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Ear Reconstruction and 3D Printing: Is It Reality?

Alexandra J. Lin[1] · Jaime L. Bernstein[1] · Jason A. Spector[1,2]

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Abstract
Purpose of Review Autologous reconstruction of microtia, the most common congenital external ear deformity, is one of the more challenging plastic surgical procedures, causing obligatory donor site morbidity and often resulting in suboptimal aesthetic outcomes. Recent advances in the fields of tissue engineering and 3D printing promise to profoundly affect the practice of reconstructive surgery.
Recent Findings 3D printed guides are already used by reconstructive surgeons during correction of complex anatomic defects. Similarly, the prosthetics industry has benefited from the ability to rapidly prototype customized pieces. Bioprinting, the ability to 3D print living tissue, is an emerging field that may soon allow the possibility of creating autologous cartilage in specific shapes.
Summary In this review, we explore the numerous ways 3D printing is being employed to address external ear deformity and how, when used in combination with cutting-edge tissue engineering technology, it may finally help us achieve the holy grail of ear reconstruction—an "off-the-shelf" auricular scaffold.

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Alexandra J. Lin and Jaime L. Bernstein have contributed equally to this work.

✉ Jason A. Spector
Jas2037@med.cornell.edu

[1] Laboratory of Bioregenerative Medicine and Surgery, Division of Plastic Surgery, Weill Cornell Medical College, 525 E 68th St, Payson 709-A, New York, NY 10065, USA

[2] Nancy E. and Peter C. Meinig School of Biomedical Engineering, Cornell University, Ithaca, NY, USA

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Introduction

Microtia is the most common congenital auricular anomaly with a prevalence of approximately 2 per 10,000 births [1••, 2••]. This external ear deformity encompasses a broad spectrum of phenotypes from mild structural anomalies to complete absence of the ear (anotia) (Fig. 1 [1••]) [1••, 2••]. Microtia and its associated disfigurement, even when minor, can cause significant psychological distress and negatively impact psychosocial functioning. [2••, 3–6, 7••] Beyond congenital anomalies, ear reconstruction is often necessary secondary to trauma or oncologic resections, with more than 1 in 500 people sustaining an acquired auricular deformity annually [2••, 8, 9].

The current gold standard for reconstruction of the external ear, which utilizes autologous tissue, has significant shortcomings [10, 11, 12•, 13, 14•]. In this procedure, costal cartilage is harvested from multiple ribs and then meticulously sculpted and assembled into a three-dimensional ear scaffold before being implanted under the periauricular skin [15, 16]. However, the resultant donor site is quite painful (especially for children), and the carving of the scaffold into a reasonable facsimile of the ear requires immense technical skill and experience [1, 17–21]. Further, the use of rib fibrocartilage instead of elastic cartilage poorly mimics the biomechanical properties of native elastic ear cartilage. Thus, surgeons have long sought an innovative solution devoid of the morbidity and technical challenges associated with autologous ear reconstruction.

Tissue engineering—"an interdisciplinary field that applies the principles of engineering and life sciences

