Available online on 15.10.2021 at http://jddtonline.info

Journal of Drug Delivery and Therapeutics

Open Access to Pharmaceutical and Medical Research

Open Access Full Text Article

Review Article

4D Printing: The Dawn of "Smart" Drug Delivery Systems and Biomedical Applications

Khan Ahmar[1]*, Iqbal Mir Javid[2], Amin Saima[3], Bilal Humaira[4], Bilquees[5], Noor Aneeza[6], Mir Bushra[7], Kaur Mahak Deep[8]

1. Department of Pharmaceutics, School of Pharmaceutical Education and Research, Jamia Hamdard, Delhi, India
2. Department of Clinical Pharmacy, King Khalid University, Aseer, Abha, Kingdom of Saudi Arabia
3. Department of Pharmaceutics, School of Pharmaceutical Education and Research, Jamia Hamdard, Delhi, India
4. Department of Pharmaceutical Sciences, University of Kashmir, Srinagar - 190006, Jammu and Kashmir, India
5. Department of Pharmaceutical Sciences, University of Kashmir, Srinagar - 190006, Jammu and Kashmir, India
6. Department of Pharmaceutical Sciences, University of Kashmir, Srinagar - 190006, Jammu and Kashmir, India
7. Department of Pharmaceutical Sciences, University of Kashmir, Srinagar - 190006, Jammu and Kashmir, India
8. Department of Pharmaceutics, Chandigarh College of Pharmacy, Landran, Mohali Punjab, India

Article Info:

Article History:

Received 08 August 2021
Reviewed 19 September 2021
Accepted 26 September 2021
Published 15 October 2021

Cite this article as:

Khan A, Iqbal MJ, Amin S, Bilal H, Bilquees, Noor A, Mir B, Kaur MD, 4D Printing: The Dawn of "Smart" Drug Delivery Systems and Biomedical Applications, Journal of Drug Delivery and Therapeutics. 2021; 11(5-S):131-137

DOI: http://dx.doi.org/10.22270/jddt.v11i5-S.5068

*Address for Correspondence:

Mr. Ahmar Khan, PhD Scholar, Department of Pharmaceutics, School of Pharmaceutical Education and Research, Jamia Hamdard, Delhi, India.

Abstract

With the approval of first 3D printed drug "spritam" by USFDA, 3D printing is gaining acceptance in healthcare, engineering and other aspects of life. Taking 3D printing towards the next step gives birth to what is referred to as "4D printing". The full credit behind the unveiling of 4D printing technology in front of the world goes to Massachusetts Institute of Technology (MIT), who revealed "time" in this technology as the fourth dimension. 4D printing is a renovation of 3D printing wherein special materials (referred to as smart materials) are incorporated which change their morphology post printing in response to a stimulus. Depending upon the applicability of this technology, there may be a variety of stimuli, most common among them being pH, water, heat, wind and other forms of energy. The upper hand of 4D printing over 3D printing is that 3D printed structures are generally immobile, rigid and inanimate whereas 4D printed structures are flexible, mobile and able to interact with the surrounding environment based on the stimulus. This capability of 4D printing to transform 3D structures into smart structures in response to various stimuli promises a great potential for biomedical and bioengineering applications. The potential of 4D printing in developing pre-programmed biomaterials that can undergo transformations lays new foundations for enabling smart pharmacology, personalized medicine, and smart drug delivery, all of which can help in combating diseases in a smarter way. Hence, the theme of this paper is about the potential of 4D printing in creating smart drug delivery, smart pharmacology, targeted drug delivery and better patient compliance. The paper highlights the recent advancements of 4D printing in healthcare sector and ways by which 4D printing is doing wonders in creating smart drug delivery and tailored medicine. The major constraints in the approach have also been highlighted.

Keywords: 4D printing, smart, drug delivery system, patient compliance, biomaterials, tailored medicine

1. INTRODUCTION:

Innovation is never-ending. With the beginning of the 3D printing era in medicine and drug delivery, 4D printing has become a reality. Based on the foundation of 3D printing, 4D printing includes a fourth dimension; that is, time. Products designed through 4D printing can often change their configuration over time in response to external or internal stimuli (such as heat, pH, temperature or even water). Conversion into this 'fourth dimension' can be a result of the smart materials (feedstock) and the predetermined design in which the products are fabricated (known as smart design).[1-5]

