Human Gene Therapy Products Incorporating Human Genome Editing

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Draft Guidance for Industry

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U.S. Department of Health and Human Services
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I. INTRODUCTION

In this guidance, we, FDA, are providing recommendations to sponsors developing human gene therapy[1] products incorporating genome editing (GE) of human somatic cells. Specifically, this guidance provides recommendations regarding information that should be provided in an Investigational New Drug (IND) application in order to assess the safety and quality of the investigational GE product, as required in Title 21 of the Code of Federal Regulations 312.23 (21 CFR 312.23). This includes information on product design, product manufacturing, product testing, preclinical safety assessment, and clinical trial design.

The contents of this document do not have the force and effect of law and are not meant to bind the public in any way, unless specifically incorporated into a contract. This document is intended only to provide clarity to the public regarding existing requirements under the law. FDA guidance documents, including this guidance, should be viewed only as recommendations, unless specific regulatory or statutory requirements are cited. The use of the word *should* in FDA guidances means that something is suggested or recommended, but not required.

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[1] Human gene therapy seeks to modify or manipulate the expression of a gene or to alter the biological properties of living cells for therapeutic use. FDA generally considers human gene therapy products to include all products that mediate their effects by transcription or translation of transferred genetic material, or by specifically altering host (human) genetic sequences. Some examples of gene therapy products include nucleic acids, genetically modified microorganisms (e.g., viruses, bacteria, fungi), engineered site-specific nucleases used for human genome editing, and ex vivo genetically modified human cells. Gene therapy products meet the definition of "biological product" in section 351(i) of the Public Health Service (PHS) Act (42 U.S.C. 262(i)) when such products are applicable to the prevention, treatment, or cure of a disease or condition of human beings. (see Federal Register Notice: Application of Current Statutory Authorities to Human Somatic Cell Therapy Products and Gene Therapy Products (58 FR 53248, October 14, 1993), https://www.fda.gov/media/76647/download).

II. BACKGROUND

Over the past 10 years, the level of interest in human GE as a scientific technology used in the treatment of human disease has increased substantially, and there has been rapid development of gene therapy products incorporating GE. While the potential of such products for the treatment of human disease is clear, the potential risks are not as well understood. To assist in the translation of these products from the bench to clinical trials, this guidance includes recommendations for how to assess the safety and quality of these products and address the potential risks of these products.

For the purpose of this guidance, human GE is a process by which DNA sequences are added, deleted, altered or replaced at specified location(s) in the genome of human somatic cells, ex vivo or in vivo, using nuclease-dependent or nuclease-independent GE technologies. Human gene therapy products incorporating GE are referred to as human GE products throughout this guidance.

FDA evaluates human GE products using a science-based approach weighing the benefits and risks of each product. The benefit-risk profile for each product depends on the proposed indication and patient population, the extent and duration of therapeutic benefit achieved, and the availability of alternative therapeutic options. Some of the specific risks associated with GE approaches include off-target editing, unintended consequences of on- and off-target editing, and the unknown long term effects of on- and off-target editing.

Human GE is a rapidly evolving field and this guidance encompasses FDA's current thinking regarding the development of human GE products for clinical studies and licensure. As the field evolves, product design advances, and we gain information on the safety of human GE products, we may revise our recommendations to take into account such changes.

III. CONSIDERATIONS FOR PRODUCT DEVELOPMENT

A. General Considerations

A GE technology may be composed of a single or multiple GE component(s). These GE components may include the nuclease, DNA targeting elements (i.e., elements used to dictate the target DNA sequence, such as guide RNA) and a donor DNA template (i.e., DNA sequence provided to repair the target sequence), if applicable. When developing a human GE product, we recommend that sponsors consider: 1) the method by which the DNA sequence change will be achieved; 2) the type of genomic modification needed for the desired therapeutic effect; and, 3) the delivery method of the human GE components.