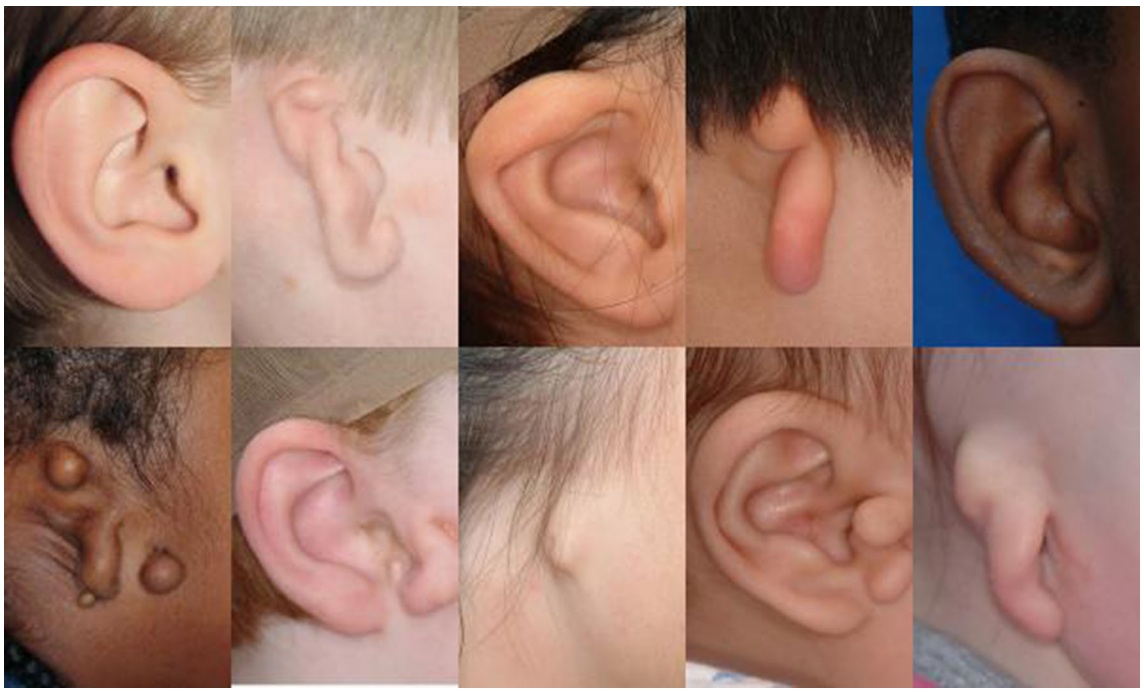

Fig. 1 Microtia and anotia. Digital photographs showing the wide range of phenotypes seen in microtia/anotia. Top left image shows a normal ear Reproduced with permission from Fig. 1 of [1••]

toward the development of biological substitutes that restore, maintain, or improve whole organ or partial tissue function" [22]—has long offered the promise of creating "off-the-shelf" tissue replacements for use in patients [23]. This approach holds particular potential for cartilage engineering as one of the major limitations for engineering tissues of clinically relevant size remains the inability to create an inherent hierarchical vasculature, a requirement obviated in cartilage, which is an avascular tissue.

Similar to the field of tissue engineering, the advent of three-dimensional printing (also known as additive manufacturing) can be traced back to the 1980s [24, 25••, 26]. The flexibility of its application has allowed engineers to design everything from intricate machine parts to self-assembling houses, and more recently it has become an important tool for innovation and rapid prototype fabrication in science and medicine. [25••, 26, 27•, 28••, 29••] Combined with brisk advances in printer technology—some printers are capable of printing in resolutions at the micron scale—it is now feasible to take a conceptual object or design and have a prototype printed within the span of a single day. [29••, 30••] In the medical world, this translates into the ability to take digital images of intricate three-dimensional patient anatomy, such as an ear or an anomalous internal organ, and transform those CT or MRI images into 3D models which can be handled and studied with a level of detail and precision previously unavailable (Fig. 2). This functionality is particularly appealing to the reconstructive surgeon as it offers the potential for more accurate and aesthetic outcomes in even the most challenging cases. [31••]

At present, 3D printing is already used clinically by some microtia surgeons as a modeling guide for creation of the scaffold during ear reconstruction, either autologous (costal cartilage) [32••, 33••] or prosthetic (MEDPOR®) [34••, 35, 36••]. Patients uninterested in or ineligible for surgical reconstruction have also benefited from the creation of custom-made prosthetic ears with contour matching previously unattainable by the traditional methods of prosthesis creation [37]. However, it is the combination of 3D printing technology with advanced tissue engineering that promises to deliver the holy grail of ear reconstruction—an "off-the-shelf" auricular scaffold composed of autologous elastic cartilage that is a perfect mirror image of the patient's contralateral ear.

Modeling the Auricle

Because of the inherent technical difficulty associated with reconstruction of the microtic auricle and the extensive experience required, very few plastic surgeons routinely perform this procedure [38••]. The traditional approach to creating a mirror image auricle involves the placement of a transparent film against the unaffected ear (or a radiograph of the ear) to hand-trace the ear's main topographic