2. BACKGROUND AND EMERGENCE OF 4D PRINTING:

In 1984, 3D printing technology was introduced by Charles (Chuck) Hull of 3D Systems Corporation in a process known as stereolithography (SLA) which employs materials, designs, and 3D printers to create 3D structures by successive layering of the materials based on a predetermined design[6]. The official term "3D printing" (formerly referring to a powder bed adhesive jetting technology) was introduced later at MIT, which developed at MIT. In 2012, 4D printing was revealed to the world by the Massachusetts Institute of Technology (MIT) at the TED Conference where Skylar Tibbits demonstrated how a static printed object transformed over time[7]. This marked the start

of the 4D printing concept, where the fourth dimension is time, highlighting that the printed structures are no longer static; rather, they are programmable and can transform independently, alter their morphology when exposed to various stimuli like light, temperature, water, and pH. 4D printing is predictable and targets shape, property, and functionality evolution which allows for self-assembly, multi-functionality, and self-repair. Since then, 4D printing has become an advanced branch of 3D printing that is gaining substantial attention from scientists and engineers of different disciplines.[8]

3. HOW 4D PRINTING WORKS?

4D printing requires the integration of a time component into the 3D printed structures. This makes the designing process more important. Furthermore, 4D structures need to be programmed prior printing to make the time-dependent transformation mechanism of smart materials possible[9]. Fig. 1 depicts the working of a 4D printer[10].

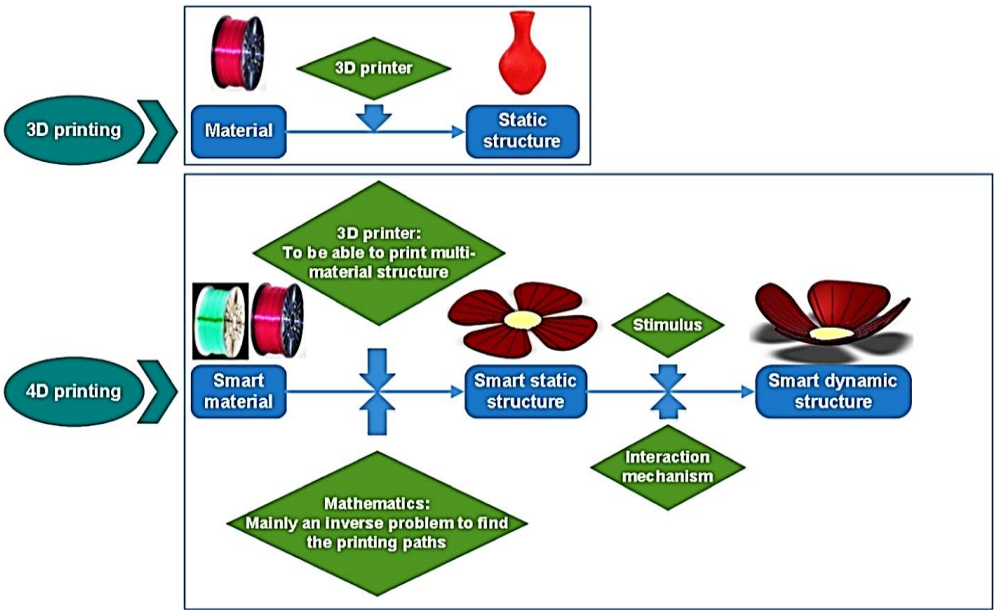

Figure 1: Working of a 4D printer.

4. SMART MATERIALS USED IN 4D PRINTING:

The smart materials used in 4D printing for drug delivery and medical device applications are appended in Table 1.

Material	Description	Stimulus	Examples	Ref
Shape Memory Alloys (SMAs)	SMAs are usually subjected to a programming process between two transformation phases of metal alloys. These phases depend on differences in temperature or magnetic field.	Thermo-responsive	TiNi (Titanium Nickle) alloy	11
Shape Memory Ceramics (SMCrs)	SMCrs exhibit either superelasticity with the ability to deform to large strains and recover, or exhibit the shape memory effect where they can transform between states that have been predefined with the aid of an external stimulus. These crack at low strains and after only few cyclic strains are applied.	Thermo-responsive	Superelastic zirconia ceramics	12,13
Shape Memory Polymers (SMPs)	SMPs withstand large amounts of deformation strain over a range of temperatures, for radical and impressive transformation versatile processing and possess abilities required for a range of AM technologies.	Thermo-responsive	Inks: Methacrylate-based monomers, photoinitiator Phenylbis (2,4,6-trimethyl benzoyl) phosphine oxide (BAPO), Sudan I (0.05 wt%) and Rhodamine B (1 wt%)	11
		UV radiation	Self-healing SMP (semicrystalline linear polymer. Polycaprolactone (PCL) incorporated into a methacrylate-based SMP system)	11
Stimuli-responsive hydrogels	Hydrogels are used to fabricate structures in which the diffusion of water into the polymer network manipulates the product morphology by causing the structure to swell up.	Water		7

5. 4D PRINTING AND SMART DRUG DELIVERY SYSTEMS:

Owing to its novelty, the use of 4D printing in developing drug delivery systems is yet to be explored fully. As of now, there are two major mechanisms by which 4D printing can be used in fabricating smart drug delivery systems viz bio-adhesion and encapsulation.[5]

