cost per dose.

Table 2: Investment cost comparison with previous literature. Smaller plants are scaled-out to match the production of the platform-specific large-scale example.

Platform		Large-scale	Intermediate-scale	MOD
mRNA	No. of plants	1	476	2,192
	Production capacity, 10^6 doses year $^{-1}$ plant $^{-1}$	8,000	17	3.65
	Total investment cost, 10^6 €	1,800	11,700	4,318
	Source	[18]	[43]	This study
Yeast	No. of plants	1	3	18
	Production capacity, 10^6 doses year^{-1} plant $^{-1}$	30	10	1.64
	Total investment cost, 10^6 €	91 - 200	81 - 174	40
	Source	[66]	[66]	This study

Besides complex construction projects, building large-scale plants requires scale-up of the process to unprecedented scale, whereas the MOD plants produce at the same scale as required for clinical trials and therefore bypass the technological risk and development time of upscaling. The first MOD plants supplying regions of most need can hence be commissioned shortly after approval of a new vaccine or pharmaceutical. Castillo *et al.* [67] reported that having an additional production capacity of one billion doses per year in early 2021 would have had a global benefit of 540 - 1,940 billion USD depending on the capacity already available at that point. Such numbers dwarf the difference in investment cost and even annual operating cost of MOD plants as compared to centralised facilities. In other words, if MOD plants allow to bring more doses to the market much more quickly than centralised plants, the potentially higher cost per dose might well be worth paying.

2.5. Facility Location - Case Study for Nigeria

The choice of batch sizes for the MOD processes was motivated by providing attractive economics while still adhering to the container concept. To give a notion of what this size means in practice, we provide an exemplary distribution of MOD plants in Nigeria, a country of the Global South with a moderate population density (see Figure 5). This particular distribution assumes mRNA production units that produce 100 batches of 10,000 doses each. To position the production units (consisting of two shipping containers each, see Section 2.1), the country is discretised into 774 points, which correspond to its local government areas (LGAs). Each of the LGAs should be provided with two vaccine doses for at least

80 % of its population during the campaign. When constraining the number of units to correspond to the exact minimum required to reach the overall vaccination rate (i.e., no overcapacity), the longest distance a vaccine dose must travel from its production site to the site of use is 81 km. When instead constraining the maximum travel distance to 50 km, 330 units are required, compared to a minimum of 310 units that would be required to supply the Nigerian population without considering travel constraints, corresponding to 94 % utilisation. Despite using simplifying assumptions such as geometrical distance between LGA centers, the results give a good impression of the practical implication of the chosen production volume per unit. Accordingly, the plant size and allocation strategy is well-suited to conduct a fast vaccination campaign for a country of Nigeria's population density.

For much lower population densities, the units supply exceedingly large areas, increasing the distance, doses are shipped (see the SI). Producing vaccines at approximately a quarter the rate as the mRNA process, the yeast-based process either leads to a longer campaign duration of about one year, or requires to position more units that produce less than 100 batches each before being collected for another campaign. The latter strategy is attractive for countries with a low population density, but increases plant off-times. The feasibility of moving plants during a campaign and its implications on productivity and economics are considered outside the scope of this work and may be considered in future work.