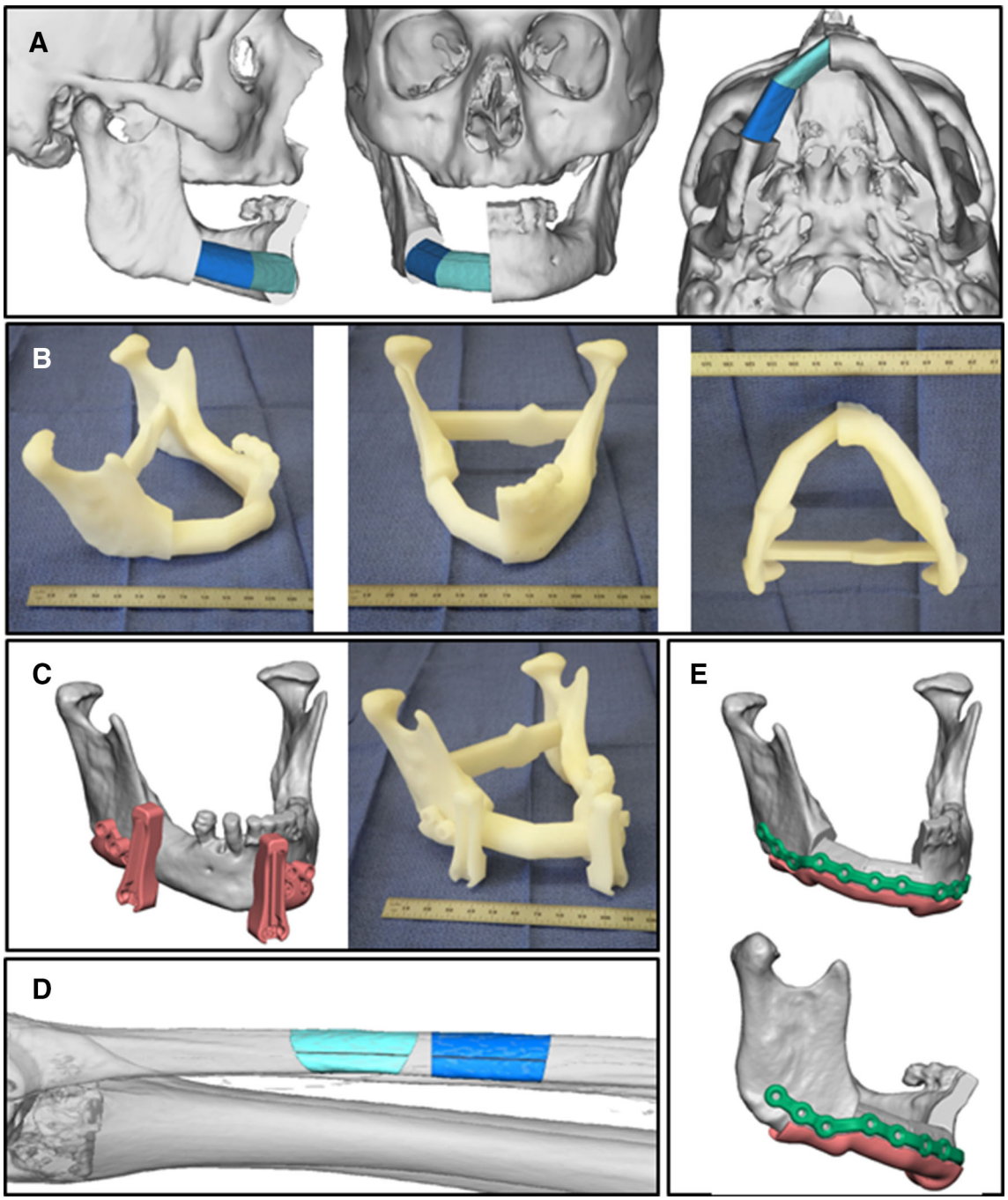

Fig. 2 3D Printing in reconstruction planning. Digital rendering and reconstruction planning of mandibular neoplasm with free fibula flap and 3D printed guides. **a** Digital images of planned reconstruction. **b** 3D printed guide of mandible showing reconstruction plan. **c** Digital and 3D printed mandible cutting guides. **d** Fibula cutting guide showing sections of bone that will be integrated into reconstructed mandible. **e** Final reconstruction with custom osseointegrated plate

features. This two-dimensional film is then used as a visual guide in the operating room to shape the harvested rib cartilage. Many limitations of this 2D model, such as lacking height, depth features, and the intricate details of the auricle (of the 14 described structures of the auricle, usually only 6–8 are captured [32••]), often resulted in suboptimal 3D outcomes. [33••]

Although surgeons have previously utilized patient-specific "3D" ear models made from plaster [39••], paper [40], or other materials [41, 42], it is only in the past few years, with the increasing availability and precision of (non-ionizing) image acquisition and printing technology, that the ability to rapidly (within hours) create a patient-specific 3D model of the ear has become feasible, leading

directly to improved patient outcomes. Although surface image acquisition includes both the skin and cartilage, the thickness of the skin over the auricle ranges from 0.8 to 1.0 mm [43]. 3D printed models of an ear, thus, are close approximations of the natural underlying elastic cartilage.

In 2016, Jeon et al. demonstrated improved outcomes in autologous reconstruction for microtia when using a 3D model as a guide [32••]. In this work, a casting technique was used to generate an alginate mold of the patient's unaffected ear (they believed that the patients were too young to remain still long enough to complete a scanning process). The mold was used to cast the patient's ear, which was subsequently laser scanned and digitally transformed into the mirror image of the unaffected ear and then 3D printed. The custom ear models were utilized in combination with the Nagata technique [16, 44•] to reconstruct the microtic ears. The group concluded that the 3D printed models differed in shape on overlaid image comparison an average of 2.31% from actual ear, whereas the 2D model differed over 16% [32••].

In the same year, Zhou et al. described the use of a three-dimensional printed template for autologous ear reconstruction [33••]. In this study, a 3D surface scanner with 0.1 mm resolution was used to acquire the details of the patient-specific auricle, which were then processed and converted to stereolithography interface format (STL) files. The data were then used to 3D print both a 2D sheet mold designed from the surface scan of the unaffected ear and a 3D ear-shaped model of the patient-specific auricle. These templates were used to assist in autologous costal cartilage auricular reconstruction, providing the surgeon with information such as height, width, and depth of the contralateral ear while reconstructing the affected ear (Fig. 3 [33••]). This new planning technique was compared to the results achieved using the traditional 2D contralateral tracing approach. When comparing their templates against the standard 2D film guide, they found that the 3D model produced significantly more accurate outcomes ($p < 0.001$) on the evaluation of five distinct indexes (symmetry, length, width, cranioauricular angle, and the substructure of the reconstructed ear) as evaluated by both the surgeons and the patients' parents [33••].