5.1. Bio-Adhesion:

5.1.1. Smart Microneedles with backward facing barbs:[14]

A major constraint in controlled long-term drug delivery or biosensing using microneedles is its low tissue adhesion. Inspired by natural microscopic structures with high tissue adhesion found in living creatures like microhooks of parasites, barbed stingers of honeybees and quills of porcupines, Daehoon *et al* fabricated microneedles with bioinspired backward-facing curved barbs for enhanced tissue adhesion using 4D printing. The microneedles with horizontal barbs were fabricated using a 3D printer. Post-printing, upon exposure to UV light as stimulus, these horizontal barbs containing resin undergo bending and curvature due to desolvation-induced deformation utilizing cross-linking density gradient in a photocurable polymer and change into backward facing barbs that result in enhanced tissue adhesion. These backward facing barbed micro needles have demonstrated 18 times more stronger tissue adhesion than the barbless microneedles. Fig. 2 depicts the schematic illustration of 4D printing approach to program deformation of horizontally printed barbs into a backward-facing shape[14].

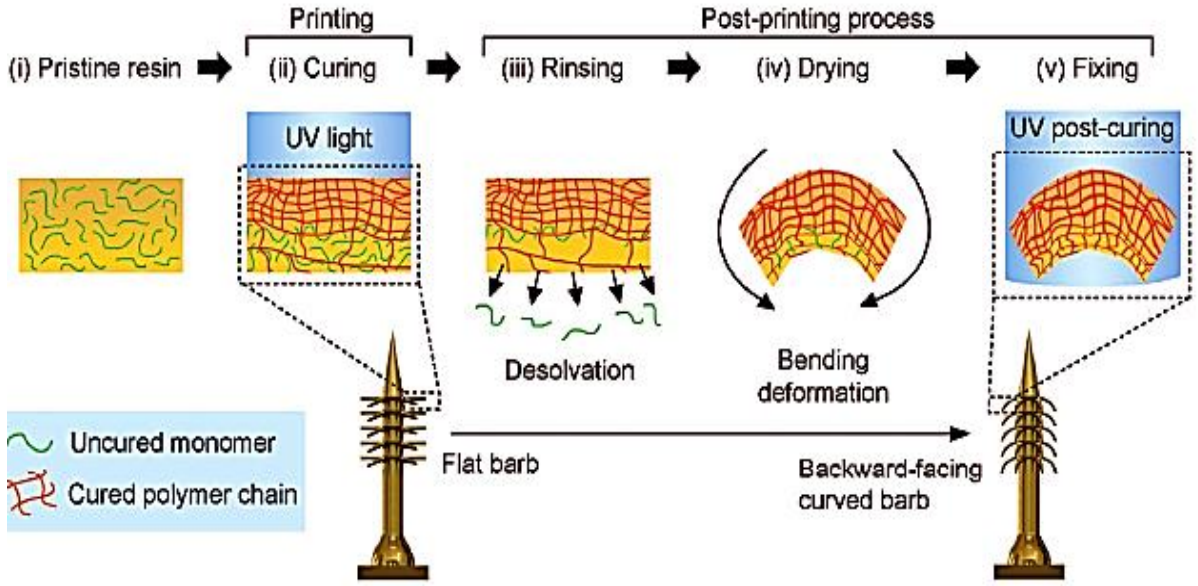

Figure 2: Schematic illustration of 4D printing approach to program deformation of horizontally printed barbs into a backward-facing shape.

The authors have also demonstrated the potential of these 4D printed backward facing barbed microneedles for robust and sustained soft tissue applications, such as transdermal drug delivery, tissue wound healing, and long-term in vivo drug delivery and biosensing.

5.1.2. Tri-layered Mucoadhesive Drug Delivery System:

Using 4D printed materials, a smart sustained drug delivery system has been fabricated by He et al. The device is provided with an outer layer of a pH-sensitive hydrogel. As it reaches the intestinal pH of 6.5, it changes its shape in such a way that it gets anchored to the intestinal wall. This has been successfully demonstrated in the porcine small intestine (using a pH 6.5 buffer), providing a longer residence time in comparison with a standard poly(ε-caprolactone) patch (103 and 72 min respectively), minimizing the drug exposure to intestinal fluid. Using this self-folding mechanism, this tri-layered mucoadhesive drug delivery system device increases the duration of adhesion of the drug-loaded mucoadhesive layer with the intestinal wall, thus resulting in more amount of drug getting traversed across the mucosal epithelium. Fig. 3 depicts this type of smart, Tri-layered Mucoadhesive drug delivery system.[15,16]

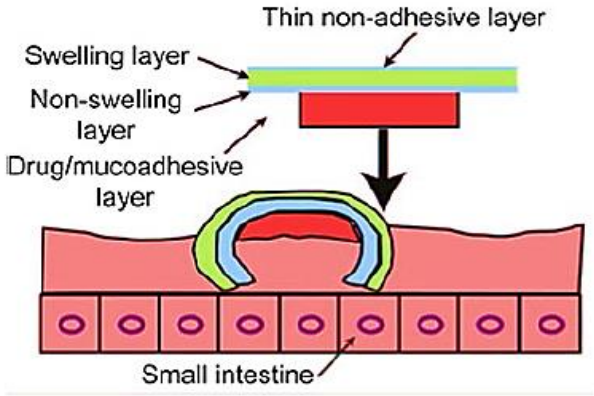

Figure 3: Tri layered Mucoadhesive drug delivery system.