1. Genome Editing methods

GE can be achieved by either nuclease-dependent or nuclease independent methods. Nuclease-dependent GE technologies introduce site-specific breaks in

the DNA, which may result in modification of the DNA sequence at the cleavage site. Some examples of nuclease-dependent GE technologies are zinc finger nucleases (ZFNs), transcription activator-like effector nucleases (TALENs), modified-homing endonucleases (meganucleases), and clustered regularly interspaced short palindromic repeat (CRISPR)-associated (Cas) nucleases. Nuclease-independent GE technologies can change a DNA sequence without cleaving the DNA. Examples of nuclease-independent GE technologies include, but are not limited to, base editing and synthetic triplex-forming peptide nucleic acids. When choosing a specific GE technology, consideration should be given to the mechanism of action (MOA), the ability to specifically target the desired DNA sequence, and the ability to optimize the GE components to improve efficiency, specificity, or stability.

2. Type and degree of genomic modification

The type of genomic modification needed for the desired therapeutic effect is another important consideration. Many GE approaches rely on intrinsic DNA damage repair pathways to perform genomic modification. Two commonly utilized DNA damage repair pathways are homology directed repair (HDR) and non-homologous end-joining (NHEJ). HDR utilizes a homologous DNA sequence to repair the DNA break. NHEJ repairs the DNA break by rejoining two ends of cleaved DNA without a homologous repair template. Both HDR and NHEJ can be used to therapeutically modify the genome (Ref. 1). However, it is important to note that NHEJ is relatively independent of the cell cycle, while HDR is most active during S/G2 phase. It is also important to keep in mind that, although these processes can be accurate, they can also result in unintended DNA insertions or deletions (indels) with possible unanticipated consequences.

We recommend considering the degree of genome modification needed for the desired therapeutic effect (i.e., therapeutic modification threshold) when developing a therapeutic product incorporating human GE. The degree of modification needed for the desired therapeutic effect will depend on the indication and the intended patient population. For some conditions, clinical data may be available to support a given therapeutic modification threshold. The potential efficacy of a human GE product will depend on its ability to achieve this therapeutic modification threshold. If clinical data supporting a therapeutic modification threshold are not available, we recommend sponsors provide a justification for the potential efficacy of the achievable modification threshold.

3. Genome Editing Component Delivery Method

When determining the optimal delivery method of the GE components, it is important to consider the advantages and limitations of each potential method (e.g., the amount of nucleic acid the delivery vector can contain, efficiency of targeted delivery, and GE component persistence and stability). With regard to

123 persistence of the GE components, the longer the persistence of certain GE
124 components (e.g., the nuclease), the greater the risk of unintended genomic
125 modifications, specifically off-target editing and chromosomal rearrangements.
126 Therefore, to limit the degree of potential off-target editing, the duration of GE
127 component persistence should be minimized to the time needed to perform the
128 desired genomic modification, to the extent possible.
129
130 The optimal method for delivering the GE component(s) may depend on whether
131 the product involves ex vivo or in vivo genomic modification. Ex vivo
132 modifications are introduced into cells while the cells are outside the body. The
133 modified cells are then administered to the patient. In vivo modifications result
134 from administration of the GE components in their final formulation to the
135 patient. Sponsors should consider whether in vivo or ex vivo genomic
136 modification is best suited to their target indication and patient population.
137
138 For ex vivo genome modification, the cell type of interest may be amenable to
139 electroporation or mechanical methods, in which case the GE components may be
140 delivered as DNA, RNA, protein or ribonucleoprotein complexes (RNPs) for
141 CRISPR/Cas9. If HDR is the repair pathway being used, the donor DNA
142 template can be supplied as a plasmid, or using a viral vector, such as adeno-
143 associated virus (AAV). The chosen method of delivery may depend on the
144 ability of the cell type of interest to be efficiently electroporated or transduced by
145 a vector and maintain acceptable levels of viability following electroporation or
146 transduction.
147
148 For in vivo genome modification, GE components may be delivered by viral
149 vectors or nanoparticles. When choosing an in vivo delivery method, it is
150 important to consider the ability of the delivery vector to target the cells/tissue of
151 interest and minimize distribution to non-targeted tissue. Consideration should
152 also be given to the ability to control expression of vector delivered GE
153 components (e.g., using tissue-specific promoters, small molecule inhibitors), if
154 appropriate. Viral vectors may support sustained expression of GE component
155 transgenes, and nanoparticles may allow the temporal delivery of GE components
156 as messenger RNAs or proteins. The potential for vector-mediated toxicity as
157 well as pre-existing immunity to the GE component and vector should also be
158 considered. The sponsor should select the appropriate delivery method based on
159 the intended use.
160
161 **B. Chemistry, Manufacturing and Controls (CMC) Recommendations**
162
163 The general CMC considerations for product manufacturing, testing and release of human
164 GE products are the same as those previously described (Ref. 2). Additional
165 recommendations specific to human GE products regarding design, manufacture and
166 testing of the GE components, as well as the drug product (DP), are described below.
167

