Ethical Viewpoint Paper

Knowing The Nano: A look at current international laws that provide guidance on assessing the toxicity of nanomaterials for Crispr-based delivery of Human Genome Editing

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Crispr, short for Clustered Regularly Interspaced Short Palindromic Repeats-associated protein 9, is a revolutionary gene editing technology that has proven to be the solution for eradicating fatal diseases by altering the human genome. As used in the genome of human, it is referred to as human genome editing, and involves alterations such as cutting out a defective gene mutation and replacing it with a corrected one, using Crispr. Although off targets, side effects and difficulties using a physical-viral based drug delivery method have been reported, there has also been a monumental development in terms of eradicating fatal diseases, using Crispr, such as Sickle Cell Disease [1], Leukemia [2] and HIV [3].
Human genome editing using Crispr has also faced many challenges such as off-targets (not reaching target genes or reaching targeted and untargeted genes), and mosaicism (the presence of more than one genotype caused by edits [4].
Arguably, however, the most challenging issue for Crispr is in its physical-viral delivery method, i.e., how to safely and efficiently deliver the components that are needed to use Crispr, which consists of a DNA cutting enzyme called Cas/9, and a short RNA that guides the enzyme to a target gene for edits.
In Part 1 of this paper, an exploration of how researchers have turned to the use of non-viral delivery method using nanomaterials to combat this challenge will be explored.
In Part 2 of this paper, a look at international laws on nanotoxicity that regulate the use of nanomaterials for human use will be explored.

Part 1: Viral delivery challenges for Crispr in the treatment of cancer treatment
Crispr has two components, Cas/9, and the guiding RNA. To achieve safe, and efficient delivery of treatment of diseases such as cancer, these two components must reach the targeted genes by passing through different physical barriers using viruses. According to researchers, a viral- based delivery technique can have complications for the patients, such as patients developing antibodies to the viruses or having pre-existing antibodies to the viruses. Viral vectors can also prompt side effects, off-target responses, and be costly.

Physical delivery challenges
Physical delivery method used in Crispr include micro-injection, electroporation, and hydrodynamic delivery, all of which, while found to be highly efficient, are also found to be beneficial if used in-vitro but not in in-vivo, a shortfall that cannot be ignored, since current laws allow for gene editing in humans but not for germline editing in human embryos [5].
Faced with these challenges, researchers have turned to the use of non-viral delivery method using nanomaterials. Also called non-viral vectors, nanocarriers such as polymers, lipids, porous silicon, mesoporous silica nanoparticles, metal-organic frameworks have been used to achieve safe and efficient delivery method using Crispr. These nanomaterials are being used in Crispr due

to their unique properties. Nanomaterials have low immunogenicity, high biocompatibility, and because of their nano size, considered ideal for delivery of the Crispr components [6].

Nano-based, non-viral delivery technique of CRISPR may be more efficient

Nanotechnology, inner engineering at nanoscale

Simply put, Nanotechnology is engineering and fabrication of things at nanoscale. Nanoscience is the study of the phenomena of materials and their properties at nanometer scale. A nanometer is 10-9 or one-billionth of a meter. In layman terms, a nanoparticle is "up to one million times thinner than a [strand of] human hair: https://www.horiba.com/en_en/science-in-action/should-you-be-worried-about-nanotoxicity/

As such, Nanoscience is the study of materials at microscopic levels: https://www.nano.gov/nanotech-101/what/definition.

As distinguished from Nanoscience, Nanotechnology, also known as molecular manipulation, is the use of nanoscience in the design, characterization, production, and application of structures, devices, and systems at the nanoscale. Nanotechnology is about the control of matter at atomic level: https://www.science.org.au/curious/nanoscience.

Part 2: Nanotoxicity: In International Law

Newness

An essential legal question relating to guidance on how to assess the risks of nanomaterials involves whether assessment of chemicals at nanoscale should be considered new. Since many chemicals in their original form have been assessed and regulated, should they then be exempt if introduced at nanoscale and exposed to both the environment, and humans? Various schools of thought argue for and against the exemption of nanomaterials as new or not new. This paper argues that since the toxicity of materials at nanoscale can be "extremely toxic", it is crucial that laws should be enacted to mandate individual assessment of nanomaterials before they are used in the treatment of diseases in humans using Crispr.