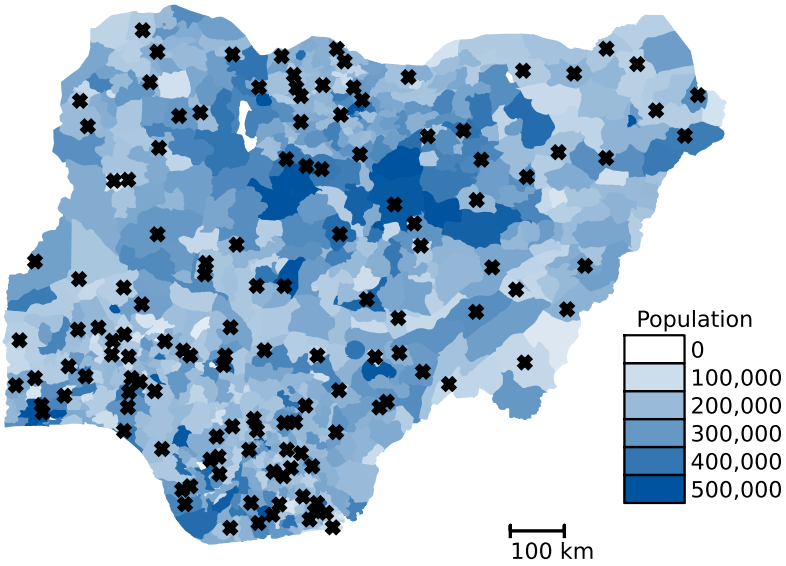


Figure 5: Optimal distribution of mRNA production units in Nigeria. Crosses mark position of one or more production units.

3. Discussion

In this work, we developed MOD vaccine production processes based on an mRNA and a yeast platform. The batch sizes were adapted to be compatible with small-scale plants that can be operated in two 20-foot

shipping containers each. The processes are based on currently available technology that matches the space constraints of the shipping containers. We thereby show that MOD manufacture of COVID-19 vaccines is technically feasible. The expected costs per dose are 20.84 € for mRNA and 8.06 € for yeast for the base scenario, but can vary substantially depending on raw material costs, dose size and the chosen production volume per MOD plant.

The cost and technical feasibility of the MOD mRNA process is highly dependent on purchase costs and availability of specific, often proprietary raw materials and equipment required due to novelty of the mRNA platform. This dependence on suppliers can also be understood as a lack of vertical integration in the MOD plants. The short development times for new mRNA vaccines and minimal process changes are strengths of the mRNA platform. Additionally, mRNA production uses smaller liquid volumes than an equivalent fermentation process, which is favorable when considering production in a constraint space. Moreover, due to the short batch duration, the mRNA MOD plants can conduct vaccination campaigns quickly without requiring any resupply after the initial setup. After that, the plants can be collected and restocked for a new campaign in another region, even though this is not necessary from an economic standpoint. Future alternative uses may be the small-scale production of cancer vaccines [68].

The production of yeast-based vaccines is associated with a longer batch duration. Consequently, to achieve a reasonable overall production volume, the plants should be operated for more than a year. This may be longer than an ideal vaccination campaign following an acute local outbreak should take. However, an appropriate overall production volume can be achieved by relocating the plants. Due to the larger fluid volumes processed, the yeast-based plant requires more frequent resupply of raw materials and SUE than the mRNA plant, but many of the required materials for this more conventional process may be available locally. Moreover, local expertise might be available with this type of process in contrast to *in vitro* transcription of mRNA [24]. As proteins structurally differ more from one another than the mRNA encoding them does, it remains to show that the yeast process allows to switch to a new product with minimal process changes.

When comparing the two considered platforms for MOD vaccine production in lower and middle income countries, we conclude that the reduced process complexity, batch duration and space demand renders the mRNA process more suitable for production in a confined space. However, if practical studies confirm that the MOD yeast process is indeed feasible, it offers reduced vaccine production costs and reliance on proprietary raw materials.

The production costs per dose produced with MOD procedures we calculated were higher than the estimated production costs for centralised production [43, 61]. Furthermore, the production costs were higher than some current market prices for COVID-19 vaccines [60]. However, savings in the cost per MOD-produced vaccine dose may, for example, be achieved by serial production of MOD plants and reduction in shipping-related loss of doses, but are currently difficult to quantify. Last but not least,

using MOD plants may provide much needed vaccine doses more quickly than centralised production does, which may ultimately justify the extra costs.

Because we have determined that MOD vaccine production is feasible and potentially attractive, future work should focus on a practical implementation of this approach. Despite our efforts to obtain accurate cost estimates for individual cost factors, we end up with a considerable uncertainty range in terms of final costs. This uncertainty range could be narrowed by establishing a pilot plant to determine the production costs more accurately. Additionally, further aspects that were not considered in this study (e.g., licensing, GMP certification, availability of trained personnel and potential political hurdles) will likely affect both the feasibility and the production costs. For example, a 2020 guideline by the FDA calls for facilities of suitable size and construction for the production of COVID-19 vaccines, which may impede the success of container-based plants [69]. Moreover, from raw material production to administering of the vaccine, there are many potential bottlenecks beyond shipping and vaccine production that should be assessed rigorously in future work.