As demonstrated by these studies, 3D printing a patient-specific auricular model not only improves aesthetic outcomes, but also reduces operating room time (Zhou et al. found that operative time decreased an average of 15 min when using a 3D printed template [33••]). Although the preoperative planning (scanning the unaffected ear, digitally remodeling or altering it, printing the model, etc.) is more costly and requires greater involvement on part of the reconstructive team, the reduction in surgical time and increased precision and accuracy of outcomes represent indisputable benefits with few, if any, disadvantages to the patient [32••, 33••].

3D Printing and Prosthesis Creation

In some instances, reconstructive options for auricular defects are limited by a lack of sufficient autologous tissue, either at the donor or recipient site, or by patient comorbidities. In such cases, patients may achieve a better result through the use of a custom prosthesis [45••, 46]. Traditional design and fabrication of craniofacial prosthetics is performed by clinical anaplastologists through a meticulous artistic and technical hand carving process (Fig. 4a [47]). Given the expertise and length of time required to produce a high-quality prosthesis, the cost of production can reach $15,000 (and may not be covered by insurance [48]).

In perhaps the most obvious example of the disruptive nature of 3D printing in the clinic, intricately shaped prosthetics, which previously had to be hand-made, can now be rapidly prototyped through a combination of 3D image acquisition and digital remodeling (Fig. 4b [37]). From these virtual renderings, it is possible to print solid prosthetic ear models [49–51]. Because prosthetics are commonly made from polydimethylsiloxane or silicone, the 3D printed ears then become an invaluable tool for the creation of custom casting molds [37, 49, 52, 53]. These personalized prostheses can be manufactured in hours at a fraction of the cost of the traditional method. As they are modeled from the patient's unaffected ear, the final prosthetic has near-perfect symmetry with regard to shape, size, and intricate anatomic features. Currently, it is not possible to directly print a skin tone-matched silicone implant, and thus the prosthetic ear still requires a custom paint job to provide natural skin tone [29••, 54, 55•, 56••, 57•]. With further technological improvements, the direct printing of prosthetics with correct color matching will soon be possible.

Although the majority of published literature on the subject of 3D printing prosthetics has involved the use of costly industrial printers, the technology exists for the creation of acceptable ear prosthetics using much cheaper printers [56••]. In 2014, He et al. created a custom prosthetic for an approximate cost of $30 using an "at-home" desktop 3D printer. The final prosthesis was not tested clinically; however, the results are promising and suggest a future in medical 3D printing that is not limited by cost or feasibility [56••]. Such inexpensive printing technology holds promise for immediate application in the developing world and other underserved populations.