Genotoxicity

Since most nanomaterials work by direct interaction with biomolecules, this also means that if nanomaterials are not assessed properly for toxicity, they would be administered directly into biomolecules that are essential for normal gene function and cell division: https://www.sciencedirect.com/science/article/abs/pii/S0169409X12002384?via%3Dihub

Tracing

Some nanomaterials are not capable of being traced after administration which makes it ultra-dangerous for human use.

Precise Interactions

In addition, the precise interactions of these nanomaterials are simply unknown.

Intravenous clinical applications

Many scientific studies have proven the negative impact of intravenous administration of nanomaterials, which constitutes the accumulation of nanomaterials in the liver, and subsequent translocation to essential areas in the human body such as the central nervous, cardiovascular, and renal systems.

While the potential efficacy of nanomaterials as used in the treatment of diseases such as cancer in humans using Crispr should be welcomed, their potential toxicity to harm humans should not be ignored. Sadly, the complexity of assessing nanomaterials means that there is currently a lack of proper guidance in terms of regulations surrounding nanomaterials [7].

Despite this, there are number of international organizations that have put forth efforts to provide guidance in this area.

UNITED NATIONS

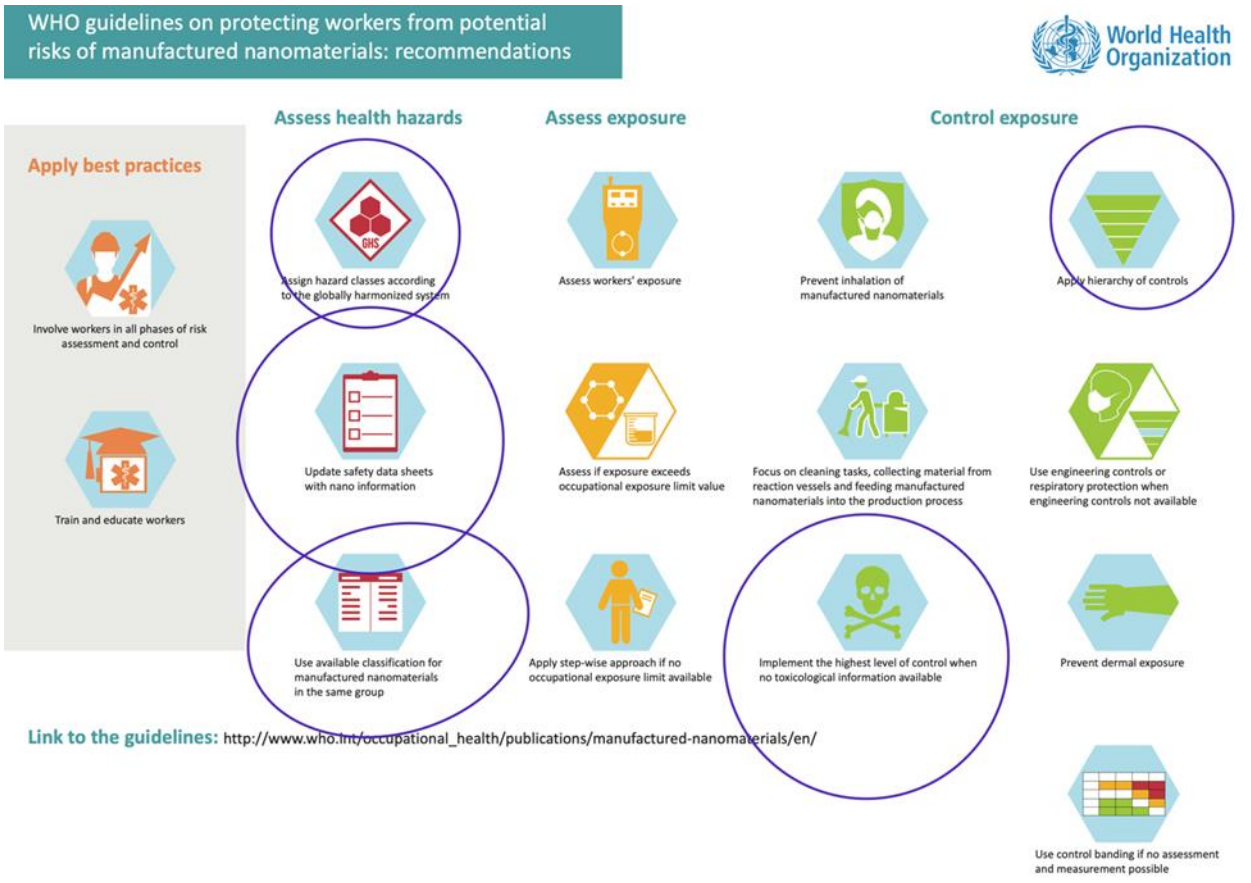
United Nations Institute for Training and Research (UNITAR)