168 1. Genome Editing Component Design
169
170 Many platforms exist to design GE components, particularly the targeting
171 elements. We recommend sponsors utilize design platforms that are most
172 applicable to their genomic target and the type of intended genomic modification.
173 A description of, and rationale for, the design and screening processes should be
174 provided in the IND. The IND should also include the sequences of the GE
175 components.
176
177 We recommend sponsors optimize the GE components to reduce the potential for
178 off-target genome modification, to the extent possible. Optimization can be
179 performed on the editor or the targeting elements, depending on the GE
180 technology being utilized. GE components, such as guide RNA, can also be
181 optimized to inhibit degradation. The optimization strategy should be described
182 in detail in the IND.
183
184 2. Genome Editing Component Manufacture and Testing
185
186 GE components can be administered in vivo using nanoparticles, plasmids, or
187 viral vectors, or they can be used to modify cells ex vivo. When administered in
188 vivo in the form of DNA, RNA and/or protein via nanoparticles, the GE
189 components are considered the active pharmaceutical ingredients or drug
190 substances. A GE component in its final formulation for in vivo administration is
191 generally considered a DP. For example, when the GE components are expressed
192 in vivo by directly administered plasmids or vectors, the plasmid or vector in its
193 final formulation encoding the GE component is considered the DP. If used to
194 modify cells ex vivo, GE component quality is considered critical for the
195 manufacture of the final product because without these components, the resulting
196 cell product would not have the same pharmacological activity.
197
198 Detailed descriptions of how each GE component is manufactured, purified and
199 tested must be provided in the IND (21 CFR 312.23(a)(7)). We recommend a
200 description of the manufacturing process and any in-process controls for each GE
201 component include a flow diagram(s) and a detailed narrative. We recommend
202 sponsors provide lists of the reagents used during these processes and certificates
203 of analysis. Descriptions of the following should be provided in the IND for each
204 GE component manufacturing site:
205
206 • The quality control and quality assurance programs in place;
207
208 • Procedures in place to ensure product tracking and segregation;
209
210 • Procedures in place to prevent, detect and correct deficiencies in the
211 manufacturing process; and
212