4. Methods

4.1. Container Location Problem

The results displayed in Figure 5 were obtained using GUROBI and its python interface GUROBIPY. Solution times were on the order of 100 s on an Intel Core i7-10510U CPU (using 4 cores @ 1.80 GHz). The results represent the globally optimal solution (optimality tolerance of 10^{-4}) to the optimisation problem given by

$$\min_{x_{i,j},z_{i,j},y_i,d_{max}} \quad d_{\max} \tag{1a}$$

$$s.t. \quad n_{\max} \leq \sum_{i \in \text{LGAs}} y_i \tag{1b}$$

$$z_{i,j} d_{i,j} \leq d_{\max} \; \forall \; i,j \in \text{LGAs} \tag{1c}$$

$$x_{i,j} \leq z_{i,j} \; \forall \; i,j \in \text{LGAs} \tag{1d}$$

$$\sum_{i \in \text{LGAs}} x_{i,j} \leq 1 \; \forall \; j \in \text{LGAs} \tag{1e}$$

$$R_{\min} \leq \sum_{i \in \text{LGAs}} x_{i,j} \; \forall \; j \in \text{LGAs} \tag{1f}$$

$$n_{\text{shots}} * \sum_{j \in \text{LGAs}} x_{i,j} p_j \leq c_{\max} * y_i \; \forall \; i \in \text{LGAs} \tag{1g}$$

$$x_{i,j} \leq 1 \; \forall \; i,j \in \text{LGAs} \tag{1h}$$

$$x_{i,j} \leq y_i \; \forall \; i,j \in \text{LGAs}. \tag{1i}$$

Therein, the objective function $d_{\max}$ describes the maximum delivery distance of a dose and the parameters $d_{i,j}\ \forall\ i,j \in$ LGAs mark the distance between two LGAs. The latter distance is simplistically determined as a geometrical distance, but can be replaced by the real driving distance or respective transport cost, if such data is available. The integer variables $y_i\ \forall\ i \in$ LGAs correspond to the number of containers positioned the respective LGA. The continuous variables $x_{i,j}\ \forall\ i,j \in$ LGAs denote the proportion of the demand in LGA j satisfied by the plants in LGA i. The binary variables $z_{i,j}\ \forall\ i,j \in$ LGAs determine if supply from i to j exists. The constant n_{shots} describes the number of doses per person, which is assumed to be 2 and the parameter $R_{\min}$ describes the minimum vaccination rate. The parameters p_j describe the population of the individual LGAs.

The constraint Eq. (1b) limits the number of containers to a pre-specified number, for example the minimum number necessary to produce enough doses for the overall population of the country. The constraints Eq. (1g) limit the number of doses produced per unit to $c_{\max}$, which is set to 1 million. Constraint Eq. (1f) sets a minimum vaccination rate R_{min}, e.g., 80%, that must be satisfied in each region to achieve local herd immunity. The last constraint Eq (1i) is not necessary but helps accelerate the solution time of the optimisation. Moreover, we set those $x_{i,j}$ to zero a priori for which the corresponding distance $d_{i,j}$ is above a certain threshold without altering the result of the optimisation.

The optimisation can easily be modified, for example by replacing the objective function by the number of containers and fixing a maximum distance driven, $d_{\max}$. This allows to calculate the minimum number of containers necessary for a given maximum driving distance.

Contributions S.S.M. conceived the study. S.S.M. and K.R. organized the research team. S.S.M.. K.K., N.I. and K.R. supervised the project. S.A.P.B. conceptualized the yeast process and prepared the process simulation. L.K., S.F. and C.S. conceptualized and designed the mRNA process. C.S. and S.F. conducted the allocation optimisation. N.I. reviewed and helped finalize both process models. S.F., S.A.P.B. and L.T. wrote the original draft of the manuscript. All authors reviewed and commented on the manuscript.