UNITAR [8] has implemented international efforts to support developing countries in nano-safety. Since 2009, and principally funded by the government of Switzerland, UNITAR has been working with the Inter-Organization Program for the Sound Management of Chemicals (IOMC), the Strategic Approach to International Chemicals Management (SAICM), and the Organization for Economic Co-Operation and Development (OECD) to raise awareness of the risks involved with the use of nanomaterials in developing countries [9].

World Health Organization (WHO)

WHO has also implemented guidelines/recommendations to protect workers from potential risks of exposure to manufactured nanomaterials [10].

While this is not directly relevant for this paper as the discussion here is about the toxicity of nanomaterials (as opposed to Occupational Safety) as used in the safe and efficient treatment of diseases such as cancer in humans using Crispr, some of WHO's guidelines merit our attention.



The circled guidelines, such as "the highest level of control when no toxicological information is available" or "Update safety data sheets with nano information," should be followed in assessing the risks of toxicity involved in using nanomaterials.

European Union Observatory for Nanomaterials (EUON) [11]

In Europe, EUON discussed the two European Union (EU) regulations that merit attention, namely, the Registration, Evaluation, Authorization and Restriction of Chemicals regulation (REACH No. 1907/2006), and the "Classification, Labelling, and Packaging" regulation (CLP No. 1272/2008) to assess the toxicity of nanomaterials: https://euon.echa.europa.eu/regulation

Under REACH, to be legally manufactured or imported in the EU, all hazardous nanomaterials must be registered. The registration must include information on the hazardous nanoforms, their impact on humans and the environment, and their exposure throughout the lifecycle.
Under CLP, the same hazardous nanoforms must not only be notified to The European Chemicals Agency (ECHA), but appropriately and clearly classified, labelled, and packaged to reflect the "possible hazards" of nanomaterials.

The rule of transparency in assessing the potential risk
In addition to the combined efforts of the organizations and the regulations, Europe also demands that companies that wish to use certain nanomaterials for commercial or scientific purposes should do the following:

- Clearly inform the above organizations on the safety of nanomaterials
- Measures needed to assess and control the potential risk
- Follow ECHA guidance on the identification and properties of nanomaterials

Knowing The Nano: A hope for more public debate in assessing nanotoxicity
In conclusion, while human Genome Editing, using Crispr has had a monumental impact on humanity, we cannot ignore the fact that just as with any new scientific innovations, Crispr as a human genome editing tool has met with challenges such as off-targets, side effects, and difficulties using a physical-viral based drug delivery technique.
While creating a safe and efficient delivery method using nanomaterials is currently being tested by Crispr researchers, they must not ignore the potential toxicity of nanomaterials as an ethical and legal challenge. In fact, creating a safe and efficient delivery method must mean that human lives must never ever be compromised in exchange for scientific innovations.
While there is currently a lack of proper uniform guidance in terms of ethics, and law surrounding nanomaterials, the four principles in bioethics, and the guidance from several international organizations should be utilized in furthering the public debate in this area.
Therefore, it is the hope of this paper that such public debates will shine light on the toxicity, as opposed to the efficacy of using nanomaterials, in the safe and efficient treatment of diseases such as cancer in humans using Crispr

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