213 • Procedures for shipping of the GE component from the component
214 manufacturing site to the final product manufacturing site.
215
216 This information is needed even if the GE component is manufactured by a
217 contract manufacturer (Ref. 3) and may also be cross-referenced if it is present in
218 an existing IND or Master File (Ref. 4). For most Phase 1 clinical investigations,
219 sponsors should follow the recommendations in FDA's Guidance for Industry:
220 CGMP for Phase 1 Investigational Drugs for the manufacture of these
221 components (see 21 CFR 210.2(c); Ref. 5). However, for later Phase studies and
222 for licensure, GE components must be manufactured according to CGMP
223 standards (21 CFR Parts 210 and 211), with particular consideration for control of
224 reagent quality, manufacturing process, and analytical methods.
225
226 We recommend each GE component be tested appropriately. In addition to
227 evaluating the sterility, identity, purity and functionality of each component, as
228 applicable, additional testing, such as that for process residuals, should be
229 included, depending on the manufacturing process. Descriptions of the analytical
230 procedures utilized for GE component testing, including the sensitivity and
231 specificity of the procedures, should be included in the IND. Sponsors should
232 also outline any in-process testing performed to ensure the quality of the
233 components, as appropriate.
234
235 We also recommend GE components be assessed for stability. Outlines of
236 stability study protocols and any available stability data should be provided in the
237 IND. Stability studies should be conducted on all GE components (e.g.,
238 lyophilized and reconstituted materials, if applicable). Stability studies should
239 include stability-indicating tests assessing critical product attributes, such as
240 purity and functionality, that may be affected during storage.
241
242 3. Drug Product Manufacture and Testing
243
244 An IND should contain a detailed description of the DP manufacturing process,
245 and any in-process controls. We recommend this description include a flow
246 diagram(s) as well as a detailed narrative. We recommend lists of the reagents
247 used during manufacture and certificates of analysis be provided. Please note that
248 for DP intended to be sterile, but that cannot be terminally sterilized, sponsors
249 should provide details on measures taken to ensure aseptic processing.
250
251 An IND should also contain a detailed description of the testing plan for the DP.
252 To ensure that the DP meets acceptable limits for identity, potency/strength,
253 quality and purity as defined in 21 CFR 312.23(a)(7)(iv), the DP testing plan
254 should incorporate evaluations that address any safety concerns introduced due to
255 the manufacturing process or identified during preclinical studies. For human GE
256 products consisting of ex vivo-modified cells, this testing should include
257 determination of GE efficiency (e.g., the degree of cleavage at the on-target site)

258 and specificity (e.g., the degree of cleavage at off-target sites). The DP should
259 also be tested for sterility.
260
261 Sponsors should describe in detail the analytical procedures used for testing the
262 DP. The descriptions should include the accuracy, precision, sensitivity, and
263 specificity of the assay (as applicable), as well as any controls and, if applicable,
264 reference materials used to ensure proper assay performance.
265
266 To help ensure product safety, the DP specifications should be developed based
267 on the starting materials, manufacturing process, desired final product attributes
268 and preclinical studies. As discussed, the DP may consist of GE components
269 intended for in vivo administration or may be composed of ex vivo-modified
270 cells. In the following sections, we provide recommendations pertaining
271 specifically to each of these human GE DP types:

272 i. In vivo -administered Human Genome Editing Drug Products

273
274 If the GE components will be expressed by a plasmid or viral vector that
275 is administered to patients in vivo, the plasmid/vector in its final
276 formulation is considered the DP and thus a complete description of
277 plasmid/vector manufacturing and testing should be provided in the IND
278 (Ref. 2).
279
280 If the GE components will be administered using nanoparticles, a
281 detailed description of the nanoparticle formulation, a description of the
282 manufacture of the nanoparticle components, as well as the DP, should
283 be provided in the IND. A description of the tests performed on each
284 nanoparticle component as well as on the DP should also be provided.
285 Please note that testing should include assays to evaluate the efficiency
286 of incorporation of each GE component into the nanoparticles. Please
287 also note that certain nanoparticles used for in vivo delivery of GE
288 components may be considered a delivery device.
289
290 When establishing potency assays for in vivo human GE DPs, we
291 recommend that assays be developed to measure the ability of the GE
292 components to perform the desired molecular genetic and downstream
293 biological modifications in the target cells or tissues. We also
294 recommend inclusion of such a potency assay in the DP stability studies.
295 Additional information on the development of appropriate potency tests
296 can be found in FDA's Guidance for Industry: Potency Tests for
297 Cellular and Gene Therapy Products (Ref. 6).
298