Acknowledgements We would like to thank Svenja Battling and Andreas Biselli for fruitful discussions and their support in setting up the process simulations. S.F. would like to thank Steffan Schlein for his consultation concerning the optimisation problem. We are thankful for the sponsorship provided by Centro CEMEX-Tec de Monterrey para el Desarrollo de Comunidades Sostenibles, Fundación FEMSA, and Fundación Banorte to enable the contribution of S.A.P.B. to this project. We further gratefully acknowledge Jochen Büchs and Alexander Mitsos for supporting this work.

Conflict of interest We have no conflicts of interest to disclose.

References

1. World Health Organization. *WHO Coronavirus (COVID-19) Dashboard* Accessed: 2022/01/16. 2022. https://covid19.who.int.
2. Garg, M., Maralakunte, M., Garg, S., Dhooria, S., Sehgal, I., Bhalla, A. S., *et al.* The Conundrum of 'Long-COVID-19': A Narrative Review. *International journal of general medicine* **14,** 2491–2506 (2021).
3. Cabrera Martimbianco, A. L., Pacheco, R. L., Bagattini, Â. M. & Riera, R. Frequency, signs and symptoms, and criteria adopted for long COVID-19: A systematic review. *International Journal of Clinical Practice* **75,** e14357 (2021).
4. Crommelin, D. J. A., Anchordoquy, T. J., Volkin, D. B., Jiskoot, W. & Mastrobattista, E. Addressing the Cold Reality of mRNA Vaccine Stability. *Journal of pharmaceutical sciences* **110,** 997–1001 (2021).
5. Park, C.-Y., Kim, K., Helble, M. & Roth, S. Getting Ready for the COVID-19 Vaccine Rollout (2021).
6. Burki, T. K. Lifting of COVID-19 restrictions in the UK and the Delta variant. *The Lancet Respiratory Medicine* (2021).
7. Wilhelm, A., Widera, M., Grikscheit, K., Toptan, T., Schenk, B., Pallas, C., *et al.* Reduced Neutralization of SARS-CoV-2 Omicron Variant by Vaccine Sera and monoclonal antibodies. *medRxiv* (2021).
8. Torjesen, I. Covid-19 will become endemic but with decreased potency over time, scientists believe. *BMJ (Clinical research ed.)* **372,** n494 (2021).
9. James, E. R. Disrupting vaccine logistics. *International health* **13,** 211–214 (2021).
10. Eccleston-Turner, M. & Upton, H. International Collaboration to Ensure Equitable Access to Vaccines for COVID-19: The ACT-Accelerator and the COVAX Facility. *The Milbank quarterly* (2021).
11. Kis, Z., Kontoravdi, C., Dey, A. K., Shattock, R. & Shah, N. Rapid development and deployment of high-volume vaccines for pandemic response. *Journal of advanced manufacturing and processing* **2,** e10060 (2020).
12. Sahin, U., Karikó, K. & Türeci, Ö. mRNA-based therapeutics–developing a new class of drugs. *Nature reviews. Drug discovery* **13,** 759–780 (2014).
13. Karbalaei, M., Rezaee, S. A. & Farsiani, H. Pichia pastoris: A highly successful expression system for optimal synthesis of heterologous proteins. *Journal of Cellular Physiology* **235,** 5867–5881 (2020).
14. Yadav, T., Srivastava, N., Mishra, G., Dhama, K., Kumar, S., Puri, B., *et al.* Recombinant vaccines for COVID-19. *Human vaccines & immunotherapeutics* **16,** 2905–2912 (2020).

15. A Comparison of Plasmid DNA and mRNA as Vaccine Technologies. *Vaccines* **7,** 37 (2019).

16. Shanmugaraj, B., Malla, A. & Phoolcharoen, W. Emergence of Novel Coronavirus 2019-nCoV: Need for Rapid Vaccine and Biologics Development. *Pathogens* **9** (2020).

17. Reardon, S. Cuba's bet on home-grown COVID vaccines is paying off. *Nature* **600,** 15–16 (2021).

18. Kis, Z., Papathanasiou, M., Calvo–Serrano, R., Kontoravdi, C. & Shah, N. A model–based quantification of the impact of new manufacturing technologies on developing country vaccine supply chain performance: A Kenyan case study. *Journal of Advanced Manufacturing and Processing* **1** (2019).