5.1.3. Theragrippers:[16]

These are temperature-sensitive multi-fingered drug delivering devices developed by Malachowski *et al*. After entry into the body at room temperature, this device spontaneously grips onto the tissue when exposed to temperatures beyond 32°C. The drug loading property is attributed to the porosity of the structure that can release

the drug for about 7 days, following the release kinetics of first order. A successful demonstration by in-vitro studies has shown enhanced sustained release of doxorubicin in comparison with a control. Some researchers have further incorporated iron oxide nanoparticles into the porous hydrogel layer of these theragrippers for magnetically guiding the drug delivery system to the target site, providing targeted drug delivery. In some cases, the devices were able to terminate cells from a fibroblast cell clump, showing their potential use in validating some form of surgery themselves.

5.2. 4D Smart Encapsulation Devices:

These are self-folding structures that shapeshift to create an enclosure in which the API or even cells can be encapsulated. Fibroblasts and β- cells of the pancreas have successfully been encapsulated and displayed viability for more than a week. Yeast cells have also been encapsulated in such devices using self-folding capsules that released their contents in response to the surrounding temperature. This technology can be utilized in the concept of enteric drug delivery, wherein, the contents are released in response to change in pH.[17-19]

5.2.1. 4D Microrobot device containing hydrogen bilayer:

A 'micro-robotic' device containing a bilayer of hydrogel has been designed by Li *et al* using conventional lithography. The first layer consists of a pH-responsive gel that can change the morphology of the structure to trap as well as release the contents. The second layer contains iron oxide particles, able to magnetically guide the device to the target site (similar to the aforementioned theragrippers)[20]. The pH of tumor tissue is approximately between 4.5 to 6.0 owing to the high rate of metabolic activity of the tumor microenvironment as well as the low partial pressure of oxygen[21-23]. This approach is utilized by the micro-robot that is designed in such a way that it releases its contents only after encountering acidic tumor tissue. Microbeads loaded with the anti-cancer drugs Paclitaxel and Docetaxel have been encapsulated within these devices and guided magnetically to the target cells. This structure has demonstrated successful release of the drug at the intended site with 70% reduction in cell viability. However, further reduction in size is needed for the device to pass throughout the circulatory system, and become viable clinically. This shows that utilizing this technology can open new doors in targeted drug delivery with minimal side effects, especially in cardio-oncology. By focussing drug release at the tumor site, the therapeutic effects of the drug is maximized and at the same time, while the amount of drug entering the systemic circulation is limited. This lowers the chances of serious side-effects that often limit the use of many anti-cancer medicines[5]. Fig. 4 depicts the encapsulating mechanism of fibroblast cells in a polymeric container.[17]

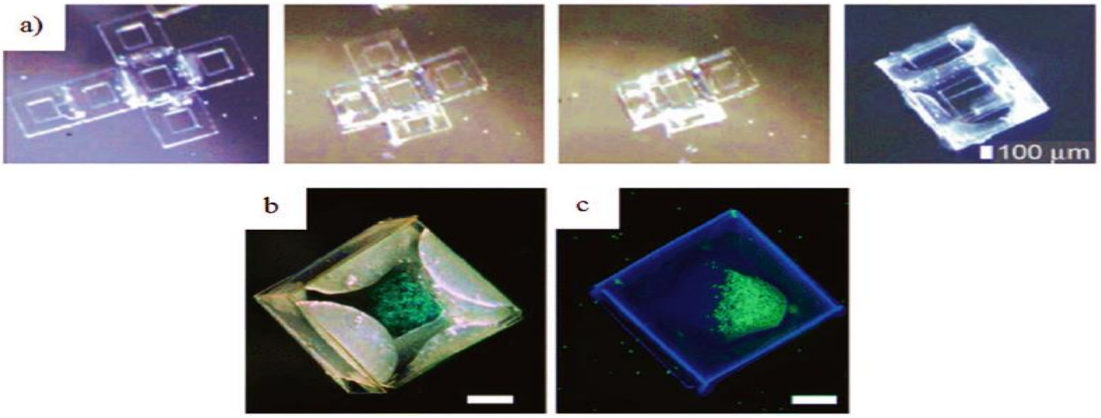

Figure 4: (a) Images showing the self-folding process of a polymeric container at 60°C (b) bright-field and (c) fluorescence z-plane stack image of a non-porous polymeric container containing stained fibroblast cells.