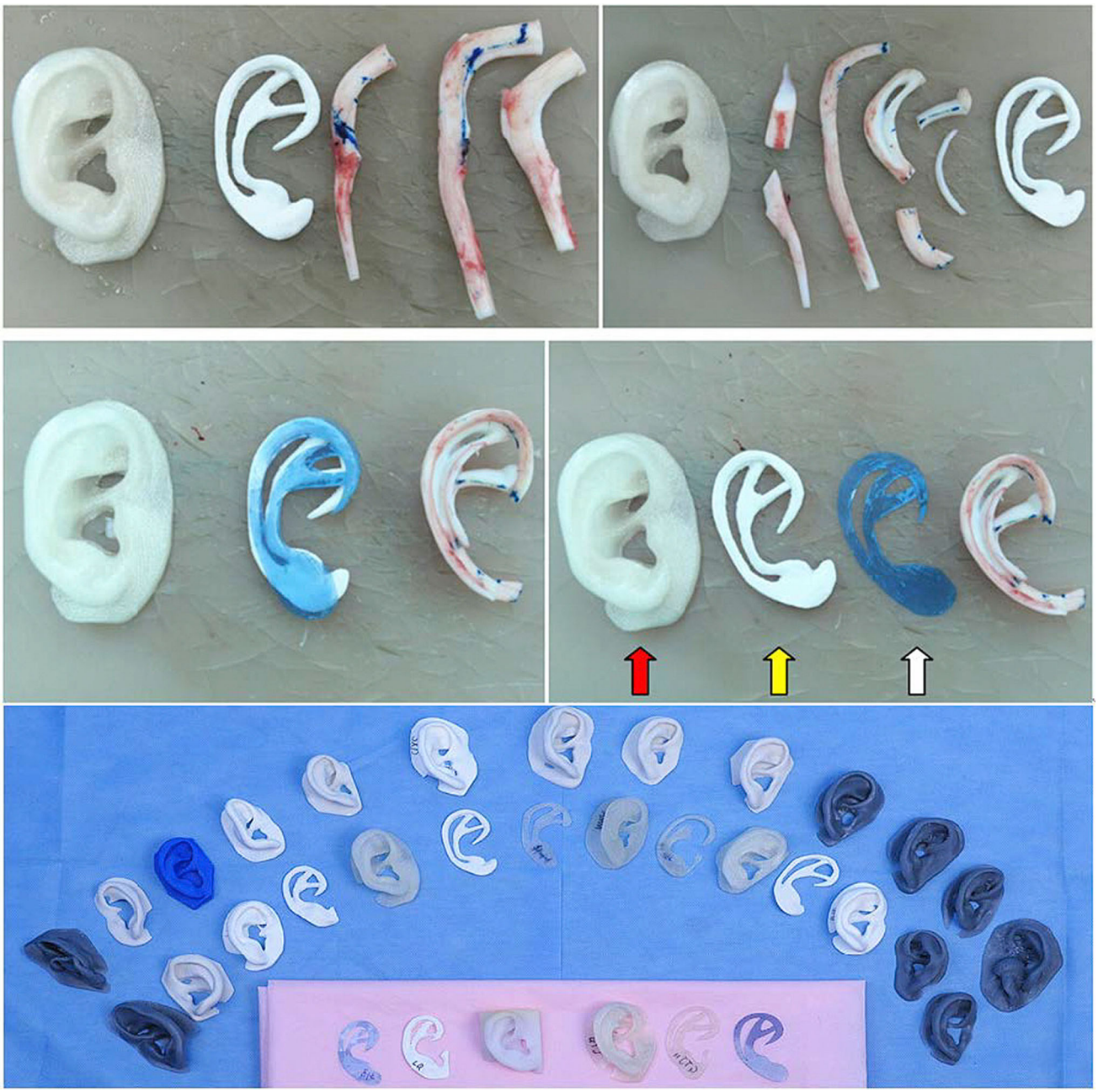

Fig. 3 Surgical modeling in auricular reconstruction. Various auricular guides used in costal cartilage reconstruction of the auricle. (Red arrow) 3D printed model, (yellow arrow) sheet molding template, and (white arrow) traditional drawn X-ray film template Reproduced with permission from Fig. 6 of [33••]

Tissue Engineering: Fulfilling the Promise?

Although direct 3D printing of auricular prostheses or models for intraoperative use has certainly resulted in incremental improvements in aesthetic outcomes as well as cost savings, the ultimate adaptation of this technology would directly print living tissue. This process, known as "bioprinting," prints cells within a biologic carrier ink. However, in order to understand how bioprinting may be applied to the fabrication of living auricles, one must first review the progress made in the field of auricular tissue engineering.

In 1997, Cao et al. published their iconic "ear on a mouse" in which a shaped polyglycolic acid/polylactic acid scaffold was seeded with bovine articular chondrocytes and incubated in vivo for up to 12 weeks [58]. Although this seminal work demonstrated the feasibility of creating cartilage in a specific framework, the authors used

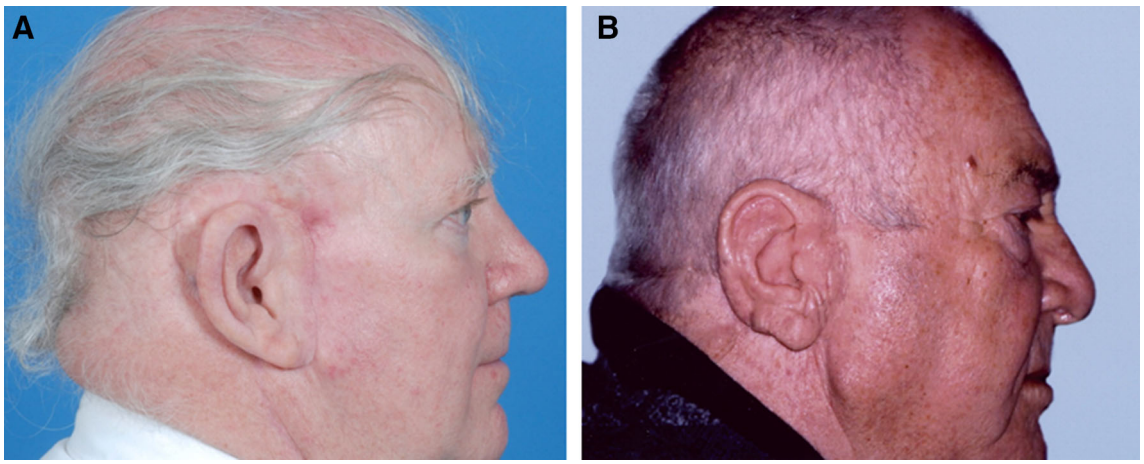

Fig. 4 3D printed prostheses. Comparison of traditional hand-sculpted silicone prosthetic ear **a** with ear made from a 3D printed mold **b** following color matching and hand-painting of dermatologic details in both examples 4A courtesy of Dr. Charles Thorne. 4B reproduced with permission from Fig. 7 of [37]

articular chondrocytes as their cell source. Further, despite the impressive image of the "ear" on the mouse, the shape was due primarily to both the scaffold material itself and support from external stenting, with very little (articular) cartilage formation. Over the past two decades since that report, considerable challenges have been overcome in the quest to develop a tissue-engineered auricular scaffold for clinical application [58].