19. Schmidt, A., Helgers, H., Vetter, F. L., Juckers, A. & Strube, J. Digital Twin of mRNA-Based SARS-COVID-19 Vaccine Manufacturing towards Autonomous Operation for Improvements in Speed, Scale, Robustness, Flexibility and Real-Time Release Testing. *Processes* **9** (2021).

20. Yazdan Panah, B., Roos, T., Kunze, M., Bertsch, F., Wochner, A., Rauen, M., *et al.* WO2020002598A1 (2020).

21. Crowell, L. E., Lu, A. E., Love, K. R., Stockdale, A., Timmick, S. M., Di Wu, *et al.* On-demand manufacturing of clinical-quality biopharmaceuticals. *Nature biotechnology* (2018).

22. Perez-Pinera, P., Han, N., Cleto, S., Cao, J., Purcell, O., Shah, K. A., *et al.* Synthetic biology and microbioreactor platforms for programmable production of biologics at the point-of-care. *Nature communications* **7,** 12211 (2016).

23. Pardee, K., Slomovic, S., Nguyen, P. Q., Lee, J. W., Donghia, N., Burrill, D., *et al.* Portable, On-Demand Biomolecular Manufacturing. *Cell* **167,** 248–259.e12 (2016).

24. Hotez, P. J. & Bottazzi, M. E. Developing a low-cost and accessible COVID-19 vaccine for global health. *PLOS neglected tropical diseases* **14,** e0008548 (2020).

25. Kumar, V. M., Pandi-Perumal, S. R., Trakht, I. & Thyagarajan, S. P. Strategy for COVID-19 vaccination in India: the country with thesecond highest population and number of cases. *NPJ vaccines* **6,** 60 (2021).

26. Intelligen. *SuperPro Designer* 2000. `https://www.intelligen.com/products/superpro-overview/`.

27. Henderson, J. M., Ujita, A., Hill, E., Yousif-Rosales, S., Smith, C., Ko, N., *et al.* Cap 1 Messenger RNA Synthesis with Co-transcriptional CleanCap® Analog by In Vitro Transcription. *Current protocols* **1,** e39 (2021).

28. ISSA, William, Joseph, AUNINS, John, Grant & Bancel, S. WO2014152031A1 (2014).

29. Thermo Scientific. *POROS™ Oligo (dT)25 Affinity Resin* Accessed: 2021/03/03. 2020. `https://www.thermofisher.com/order/catalog/product/A47382?de&en#/A47382?de&en`.

30. Richner, J. M., Himansu, S., Dowd, K. A., Butler, S. L., Salazar, V., Fox, J. M., *et al.* Modified mRNA Vaccines Protect against Zika Virus Infection. *Cell* **168,** 1114–1125.e10 (2017).

31. Precision NanoSystems. *NanoAssemblr GMP system* Accessed: 2021/03/16. 2020. https://www.precisionnanosystems.com/platform-technologies/product-comparison/gmp-system.

32. Blakney, A. K., McKay, P. F., Yus, B. I., Aldon, Y. & Shattock, R. J. Inside out: optimization of lipid nanoparticle formulations for exterior complexation and in vivo delivery of saRNA. *Gene Therapy* **26,** 363–372 (2019).

33. Gkionis, L., Campbell, R. A., Aojula, H., Harris, L. K. & Tirella, A. Manufacturing drug co-loaded liposomal formulations targeting breast cancer: Influence of preparative method on liposomes characteristics and in vitro toxicity. *International journal of pharmaceutics* **590,** 119926 (2020).

34. Bancel, S., William, J., John, Aunins, John Grant & Charkraborty, T. WO2014152027A1

35. Crommelin, D. J. A., Sindelar, R. D. & Meibohm, B. *Pharmaceutical Biotechnology* (Springer International Publishing, Cham, 2019).