5.3. 4D Bioprinting:

Bioprinting is presently being utilized extensively in 3D printing. With the help of 3D bioprinting, the fabrication of living structures is already old school. However, by transporting this concept into 4D printing, it is possible to fabricate living structures that will have an added feature of being able to respond to their external environment. As of now, Hydrogels as smart materials have been extensively employed in 4D bioprinting mainly because of their osmotic properties and high porosity[24]. As a matter of fact, these hydrogels are being utilized in scaffold-free 4D bioprinting of cells that have the ability to self-assemble, resembling normal tissue growth. This has been demonstrated in one study wherein chondrocytes were encapsulated inside a cylindrical hydrogel, enabling the tissue strands inside the hydrogel lattice to merge over time, producing viable tissues.[25] This proliferation of cells inside hydrogel lattices is referred to as "Bio-ink".[26] All of this signifies that 4D printing is stepping forward to a level wherein selected bio-inks could be used to print complex organs composed of many tissues that will respond to external as well as internal stimuli, similar to that of real physiological organs. Some researchers have used these smart hydrogels to create muscular tissue[25]. The tissue displayed contraction in response to an influx of calcium ions, much like normal muscular physiology. This demonstrates the potential of 4D printing in improving 3D bioprinting, creating complex structures responsive to their environment. Although a lot of work still needs to be done to achieve this aim, research done so far demonstrates the unique ability of this technology to create stimuli-responsive structures in which the cells can mature.[5]

5.3.1. 4D Smart Airway Splints for tracheobroncho-malacia:

Children with tracheobronchomalacia, a disease that causes the windpipe to collapse during breathing, have benefited from 4-D printing technology at CS Mott Children's Hospital, part of the University of Michigan. Before the invention of these 4D printed splints, babies with severe tracheobronchomalacia had little chance of survival. The first clinical assessment came into sight in the journal "Science Translational Medicine". This device referred to as

"airway splint", is a porous, hollow tube that can be sewn around the affected trachea and bronchi to keep them open. Initially, the size of some of the baby bronchi is no longer than a pencil lead. The expandable shape of the airway splint allows the airways to grow with time— typically doubling in size — until they are strong enough to support themselves. A child normally comes out of this condition by the age of three. The splint is made up of biodegradable material (polycaprolactone) which then dissolves harmlessly in the body[27]. Fig. 5 shows the pictures of three 4D printed tracheal airway splints (that grow with time) created using CT scans and 3D imaging software.[27,28]

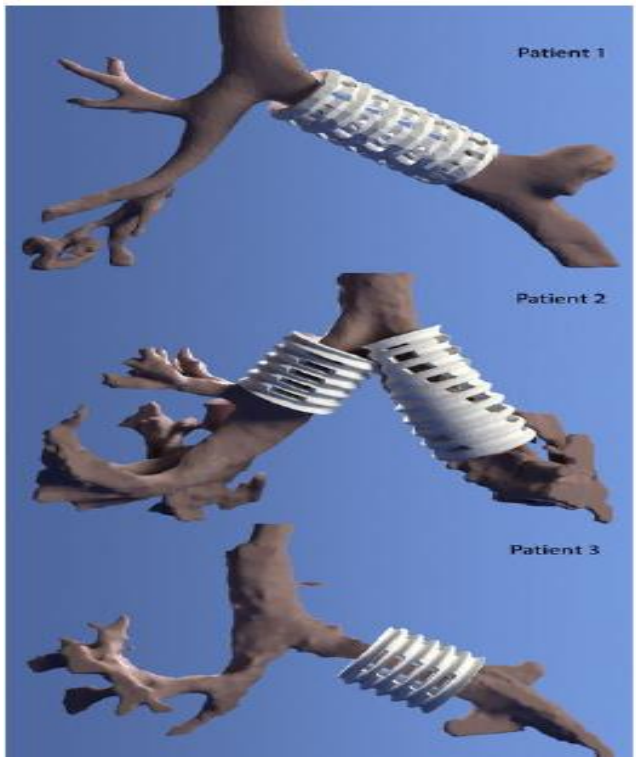

Figure 5: Images of the three 4D printed trachea implants created using CT scans and 3D imaging software.

5.3.2. 4D Bioprinting makes the smallest synthetic blood vessels till date:[29]

At this point of the technology's development, typical 3D bioprinters have the capacity to make tubes at a diameter scale of tens to hundreds of microns. If we compare, the diameter of the smallest blood vessels in the human body is about 5 microns. To achieve the fabrication of such small structures is a significant challenge to the technology, and the ability to create complex tissues for therapeutic treatments.

4D bioprinted vessels created in the Ionov Lab essentially doubles the resolution of a typical tube by allowing the diameter to be created in the act of self-folding. This method is capable of creating vessels with an average internal diameter of 20 microns which is not yet achievable by other existing bioprinting/biofabrication approaches. Additionally, the material used (made from alginate and hyaluronic acid) is not having any negative effect on the viability of the printed cells. This has been proved by supporting the survival of mouse bone marrow for up to 7 days.