Successful engineering of an ear scaffold requires the careful manipulation of several variables including an appropriate cell source (i.e., auricular chondrocytes) seeded in/onto a suitable biocompatible scaffold that not only can be formed into an accurate facsimile of a patient's ear but that can also degrade over time to be replaced by the deposition of elastic cartilage matrix. Further, this process must be finely tuned to ensure that the rate of degradation does not exceed the rate of deposition of elastic cartilage matrix or else the scaffold will lose its size and/or intricate shape.

Several authors have utilized 3D printing to fabricate auricular scaffolds made of various biocompatible materials onto which chondrocytes may then be seeded. Synthetic polymers such as polylactic acid (PLA) [59, 60], polylactic glycolic acid (PLGA) [58, 61, 62], and polycaprolactone (PCL) [63••, 64••, 65••] are praised for the level of control available in the fabrication process and their reliably reproducible results [2••, 65••, 66, 67, 68••]. The initial stiffness of these materials resists the deforming and contractile forces exerted both extrinsically and intrinsically, respectively. However, limitations of these polymers for use as scaffolding material include the inability to encapsulate cells within them, thus limiting the seeding density to the number of cells that can fit on the surfaces of the construct. Furthermore, as these materials degrade, they are not replaced with cartilaginous matrix, leaving the final construct shape in doubt. Accordingly, despite numerous reports that have demonstrated the development of cartilage to some extent on such scaffolds, their use in this context for clinical application seems unlikely [2••, 66, 69•].

In contrast, the use of naturally derived biologic materials as scaffolding, such as collagen [31••, 70••, 71••, 72••, 73], alginate [74, 75•, 76], and chitosan [62, 77, 78], has been explored with increasing success over the past few years. Although biologic scaffolds are more delicate and may introduce greater variability in their creation, chondrocytes may be encapsulated within the matrix at a sufficient density to allow for efficient transformation of the matrix into mature elastic cartilage.

Alginate hydrogels are one of the most studied biomaterials utilized in tissue engineering due to their durability and relative ease of use; however, chondrocytes cannot attach to alginate and therefore do not grow as well in 3D culture [79, 80]. Furthermore, because alginate is a polysaccharide derived from seaweed, chondrocytes are unable to degrade the polymer as mammalian cells lack the enzyme alginase [81••]. In vitro, it is possible to dissolve alginate using various chelating agents such as EDTA or sodium citrate, but this process would not be possible in vivo [79, 81••]. Thus, the translational potential of alginate-based hydrogels for auricular tissue engineering seems limited.

In contrast, collagen hydrogels are readily degraded (at a tunable rate) to their constituent amino acids in mammals by various collagenases and metalloproteinases [81••]. As the most abundant protein in humans (comprising approximately 30% of all protein in the human body) and the major component of the extracellular matrix, type I collagen provides an ideal environment for 3D culture of chondrocytes in vitro and in vivo [80, 81••].

In 2014, Reiffel et al. reported the fabrication of high-fidelity full-size pediatric auricular constructs using bovine auricular chondrocytes [31••]. Through the combination of 3D photogrammetry and 3D printing, they were able to create a 7-piece mold of the digitized ear image. Ear-shaped constructs were then fabricated from chondrocytes encapsulated within type I collagen via the process of injection molding (Fig. 5). The constructs were implanted in nude rats and, after 3 months in vivo, scaffolds demonstrated histologic, biomechanical, and biochemical qualities identical to those of native (bovine) auricular cartilage [31••]. Follow-on studies demonstrated the effective "permanence" of these scaffolds at 6 months [70••] as well as the effectiveness of this approach using human auricular chondrocyte [82••].

With the advent of bioprinters capable of printing biologic inks (such as collagen containing encapsulated chondrocytes), we are now at the cusp of being able to "print" high-fidelity custom auricular scaffolds on demand. However, printing a living auricular-shaped scaffold is only the first step. For translation to the clinic, strategies to mitigate loss of size and topographic definition of the maturing scaffold must be developed.

The need for architectural stability and maintenance of topographic detail has long been a challenge in auricular tissue engineering. Cao et al. initially supported their xenografted constructs with fixed, external stents, but on removal of the stents the constructs contracted and lost architectural detail. [58] Khan et al. have attempted to increase the rate of cartilage maturation by applying various growth factors to their constructs such as fibroblast growth factor (FGF)-2 and transforming growth factor beta-1 (TGF- β 1) [83, 84]. Others have developed methods of creating hybrid constructs consisting of degradable 3D printed external or integrated polymers, i.e., 3D printed "cages," with internal spaces that can support the delicate structure of chondrocyte-seeded collagen hydrogels [85–87, 88••].

