*Supplemental Information*

## Supplement 1: Fundamental Quality Considerations.

Process parameters are variables or conditions of the manufacturing process, typically physical or chemical conditions (e.g., temperature, pH, process time, column flow rate, reagent concentration). If the variability of a process parameter has a significant impact on a CQA, the process parameter is identified as critical (CPP), and it is monitored or controlled to ensure desired product quality. Attributes can be critical or not critical based on the risk to the patient, should the product performance fall outside the acceptable range for that attribute [1]. Based on the available information, quality attributes may be ranked to determine a "Continuum of Quality" attribute [2,3], the outcome of which may be indicative complexity of structure-function relationships and the uncertainty associated with attribute classification [4].

Product quality based on QbD is assured by performance of the selected attributes and parameters within certain boundaries termed as "Design Spaces." A design space is specific to an attribute, unit operation or process, and it can be considered as a link between CQA and CPP [5]. These relationships can be determined by applying risk assessment and design of experiments (DOE) [6,7]. In the latter approach, controlled input factors are systematically varied to determine their effects on the output responses, which allows determination of the most important input factors, identification of input factors setting leading to optimized output responses, and the elucidation of interactions between input factors [8]. Application of design requires determination unclassified parameters that do not interact and, therefore, can be excluded from the DOE. Thus, DOE screening can be used to identify interactions between process parameters. In the absence of interaction, single-variable intervals can be directly added to the design space.

Also important in product development is the Process Analytical Technology (PAT) introduced by the FDA that recommends understanding of pharmaceutical manufacturing processes to increase product quality [9]. Contrary to traditional processes that are often the result of empirical design, PAT promotes introduction of quality into the pharmaceutical manufacturing process early at the design stage of the manufacturing process, as well as on-line monitoring and controls to maintain high product quality during manufacturing operations. Thus, it is a valuable tool for the implementation of QBD as it focuses on real-time measurement and control of CQAs to achieve desired product characteristics [10]. For biological proteins, the extent of complexity multiplies the analytical quality issues, where application of PAT is deemed beneficial for bioprocess monitoring and control both upstream and downstream processes [11–14]. Complex nature of biologics may also create additional quality attributes that can affect the safety and efficacy of biology. CQAs for biological molecules may extend to include post-translational modifications that occur in the cellular environment or during the manufacturing process. Recombinant proteins are produced by living cells, which can also modify the protein structure based on their growth environment.

## Supplement 2: Nanotechnology.

As stated earlier in the paper for the combination products, the nanotechnology-based health products may also be classified as drug, device, or a combination products-based on the principal mechanism of action for achieving its intended use. The medicinal products (Drugs) assert their therapeutic action through pharmacological, immunological, and biochemical mechanism, and devices function predominantly through physical or mechanical means [15]. In combination products, both the drug and device have complementary roles. It is working of both entities in concert that assures safety, efficacy, and stability of the drug products. The FDA 2022 guidance [16] provides general recommendations for development of human drug products containing nanomaterials including drug substances, or excipients, or carriers for the drug substances. To date, the Agency has not provided a regulatory definition on nanomedicine/material. However, it describes nanomaterials as the material that has been engineered to have one or more dimension within the nanoscale range, typically to control size-dependent properties or phenomena. Normally, this range is up to 100 nm. However, FDA has not established a regulatory definition for "nanomaterial." For drug products that contain nanomaterials, if the size is engineered to exhibit specific properties or phenomena, the FDA may consider products having one or more dimensions up to 1,000 nm (or 1 micron) to be nanomaterials [17]. Though the guidance is applicable to both small molecules and biological drugs, it does not apply to biological products composed of proteins, cells, viruses, nucleic acids, or other biological materials that naturally occur at particle sizes ranging up to 1 micrometer (1000 nm), such as gene therapy or vaccine products, unless a material that has been deliberately manipulated to have

dimensions between 1–100 nm or to exhibit dimension-dependent properties or phenomena up to 1 micrometer, is also present in the product. However, it is important to exercise caution that the Agency would consider the drug product a nanomaterial even if the active occurs naturally at particle size > 1 μM if it contains excipients(s) with particle size in the nano range, or even if particles in the nanoscale range are due to conventional manufacture or storage. Further clarification about what the FDA considers as nonmaterial and the rational for its recommendation appears in its 2014 guidance [18].

Considerations for nanomaterials are further complicated by the range of nanomaterials that can be detected and their varied pharmacological properties. Since the registration of the first product, nanomedicine has progressed into various forms including dendrimers, nanocrystals, emulsions, liposomes, solid lipid nanoparticles, micelles, and polymeric nanoparticles as reported by Choi and Han [19]. A summary of consideration for evaluation of the nanomedicines by the FDA and EMA can be found in the same article.