Fig. 6 depicts the self-folding progress of the 4D bioprinted vessels (i*mage via Advanced Materials)*

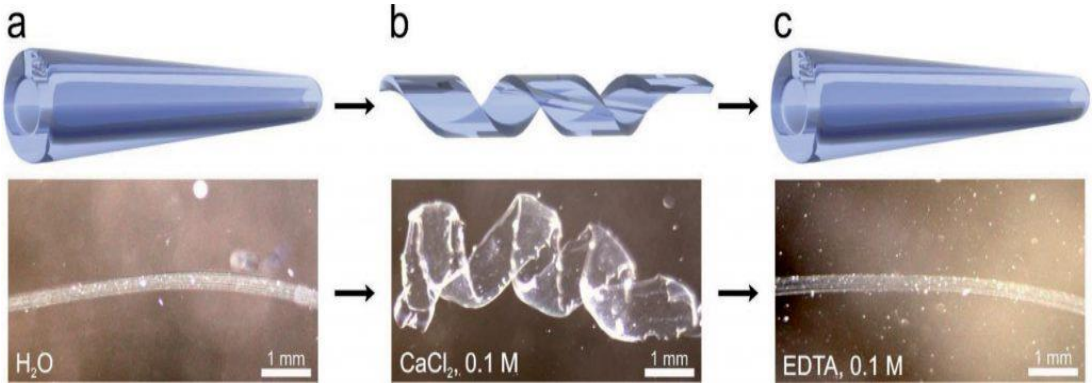

Figure 6: Self-folding progress of the 4D bioprinted vessels (i*mage via Advanced Materials)*

Figure 7 shows the screenshot taken from the Video clip demonstrating living, beating heart cells inside a self-rolled polymer tube. (*Video via leonid lonov*)

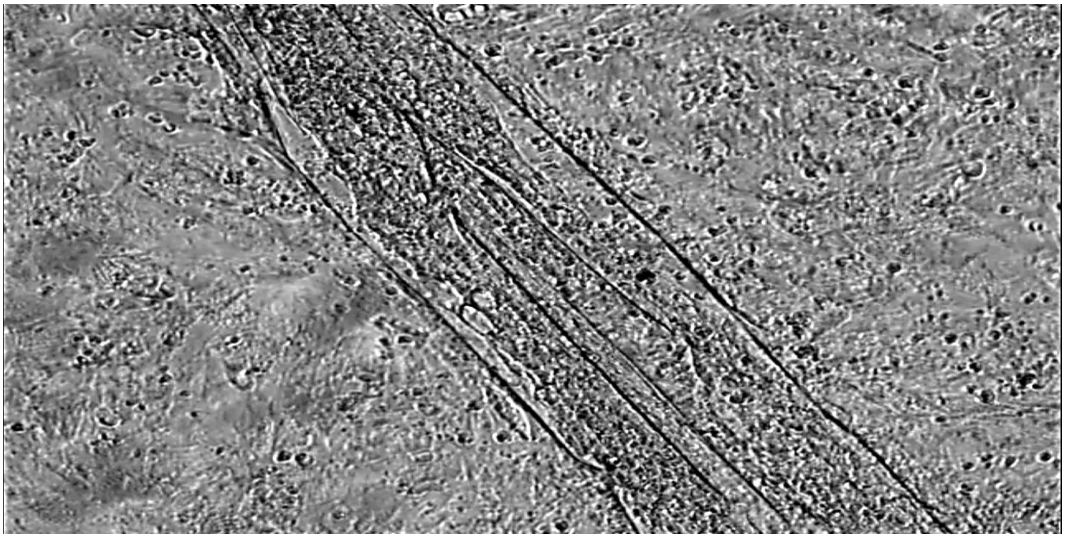

Figure 7: Screenshot taken from the Video clip demonstrating living, beating heart cells inside a self-rolled polymer tube. (*video via leonid lonov*)

6. SUMMARY OF 4D PRINTING IN SMART DRUG DELIVERY AND BIOMEDICAL DEVICES:

The summary of 4D printing in the fabrications of smart drug delivery systems and medical devices is appended in Table 2.