In the pursuit of a 3D bioprinted ear, a bioink must be stiff enough to capture the intricate topography of the auricle [2••]. A Swedish group addressed this hurdle by mixing cellulose nanofibrils into alginate, effectively stiffening the bioink. They were able to 3D bioprint an ear-shaped scaffold that demonstrated viable chondrocytes after printing [89••]; however, the longevity of this method and performance in vivo are unknown.

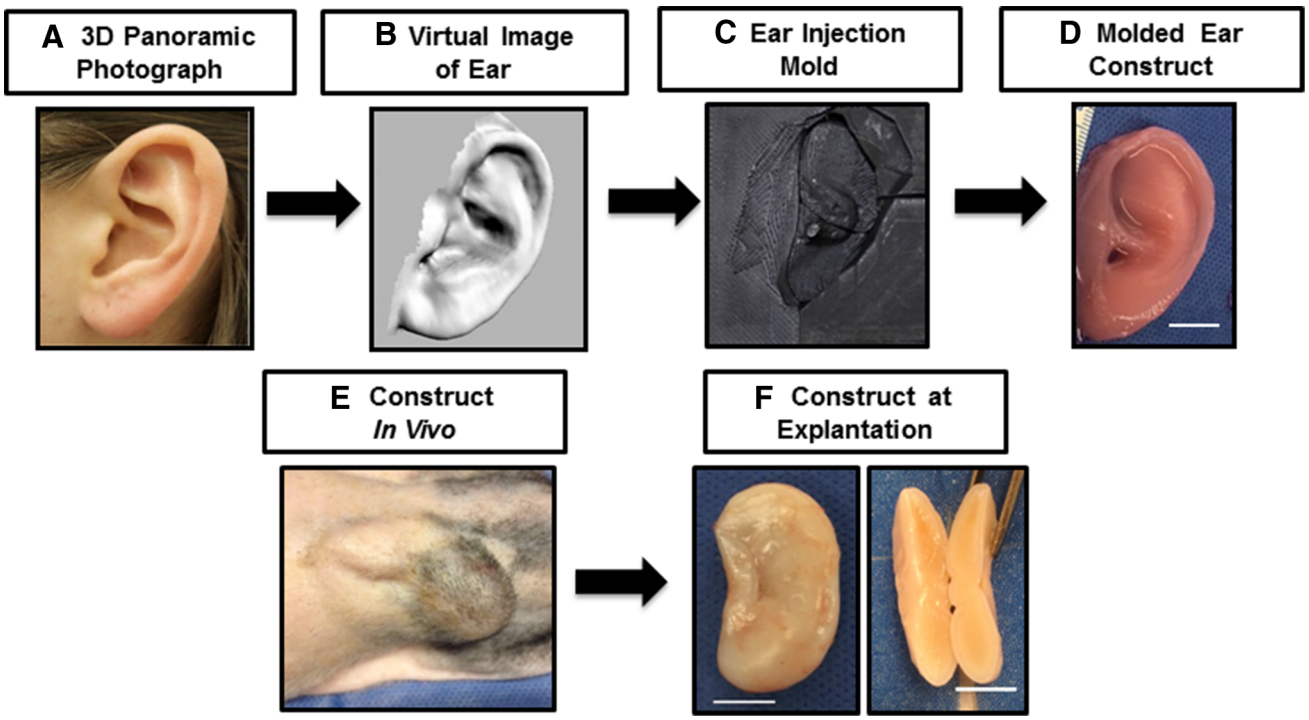

Fig. 5 Injection molding of chondrocyte-seeded type I collagen hydrogels. **a**, **b** Normal ear is digitized with a 3D surface scanner. **c** Using this 3D rendering, a negative mold is created. **d** Type I collagen seeded with bovine auricular chondrocytes is injected into the mold forming a hydrogel construct in the shape of an ear. **e** Construct implanted in vivo in nude rats for up to 6 months. **f** Explanted construct demonstrates the formation of neocartilage on gross appearance. Scale bars = 1 cm

Other investigators have employed support material that can be removed or that degrades after the scaffold is established [89••]. Park et al. have printed with alginate on a PCL scaffold, demonstrating higher chondrocyte survival in vitro in the bioprinted scaffold as compared to the hand-seeded scaffold. In vivo, their bioprinted scaffolds exhibited more robust neocartilage formation [90••]. Other approaches include printing alternate layers of electrospun PCL nanofibers with fibrin–collagen hydrogel to increase the strength of the construct and allow the hydrogel to withstand naturally occurring mechanical forces in vivo. The resulting constructs formed cartilage-like material over the course of several months [29••, 64••]. The question of whether the integration of non-permanent synthetic material, such as PCL, would affect the final architecture of the neocartilage over the long term remains to be determined.