## Supplement 3: Nebulizers.

Nebulizers convert liquid drug solutions into a fine mists suitable for inhalation into the lungs. Since their introduction in the 19th century [20,21] nebulizers have been widely used for inhaled delivery of small molecules for treatment of respiratory diseases like asthma and COPD. However, continued technological advancements have expanded their application to include the respiratory delivery of biologic therapeutics, including proteins, peptides, and nucleic acids [22,23].

Based on their aerosolization mechanisms, marketed nebulizers are broadly classified as Jet Nebulizers (JN), Ultrasonic Nebulizers (UN), and Vibrating Mesh Nebulizers (VMNs). Aerosol generation in JN, UN, and VMN devices is achieved through compressed gas, vibrating piezoelectric crystals, and vibrating mesh membrane, respectively. Ultrasonic nebulizers may induce heating of the liquid formulation during operation, which can destabilize thermolabile drugs and, therefore, are generally unsuitable for oxygen-sensitive or protein-based formulations [24]. In contrast, VMNs generate aerosols under relatively low shear conditions and without significant thermal stress, making them more suitable for preserving the structural integrity and biological activity of sensitive macromolecules of biological origin [25,26]. Nebulization can also be achieved using high-frequency acoustic waves propagating along the surface of a liquid. Surface acoustic wave (SAW) nebulizers offer advantages comparable to those of vibrating mesh nebulizers, while being less susceptible to clogging [27]. In a related approach, hybrid resonant acoustic (HYDRA) nebulizers employ a combination of surface and bulk acoustic waves, resulting in significantly higher nebulization rates than SAW systems, reduced shear stress during aerosol generation, and improved lung deposition efficiency [28,29].

The efficiency of protein drug delivery and the preservation of molecular integrity during nebulization vary considerably among different nebulizer technologies [30–34]. In addition, drug delivery from nebulizers is influenced by patient and device-related factors, including disease state, breathing patterns, aerosol output rate, and droplet size distribution [35,36].

Limited device portability has historically constrained the widespread use of nebulizers. Consequently, recent advances in nebulizer technology have focused on the development of more compact, lightweight devices incorporating smart features and built-in power sources to enable greater mobility and convenience**.** Dance-501 (Dance Biopharm, Brisbane, CA, USA) is an active vibrating mesh associated with high pulmonary deposition from 25 to 275 μL of medication upon breath actuation at inspiratory flows of 7–14 L/min [37]. Among the actively synchronized handheld nebulizers, the I-Neb® oral inhaler system (Philips Respironics, Amsterdam, Netherlands) represents a platform integrating a vibrating mesh nebulizer with Adaptive Aerosol Delivery (AAD®) technology [38,39]. AAD® synchronizes aerosol generation with the patient's breathing pattern, pulsing aerosol only during active inhalation. The system operates in both Tidal Breathing and Target Inhalation modes, incorporates patient feedback and adherence monitoring, and minimizes fugitive aerosol emissions, thereby enhancing dosing precision and reproducibility [40]. Other actively synchronized handheld nebulizer systems includeAERx (Aradigm Corporation [41,42], Hayward, CA, USA). Aerodose® (Aerogen Limited, Galway, Ireland) [43], BREELIB® and advance version of the FOX® smart nebulizer (Vectura Group(Chalfont St Giles, UK)/Phillips Medsize(Hudson, WI, USA)) for delivery of iloprost [44], as well as the Tyvaso® Optineb® [45], a pulsed ultrasonic nebulizer used for treprostinil delivery in the treatment of pulmonary arterial hypertension. The device manufactured by NEBU-TEC med. Produkt E. K. GmbH (Eisenfeld, Germany) enables dose administration using a predefined number of breaths delivered at fixed intervals. In addition, it provides both auditory and visual feedback on inhalation technique and indicates the remaining number of breaths required to complete treatment, thereby supporting correct use and adherence.

Furthermore, development of breath-actuated nebulizers (BANs) represents a significant advancement in aerosol drug delivery that synchronizes medication release with the patient's inspiratory phase. By generating aerosol exclusively during inhalation, BANs increase the fraction of drug effectively inhaled by the patient and substantially reduce medication loss during expiration [46]. Devices such as the AeroEclipse® not only improve delivery efficiency but also reduce environmental contamination and inadvertent exposure of caregivers to aerosolized medication [47]. Clinical studies suggest that breath-actuated nebulizers may shorten treatment time and improve patient satisfaction compared with standard nebulizers, with some evidence of fewer adverse events and more efficient delivery [48].

The application of BAN technology to the pulmonary delivery of biologic therapeutics is particularly promising, as it can substantially reduce the waste of costly biologic formulations associated with continuously operating nebulizers. Certain next-generation BAN systems that employ breath-actuated vibrating mesh nebulizers that generate low-shear aerosols might help preserve the structural integrity and biological activity of sensitive proteins, antibodies, and other biologics during aerosolization. For example, smart BAN platforms such as AdheResp® (HCmed Innovations, Taiwan) are designed with features that support the delivery of temperature-sensitive biologics by minimizing heat exposure during aerosol generation [49,50].

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