Table 2: Summary of 4D Printing in Smart drug delivery and Medical devices

Mechanism	Stimulus	Delivery System/ Device	Ref
Bioadhesion	UV Light	Smart Microneedles with backward facing barbs	Daehoon *et al*
	pH	Tri-layered Mucoadhesive Drug Delivery System	He *et al*
	Temperature	Theragrippers	Malachowski *et al*
	Temperature	Iron Oxide Nanoparticles incorporated theragrippers	Breger *et al*
Encapsulation	pH	Microrobot device containing hydrogen bilayer	Li *et al*
	pH or Temperature	Self-Folding Capsule	Stoychev *et al*
Bioprinting		Smart Airway Splints for tracheobronchomalacia	Morrison *et al*
		4D bioprinted vessels	Alina *et al*
		Bio Ink	Mironov *et al*
		Smart Hydrogels	Yu *et al*

7. MAJOR CONSTRAINTS OF 4D PRINTING: [30]

- 4D printing technology may replace ample number of complex manufacturing processes which otherwise require large manpower. Hence, there will be no need of such manpower which would result in unemployment and financial crisis.
- At present, this technology may be approved for use only in a few specialized areas such as outer space stations, military, healthcare and war zones. Bringing 4D technology into routine practice may take decades.
- 4D printing may have gone a step further in the field of bioprinting. But, there is lack of research for identifying unknown complications associated with 4D bioprinting of organs and tissues.
- Another major constraint in 4D printing is the high initial cost partially due to there being only a few companies currently developing techniques that support 4D. After commercialization, there may be a reduction in the cost of 4D printing.

8. CONCLUSION:

The applications of 4D printing in the healthcare system and drug designing are going to be enormous. The medical industry accepted to make use of 4D printing, as shape changing 4D prints are expected to be more compatible with the body than rigid metal supports. The “smart” drug delivery systems fabricated via 4D printing will have better patient compliance, low toxicity, low manufacturing cost as well as targeted delivery. To conclude, 4D printing is the future that is going to revolutionize the healthcare industry. Although there is much more to explore and research is going on all across the globe to find new ways by which 4D printing can be used as a valuable tool in healthcare sector as well as in the development of drug delivery systems.

9. ACKNOWLEDGEMENTS:

This work has been funded by the Deanship of King Khalid University. The authors wish to thank the reviewer(s)/editor(s) of this article for their constructive reviews. The authors are also grateful to the Department of Pharmaceutical Sciences, School of Applied Science and Technology, University of Kashmir as well as King Khalid University, Kingdom of Saudi Arabia.

10. COMPETING INTERESTS:

Authors have declared that no competing interests exist.

11. REFERENCES:

1. Tren SJ, Awad A, Goyanes A, Gaisford S, Basit AW. 3D Printing Pharmaceuticals : Drug Development to Frontline Care. Trends Pharmacol Sci. 2018; 39(5):440-51. https://doi.org/10.1016/j.tips.2018.02.006

2. Awad A, Tren SJ, Gaisford S, Basit AW. 3D printed medicines : A new branch of digital healthcare. Int J Pharm. 2018; 548(July):586-96. https://doi.org/10.1016/j.ijpharm.2018.07.024

3. Zema L, Melocchi A, Maroni A, Gazzaniga A. Three-Dimensional Printing of Medicinal Products and the Challenge of Personalized Therapy. J Pharm Sci [Internet]. 2017; 106(7):1697-705. Available from: https://doi.org/10.1016/j.xphs.2017.03.021

4. Awad A, Trenfield SJ, Goyanes A, Gaisford S, Basit AW. Reshaping drug development using 3D printing. Drug Discov Today [Internet]. 2018; 23(8):1547-55. Available from: https://doi.org/10.1016/j.drudis.2018.05.025

5. Firth J, Gaisford S, Basit AW. A New Dimension : 4D Printing Opportunities in Pharmaceutics. In: Jack Firth, Simon Gaisford AWB, editor. 3D Printing of Pharmaceuticals [Internet]. p. 153-62. Available from: https://doi.org/10.1007/978-3-319-90755-0_8

6. Li X, Shang J, Wang Z. Intelligent materials: A review of applications in 4D printing. Assem Autom. 2017; 37(2):170-85. https://doi.org/10.1108/AA-11-2015-093

7. Tibbits S. 4D printing : another dimension. 2018; 1-4.

8. Kuang X, Roach DJ, Wu J, Hamel CM, Ding Z, Wang T, et al. Advances in 4D Printing : Materials and Applications. 2019; 1805290:1-23. https://doi.org/10.1002/adfm.201805290

9. JO W, CHU K, LEE H, MOON M. 3D and 4D printing technologies: an overview. Mater Matters. 2016; 11(2):56-8. https://doi.org/10.1089/3dp.2015.0039

10. Momeni F, N SMMH, Liu X, Ni J. A review of 4D printing. Mater Des [Internet]. 2017; 122:42-79. Available from: \ https://doi.org/10.1016/j.matdes.2017.02.068

11. Lu HZ, Yang C, Luo X, Ma HW, Song B, Li YY, et al. Materials Science & Engineering A Ultrahigh-performance TiNi shape memory alloy by 4D printing. 2019; 763(May). https://doi.org/10.1016/j.msea.2019.138166