299 ii. Ex vivo-modified Human Genome Editing Drug Products
300
301 When describing the manufacturing processes for ex vivo -modified
302 human GE DPs, descriptions of process controls and in-process testing
303 should be included for critical steps that may have significant impact on
304 the efficiency or specificity of editing (e.g., RNP formation step in the
305 case of CRISPR-mediated editing). Acceptance criteria or limits should
306 be provided and justified.
307
308 Testing of ex vivo-modified human GE DPs should include evaluation
309 of the following:
310
311 - On-target editing efficiency, including characterization of the
312 editing events occurring at the on-target site;
313
314 - Off-target editing frequency;
315
316 - Chromosomal rearrangements;
317
318 - Residual GE components; and
319
320 - Total number of genome-edited cells.
321
322 We also recommend that the number of edited cells or the frequency of
323 GE be monitored during stability testing of ex vivo-modified human GE
324 DP.
325
326 When establishing potency tests for ex vivo-modified human GE DP, we
327 recommend assays be developed that measure the properties of the cells
328 and the intended functional outcomes of the genomic modifications
329 resulting from GE. For example, we recommend that potency assays for
330 a genome-edited $CD34^+$ hematopoietic stem/progenitor cell product
331 measure both the stem/progenitor cell activity and the functional
332 outcome of the GE. In some instances, surrogate potency tests may be
333 acceptable; however, it is critical that the data provided supports a
334 correlation between the output of the surrogate potency test and the
335 functional outcome of the GE (Ref. 6).
336
337 Please note that if the ex vivo-modified human GE DP is an allogeneic
338 human cell product, where a product lot is meant to treat multiple
339 patients, additional testing and establishment of acceptance criteria may
340 be appropriate. For example, in addition to meeting the donor eligibility
341 screening and testing criteria outlined in 21 CFR Part 1271, Subpart C,
342 additional donor screening and testing may be warranted. More
343 extensive analysis of the GE events occurring at both on- and off-target

sites, additional adventitious agent testing, establishment of stringent acceptance criteria for the number of alloreactive lymphocytes and absence of aberrant growth (i.e., if the DP is an allogeneic T cell product) may also be warranted.

Additional in-process, lot release, and characterization testing may be needed for more complex products (e.g., products incorporating multiple rounds of genome editing or the creation of multiple cell banks).

IV. CONSIDERATIONS FOR PRECLINICAL STUDIES

The overall objectives of a preclinical program for an investigational human GE product are generally the same as those described for gene therapy products in FDA's Guidance for Industry: Preclinical Assessment of Investigational Cellular and Gene Therapy Products (Ref. 7) ("Preclinical Assessment Guidance"). These objectives include: 1) identification of a biologically active dose range; 2) recommendations for an initial clinical dose level, dose-escalation schedule, and dosing regimen; 3) establishment of feasibility and reasonable safety of the proposed clinical route of administration (ROA); 4) support for the target patient population; and, 5) identification of potential toxicities and physiologic parameters that help guide clinical monitoring for a particular investigational product. More details for these general considerations in preclinical studies are available in the above noted guidance (Ref. 7). The following general elements should be incorporated into the preclinical development program for an investigational GE product:

- Preclinical in vitro and in vivo proof-of-concept (POC) studies are recommended to establish feasibility and support the scientific rationale for administration of the investigational human GE product in a clinical trial.

    - The use of in vitro models should be considered for evaluating the activity of a GE product in the target cell type(s) for genomic modification.

    - The animal species and/or models selected for in vivo studies should demonstrate a biological response to the investigational GE product or species-specific surrogate product (See section IV.A of this guidance for further discussion). Given the differences in the genomic sequences between humans and animals, analysis of the biological activity may be done in a species-specific context and applied to the clinical product, as appropriate.

- We recommend preclinical safety studies be designed to identify potential risks associated with administration of the GE product. Potential toxicities may be related to the delivery modality for the GE components, expression of the GE components, modification of the genomic structure, and/or expression of the gene product.

- The safety assessment should include identification and characterization of off-target activity, chromosomal rearrangements, and their biological consequences, as feasible.

- In vivo preclinical safety studies for an investigational GE product should incorporate elements of the planned clinical trial (e.g., dose range, ROA, delivery device, dosing schedule, evaluation endpoints), to the extent feasible. Study designs should be sufficiently comprehensive to permit identification, characterization, and quantification of potential local and systemic toxicities, their onset (i.e., acute or delayed) and potential resolution, and the effect of dose level on these findings.

- We recommend biodistribution studies be conducted to characterize the distribution, persistence, and clearance of the GE product, as well as any expressed GE components in vivo. Evaluation of the biodistribution profile of the edited genetic sequence and persistence of the gene product may provide additional information on the extent of editing activity in target and non-target tissues.