36. Invitrogen Corporation. *Pichia Fermentation Process Guidelines* Accessed: 09/11/2021. 2002. https://tools.thermofisher.com/content/sfs/manuals/pichiaferm_prot.pdf.

37. V. Looser, B. Bruhlmann, F. Bumbak, C. Stenger, M. Costa, A. Camattari, *et al.* Cultivation strategies to enhance productivity of Pichia pastoris: A review. *Biotechnology Advances* **33,** 1177–1193 (2015).

38. Kastilan, R., Boes, A., Spiegel, H., Voepel, N., Chudobová, I., Hellwig, S., *et al.* Improvement of a fermentation process for the production of two PfAMA1-DiCo-based malaria vaccine candidates in Pichia pastoris. *Scientific Reports* **7,** 11991 (2017).

39. Lee, J., Liu, Z., Chen, W.-H., Wei, J., Kundu, R., Adhikari, R., *et al.* Process development and scale-up optimization of the SARS-CoV-2 receptor binding domain–based vaccine candidate, RBD219-N1C1. *Applied Microbiology and Biotechnology* **105,** 4153–4165 (2021).

40. Chen, W.-H., Chag, S. M., Poongavanam, M. V., Biter, A. B., Ewere, E. A., Rezende, W., *et al.* Optimization of the Production Process and Characterization of the Yeast-Expressed SARS-CoV Recombinant Receptor-Binding Domain (RBD219-N1), a SARS Vaccine Candidate. *Journal of pharmaceutical sciences* **106,** 1961–1970 (2017).

41. Government of India. *Permission to Conduct Clinical Trial of new Drug or Investigational New Drug* 2020.

42. Kitney, R. I., Bell, J. & Philp, J. Build a Sustainable Vaccines Industry with Synthetic Biology. *Trends in Biotechnology* (2021).

43. Kis, Z., Kontoravdi, C., Shattock, R. & Shah, N. Resources, Production Scales and Time Required for Producing RNA Vaccines for the Global Pandemic Demand. *Vaccines* **9** (2020).

44. Whitford, W. G., Petrich, M. A. & Flanagan, W. P. in *Single–Use Technology in Biopharmaceutical Manufacture* 169–179 (John Wiley & Sons, Ltd, 2019).

45. Shukla, A. A. & Gottschalk, U. Single-use disposable technologies for biopharmaceutical manufacturing. *Trends in Biotechnology* **31,** 147–154 (2013).

46. World Health Organization. Supplementary guidelines on good manufacturing practices for heating, ventilation and air conditioning systems for non-sterile pharmaceutical dosage forms. *WHO Technical Report Series, No. 961, 2011.*

47. Stéphane Mabic, I. K. *Production of ultrapure water for RNase-sensitive applications via ultrafiltration* 2012.

48. Teega, Psalm Baribefe and Ahiakwo, C.O. Evaluation of Electrical Power outages in Port Harcourt, Nigeria. *International Journal of Engineering and Modern Technology* **5** (2019).

49. Fahr, S., Thiel, L. & Sengoba, C. *Mobile on-Demand (MOD) mRNA Vaccine Mobile on-Demand (MOD) mRNA Vaccine Production: A Design and Optimal Location Study* 2021. `https://web.fe.up.pt/~fgm/eurecha/scp_2020/eurecha2020_mainreport_1stprize.pdf`.

50. dolomite. *Telos™ High-Throughput Systems* Accessed: 2021/12/03. 2021. `%7Bhttps://www.dolomite-microfluidics.com/microfluidic-systems/telos-high-throughput%7D`.

51. Caballero-Aguilar, L. M., Duchi, S., Quigley, A., Onofrillo, C., Di Bella, C. & Moulton, S. E. Microencapsulation of growth factors by microfluidic system. *MethodsX* **8,** 101324 (2021).

52. Pollet, J., Chen, W.-H. & Strych, U. Recombinant protein vaccines, a proven approach against coronavirus pandemics. *Advanced drug delivery reviews* **170,** 71–82 (2021).

53. Douglas, R. G., Samant, V. B., Plotkin, S. A., Orenstein, W. A., Offit, P. A. & Edwards, K. M. The Vaccine Industry. *Plotkin's Vaccines,* 41–50.e1 (2018).