12. Siang Y, Ting W, Poh L, Wu Y, Lai Y, Li H. Acta Biomaterialia 4D Printing and Stimuli-responsive Materials in Biomedical Applications. Acta Biomater [Internet]. 2019; 92:19-36. Available from: https://doi.org/10.1016/j.actbio.2019.05.005

13. Alan Lai, Zehui Du, Chee Lip Gan CAS. Shape Memory and Superelastic Ceramics at Small Scales. American Association for the Advancement of Science [Internet]. 2013 Sep; 341(6153):1505-8. Available from: www.sciencemag.org https://doi.org/10.1126/science.1239745

14. Han D, Morde RS, Mariani S, Mattina AA La, Vignali E, Yang C, et al. 4D Printing of a Bioinspired Microneedle Array with Backward-Facing Barbs for Enhanced Tissue Adhesion. 2020; 1909197. https://doi.org/10.1002/adfm.201909197

15. He H, Guan J, Lee JL. An oral delivery device based on self-folding hydrogels. 2006; 110:339-46. https://doi.org/10.1016/j.jconrel.2005.10.017

16. Malachowski K, Breger J, Kwag HR, Wang MO, Fisher JP, Selaru FM, et al. Stimuli-Responsive Theragrippers for Chemomechanical Controlled Release ** Angewandte. 2014; 1-6. https://doi.org/10.1002/ange.201311047

17. Azam A, Laflin KE, Jamal M, Fernandes R, Gracias DH. Self-folding micropatterned polymeric containers. 2011; 51-8. https://doi.org/10.1007/s10544-010-9470-x

18. Stoychev G, Puretskiy N, Ionov L. Self-folding all-polymer thermoresponsive microcapsules. Soft Matter. 2011; 7(7):3277-9. https://doi.org/10.1039/c1sm05109a

19. Yoon C, Xiao R, Park J, Cha J. Functional stimuli responsive hydrogel devices by self-folding. 094008. https://doi.org/10.1088/0964-1726/23/9/094008

20. Li H, Go G, Ko SY, Park J, Park S. Magnetic actuated pH-responsive hydrogel- based soft micro-robot for targeted drug delivery. Smart Mater Struct [Internet]. 25(2):27001. Available from: https://doi.org/10.1088/0964-1726/25/2/027001

21. Manchun S, Dass CR, Sriamornsak P. Targeted therapy for cancer using pH-responsive nanocarrier systems. Life Sci [Internet]. 2012; 90(11-12):381-7. Available from: https://doi.org/10.1016/j.lfs.2012.01.008

22. Swietach P, Vaughan-jones RD, Harris AL, Hulikova A. The chemistry , physiology and pathology of pH in cancer. Philos Trans R Soc B. 2014; 1-10. https://doi.org/10.1098/rstb.2013.0099

23. Tong Z, Luo W, Wang Y, Yang F, Han Y, Li H, et al. Tumor Tissue-Derived Formaldehyde and Acidic Microenvironment Synergistically Induce Bone Cancer Pain. 2010; 5(4). https://doi.org/10.1371/journal.pone.0010234

24. Jadhav RG, Das AK. 10. Four dimensional printing in healthcare [Internet]. 3D Printing in Medicine. Elsevier Ltd; 2017. 207-218 p. Available from: https://doi.org/10.1016/B978-0-08-100717-4.00010-7

25. Yu Y, Moncal KK, Li J, Peng W, Rivero I, Martin JA, et al. Three-dimensional bioprinting using self-assembling scalable scaffold-free " tissue strands " as a new bioink. Nat Publ Gr [Internet]. 2016;(February):1-11. Available from: https://doi.org/10.1038/srep28714

26. Mironov V, Visconti RP, Kasyanov V, Forgacs G, Drake CJ, Markwald RR. Biomaterials Organ printing : Tissue spheroids as building blocks q. Biomaterials [Internet]. 2009; 30(12):2164-74. Available from: https://doi.org/10.1016/j.biomaterials.2008.12.084

27. Morrison RJ, Hollister SJ, Niedner MF, Mahani MG, Park AH, Mehta DK, et al. Mitigation of tracheobronchomalacia with 3D-printed personalized medical devices in pediatric patients. Sci Transl Med. 2015; 7(285):1-12. https://doi.org/10.1126/scitranslmed.3010825

28. Chu H, Yang W, Sun L, Cai S, Yang R, 4D Printing : A Review on Recent Progresses, Micromachines. 2020; 11:1-30. https://doi.org/10.3390/mi11090796

29. Kirillova A, Maxson R, Stoychev G, Gomillion CT, Ionov L. 4D Biofabrication Using Shape-Morphing Hydrogels. Adv Mater. 2017; 1703443:1-8. https://doi.org/10.1002/adma.201703443

30. Alexa V, Raiu SA. Advancements in the Research of 4D Printing-A Review ADVANCEMENTS IN THE RESEARCH OF 4D PRINTING-A REVIEW. In: IOP Conference Series: Materials Science and Engineering. 2018