Specific recommendations for the characterization of activity and safety of a GE product are as follows:

A. Product Evaluated in Preclinical Studies

- The investigational human GE product should be evaluated in the definitive POC and safety studies, when feasible.

- Due to differences in the genomic sequences between animals and humans, POC and/or safety studies may warrant the use of a surrogate GE product (e.g., substitution of the human elements including GE components, promoter(s), and transgene(s) for the respective species-specific elements in the GE product) in situations where administration of the investigational human GE product would not be informative. We recommend sponsors provide scientific justification for the administration of a surrogate GE product, and establish biological relevance of the surrogate compared to the investigational human GE product.

- For ex vivo-modified GE products, the clinical cell source should be used for the definitive preclinical studies. If an alternative cell source is used in any studies, scientific justification should be provided for the cell source selected.

- Each GE product lot evaluated in the preclinical studies should be characterized according to appropriate specifications, consistent with the stage of product development. This information will be critical to establish comparability of the product used in preclinical studies to the clinical product, if necessary.

432 ### B. Assessment of Activity
433
434 We recommend preclinical in vitro and in vivo POC studies assess the following:
435
436 - Specificity and efficiency of editing in target and non-target cells;
437
438 - Functionality of the corrected or expressed gene product (e.g., protein, RNA), if
439 applicable;
440
441 - Editing efficiency required to achieve the desired biological activity or
442 therapeutic effect;
443
444 - Durability of the genomic modification and resulting biological response; and
445
446 - Effects of genetic variation on editing activity across the target population.
447
448 ### C. Assessment of Safety
449
450 We recommend preclinical studies be conducted to identify and characterize the risk of
451 GE at on- and off-target loci and include the following:
452
453 - Identification of off-target editing activity, including the type, frequency, and
454 location of all off-target editing events.
455
456 - The use of multiple orthogonal methods (e.g., in silico, biochemical,
457 cellular-based assays) that include an unbiased genome-wide analysis is
458 recommended for identification of potential off-target sites. When
459 possible, the analysis should be performed using the target human cell
460 type(s) from multiple donors.
461
462 - Verification of bona fide off-target sites should be conducted using
463 methods with adequate sensitivity to detect low frequency events. The
464 analysis should be performed using the target human cell type(s) from
465 multiple donors.
466
467 - Appropriate controls should be included to confirm the quality of the
468 assay and to assure interpretability of the results and its suitability for the
469 intended use.
470
471 - Assessment of genomic integrity, including chromosomal rearrangements, large
472 insertions or deletions, integration of exogenous DNA, and potential
473 oncogenicity or insertional mutagenesis. For ex vivo-modified cells, this may
474 include assessment for clonal expansion and/or unregulated proliferation.
475

476 • Evaluation of the biological consequences associated with on- and off-target
477 editing, as feasible.
478
479 • Immunogenicity of the GE components and gene product expressed.
480
481 • Characterization of the kinetic profile of GE components expression and editing
482 activity.
483
484 • Assessment of viability and any selective survival advantage of the edited cells.
485
486 • Preservation of cell functionality following GE (e.g., differentiation capacity for
487 progenitor cells).
488
489 • Evaluation of the potential for inadvertent germline modification.
490
491
492 **V. CONSIDERATIONS FOR CLINICAL STUDIES**
493
494 We recommend that clinical development programs of human GE products address both the risks
495 associated with the gene therapy product itself as well as the additional risks associated with the
496 GE, including unintended consequences of on- and off-target editing, which may be unknown at
497 the time of product administration. Clinical trial design should include appropriate patient
498 selection, an efficient and safe approach to product administration (including data-based dosing,
499 dose schedule, and treatment plan), adequate safety monitoring, and an appropriate choice of
500 endpoints. Additionally, long term follow-up is recommended for clinical trial subjects receiving
501 human GE products for evaluation of clinical safety. In general, the overall study design,
502 assessment of adverse events (AEs) and subject follow-up plans should be well described in the
503 IND. The overall considerations for clinical trial design for GE products are similar to those
504 outlined for other cellular and gene therapy products (Ref. 8) and are briefly described in section
505 V.A-F of this guidance.
506
507 **A. Study Population**
508
509 Selecting the appropriate study population ensures maximum benefit, while minimizing
510 the potential risk to subjects. We recommend the choice of study population be well
511 supported based on the product MOA and study rationale, along with balancing the
512 potential risks of the product. Human GE products may have significant risks and an
513 uncertain potential for benefits. Therefore, first-in-human trials involving such products
514 generally should be designed to enroll only subjects for whom no other treatment options
515 are available or acceptable. Factors to consider in determining the study population
516 include:
517
518 • The MOA of the product in the context of a specific disease;
519
520 • The anticipated duration of therapeutic benefit;