54. Pfizer. *Pfizer and BioNTech Announce Positive Topline Results From Pivotal Trial of COVID-19 Vaccine in Children 5 to 11 Years* Accessed: 2021/10/26. 2021. `https://www.pfizer.com/news/press-release/press-release-detail/pfizer-and-biontech-announce-positive-topline-results`.

55. Issa, W. J., Barberio, J. L., John Grant Aunins & Noubar B. Afeyan US10590161B2 (2020).

56. Precision Nanosystems. *Personal Communication on Costs of LNP formation Device (NanoAssemblr Platform)* (ed Thiel, L.) Accessed: 2021/02/24. 2021.

57. Lemos-Pérez, G., Chávez-Valdés, S., González-Formental, H., Freyre-Corrales, G., Vázquez-Arteaga, A., Álvarez-Acevedo, B., *et al.* Elevated antibody titers in Abdala vaccinees evaluated by Elecsys® anti-SARS-CoV-2 S highly correlate with UMELISA SARS-CoV-2 ANTI RBD, ACE-2 binding inhibition and viral neutralization assays. *medRxiv* (2021).

58. Chen, W.-H., Tao, X., Agrawal, A. S., Algaissi, A., Peng, B.-H., Pollet, J., *et al.* Yeast-expressed SARS-CoV recombinant receptor-binding domain (RBD219-N1) formulated with aluminum hydroxide induces protective immunity and reduces immune enhancement. *Vaccine* **38,** 7533–7541 (2020).

59. Kis, Z., Tak, K., Ibrahim, D., Papathanasiou, M. M., Chachuat, B., Shah, N., *et al.* Pandemic-response adenoviral vector and RNA vaccine manufacturing. *medRxiv* (2021).

60. Unicef. *COVID-19 Vaccine Market Dashboard: Price* 2021. `https://www.unicef.org/supply/covid-19-vaccine-market-dashboard`.

61. Bottazzi, M. E. *Webinar: Recientes avances sobre las vacunas COVID-19. Facultad de Ciencias Químicas y Farmacia y la Dirección de Investigación Científica Humanística y Tecnológica* 2021.

62. Mvundura, M., Kien, V. D., Nga, N. T., Robertson, J., van Cuong, N., Tung, H. T., *et al.* How much does it cost to get a dose of vaccine to the service delivery location? Empirical evidence from Vietnam's Expanded Program on Immunization. *Vaccine* **32,** 834–838 (2014).

63. Lee, B. Y., Haidari, L. A., Prosser, W., Connor, D. L., Bechtel, R., Dipuve, A., *et al.* Re-designing the Mozambique vaccine supply chain to improve access to vaccines. *Vaccine* **34,** 4998–5004 (2016).

64. Haidari, L. A., Wahl, B., Brown, S. T., Privor-Dumm, L., Wallman-Stokes, C., Gorham, K., *et al.* One size does not fit all: The impact of primary vaccine container size on vaccine distribution and delivery. *Vaccine* **33,** 3242–3247 (2015).

65. Lydon, P., Raubenheimer, T., Arnot-Krüger, M. & Zaffran, M. Outsourcing vaccine logistics to the private sector: The evidence and lessons learned from the Western Cape Province in South-Africa. *Vaccine* **33,** 3429–3434 (2015).

66. World Health Organization. *Establishing Manufacturing Capabilities for Human Vaccines: Key cost drivers and factors to consider when planning the establishment of a vaccine production facility* 2017.

67. Castillo, J. C., Ahuja, A., Athey, S., Baker, A., Budish, E., Chipty, T., *et al.* Market design to accelerate COVID-19 vaccine supply. *Science (New York, N.Y.)* **371,** 1107–1109 (2021).

68. Grunwitz, C. & Kranz, L. M. in *Cancer Vaccines* (eds Savelyeva, N. & Ottensmeier, C.) 145–164 (Springer International Publishing, Cham, 2017).

69. FDA/CBER. Development and Licensure of Vaccines to Prevent COVID-19: Guidance for Industry (2020).