521
522 - The availability and effectiveness of alternative therapeutic options for the
523 patient population;
524
525 - Subjects with severe or advanced disease may be more willing to accept the
526 risks of an investigational human GE product. However, these subjects may
527 be predisposed to experiencing more AEs or be receiving concomitant
528 treatments, which could make the safety or effectiveness data difficult to
529 interpret. Therefore, in some instances, subjects with less advanced or more
530 moderate disease may be appropriate for inclusion in first-in-human clinical
531 studies.
532
533 **B. Dose and Dose Schedules**
534
535 Adopting well established, safe, and effective product delivery methods is important for
536 minimizing any potential AEs related to product delivery to target tissues. Both the
537 delivery and the proposed dose schedules should be supported by comprehensive
538 preclinical data and, where available, guided by previous clinical experience from similar
539 products, including cellular or gene therapy products that may or may not have been
540 genome edited. Additional aspects of dose and regimen for clinical trials evaluating
541 human GE products are similar to those for other cellular and gene therapy products and
542 can be found in section IV.D of FDA's Considerations for the Design of Early-Phase
543 Clinical Trials of Cellular and Gene Therapy Products; Guidance for Industry (Ref. 8).
544
545 **C. Treatment Plan**
546
547 We recommend that any risk(s) anticipated in association with the GE product be
548 mitigated by staggered subject enrollment, with a specified time interval between product
549 administration to sequential subjects within and between cohorts. The staggering interval
550 should be of sufficient duration to monitor for acute and subacute AEs prior to treating
551 additional subjects at the same dose, or prior to increasing the dose in subjects treated
552 subsequently. The staggering interval should also take into account the expected duration
553 of activity of the human GE product.
554
555 Selection of study cohort size depends on the size of the proposed patient population and
556 the amount of acceptable risk in that study population for the GE product. In addition,
557 other considerations, such as assessments of tolerability, feasibility, and pharmacologic
558 activity may influence choice of cohort size. Additional cohort size considerations are
559 outlined in section IV.E.2 of FDA's Considerations for the Design of Early-Phase
560 Clinical Trials of Cellular and Gene Therapy Products; Guidance for Industry (Ref. 8).

561
562 **D. Monitoring and Follow-Up**
563
564 1. Assessment of Product-Related Adverse Events
565
566 A thorough safety monitoring strategy, with a well-defined toxicity grading
567 system, and a toxicity management plan is crucial for clinical trials evaluating
568 human GE products. Specific consideration should be given for adequate
569 monitoring of any off-target editing and adequate assessment of the outcomes of
570 unintended consequences of on- and off-target editing. Additional monitoring
571 should capture AEs related to aberrant cellular proliferation, immunogenicity, and
572 tumorigenicity. Such AEs should be anticipated from pre-clinical studies, if
573 possible, and toxicity grading and management strategy should be outlined in the
574 clinical protocol.
575
576 Applicable reporting requirements outlined in 21 CFR 312.32 for adverse
577 experiences associated with the use of the human GE product must be followed.
578 Additional information concerning good clinical practice can be found in FDA's
579 E6(R2) Good Clinical Practice: Integrated Addendum to ICH E6(R1); Guidance
580 for Industry (Ref. 9).
581
582 2. Long Term Follow-Up
583
584 Prior to enrolling, subjects should be asked to provide voluntary, informed
585 consent to long term follow-up (LTFU). As discussed, the long term effects of
586 intended, as well as unintended, editing at on- and off-target loci may be unknown
587 at the time of GE product administration. Therefore, we recommend that
588 sponsors conduct LTFU at least 15 years after product administration, as outlined
589 in FDA's Long Term Follow-Up After Administration of Human Gene Therapy;
590 Guidance for Industry (Ref. 10).
591
592 **E. Study Endpoints**
593
594 We recommend that study endpoints be based on the proposed indication. For efficacy
595 studies, the primary endpoint should also reflect a clinically meaningful effect of the GE
596 product. The experience gained from early-phase clinical studies can help guide the
597 selection of a primary endpoint for late-phase studies. Further information may be
598 obtained from FDA's Guidance for Industry: Providing Clinical Evidence of
599 Effectiveness for Human Drugs and Biological Products (Ref. 11).
600
601 **F. Special Considerations for Research Involving Children**
602
603 When possible, clinical studies should enroll individuals who can understand and consent
604 to the study procedures and risks. For clinical investigations involving children,
605 associated with greater than minimal risk, a reviewing Institutional Review Board must

606 find, among other things, that these risks are justified by the anticipated direct clinical
607 benefit to the children (21 CFR 50.52). Such prospect of direct benefit should be
608 evidence-based (e.g., from adult humans or appropriate animal models). Therefore, it is
609 important to enroll at least an initial cohort of adult subjects, whenever feasible, to obtain
610 preliminary data on safety and feasibility, bioactivity, and preliminary efficacy to support
611 enrollment of pediatric subjects. If enrollment of pediatric subjects is justified, then an
612 effort should be made to enroll adolescents prior to enrollment of younger children and
613 infants, as appropriate for the specific disease of interest.
614
615
616
VI. COMMUNICATION WITH FDA

617
618 We recommend sponsors of human GE products communicate with the Office of Tissues and
619 Advanced Therapies (OTAT) in the Center for Biologics Evaluation and Research (CBER) early
620 in product development, before submission of an IND, to discuss the product-specific
621 considerations for transitioning these products to the clinical phase of product development.
622 There are different meeting types that can be used for such discussions, depending on the stage
623 of product development and the issues to be considered. These include pre-IND meetings prior
624 to submission of the IND (Ref. 12), and INitial Targeted Engagement for Regulatory Advice on
625 CBER producTs (INTERACT) meetings, which can be used earlier in development to discuss
626 issues such as preclinical development or manufacturing, so that sponsors can obtain non-
627 binding regulatory advice.[2]
628
629

[2] For additional information about INTERACT meetings, please see https://www.fda.gov/vaccines-blood-biologics/industry-biologics/interact-meetings.

630 **VII. REFERENCES**
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670
671 * When finalized, this guidance will represent FDA's current thinking on this topic.

672
APPENDIX
673
Abbreviations and Acronyms
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Term	Description
AAV	Adeno-Associated Virus
AE	Adverse Event
Cas	CRISPR-associated
CBER	Center for Biologics Evaluation and Research
CGMP	Current Good Manufacturing Practice
CMC	Chemistry, Manufacturing and Controls
CRISPR	Clustered Regularly Interspaced Short Palindromic Repeat
DNA	Deoxyribonucleic Acid
DP	Drug Product
FDA	Food and Drug Administration
GE	Genome Editing
HDR	Homology Directed Repair
ICH	International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use
IND	Investigational New Drug
Indels	Insertions or Deletions
INTERACT	INitial Targeted Engagement for Regulatory Advice on CBER producTs
LTFU	Long Term Follow-Up
MOA	Mechanism of Action
NHEJ	Non-Homologous End-Joining
OTAT	Office of Tissues and Advanced Therapies
PHS	Public Health Service
POC	Proof-of-Concept
RNA	Ribonucleic Acid
RNP	Ribonucleoprotein Complex
ROA	Route of Administration
TALEN	Transcription Activator-Like Effector Nuclease
ZFN	Zinc Finger Nuclease

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