An alternative to integrated support is the use of sacrificial support material. In 2014, Lee et al. proposed polyethylene glycol (PEG) as a feasible support material when used in combination with their PCL and cell-laden hydrogels [91••]. PEG can be sacrificed by the addition of aqueous solutions and has no effect on cell viability. The shortcoming of sacrificial support is that it must be sacrificed prior to scaffold implantation and therefore cannot contribute to the maintenance of scaffold geometry in vivo.

Morris et al. have proposed a method employing stereolithography in the fabrication of an ear-shaped scaffold from natural chitosan and synthetic polyethylene glycol diacrylate [92••]. They have shown that their scaffolds maintained cell viability within the hybrid material over the long term, but the use of chitosan (non-biodegradable in mammals) as their biologic scaffolding material does not provide the most suitable environment for the proliferation of chondrocytes and development of a natural extracellular matrix.

Conclusions

Which of these various tissue engineering approaches will ultimately yield the "off-the-shelf" auricular scaffold remains to be seen. Nonetheless, it is clear that 3D printing has fostered the creation of sophisticated scaffolds, either as supports or as the construct itself in a variety of synthetic, biologic, and hybrid materials. As the technology continues to improve, we can expect translational work to follow in close succession.

Compliance with Ethics Guidelines

Conflict of interest The authors declare no conflicts of interest relevant to this manuscript.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

References

Papers of particular interest, published recently, have been highlighted as:

- • Of importance
- •• Of major importance

1. ••Luquetti D, Heike C, Hing A, et al. Microtia: epidemiology and genetics. Am J Med Genet A. 2013;158A(1):124–39.
2. ••Jessop Z, Javed M, Otto I, et al. Combining regenerative medicine strategies to provide durable reconstructive options: auricular cartilage tissue engineering. Stem Cell Res Ther. 2016;7(1):19.
3. Horlock N, Vögelin E, Bradbury E, et al. Psychosocial outcome of patients after ear reconstruction: a retrospective study of 62 patients. Ann Plast Surg. 2005;54(5):517–24.
4. Du J, Zahuang H, Chai J, et al. Psychological status of congenital microtia patients and relative influential factors: analysis of 410 cases. Zhonghua Yi Xue Za Zhi. 2007;87(6):383–7.
5. Jiamei D, Jiake C, Hongxing Z, et al. An investigation of psychological profiles and risk factors in congenital microtia patients. J Plast Reconstr Aesthetic Surg. 2008;61(1):S37–43.
6. Li D, Chin W, Wu J, et al. Psychosocial outcomes among microtia patients of different ages and genders before ear reconstruction. Aesthetic Plast Surg. 2010;34(5):570–6.
7. •Johns A, Lucash R, Im D, Lewin S. Pre and post-operative psychological functioning in younger and older children with microtia. J Plast Reconstr Aesthetic Surg. 2015;68(4):492–7.
8. Gault D. Post traumatic ear reconstruction. J Plast Reconstr Aesthetic Surg. 2008;61(1):S5–12.
9. Bhandari P. Total ear reconstruction in post burn deformity. Burns. 1998;24(7):661–70.
10. Thomson H, Kim T, Ein S. Residual problems in chest donor sites after microtia reconstruction: a long-term study. Plast Reconstr Surg. 1995;95(6):961–8.
11. Kawanabe Y, Nagata S. A, new method of costal cartilage harvest for total auricular reconstruction: part I. Avoidance and prevention of intraoperative and postoperative complications and problems. Plast Reconstr Surg. 2006;117(6):2011–8.
12. •Sun Z, Yu X, Chen W, et al. Costal cartilage assessment in surgical timing of microtia reconstruction. J Craniofac Surg. 2017;28(6):1521–5.
13. Park C, Yoo Y, Hong S. An update on auricular reconstruction: three major auricular malformations of microtia, prominent ear and cryptotia. Curr Opin Otolaryngol Head Neck Surg. 2010;18(6):544–9.
14. •Go Y, Kang Y, Hwang J, Oh K. Management of chest deformity caused by microtia reconstruction: comparison of autogenous diced cartilage versus cadaver cartilage graft partial filling techniques. J Plastic Reconstr Aesthetic Surg. 2017;70(1):104–9.
15. Shieh S, Terada S, Vacanti J. Tissue engineering auricular reconstruction: in vitro and in vivo studies. Biomaterials. 2004;25:1545–57.
16. Walton R, Beahm E. Auricular reconstruction for microtia: part II. Surgical techniques. Plast Reconstr Surg. 2002;110(1):234–49.
17. Haisch A, Klaring S, Groger A, et al. A tissue-engineering model for the manufacture of auricular-shaped cartilage implants. Eur Arch Orohinolaryngol. 2002;259:316–21.

18. Liu Y, Zhang L, Zhou G, Li Q, Liu W, et al. In vitro engineering of human ear-shaped cartilage assisted with CAD/CAM technology. Biomaterials. 2009;31:2176–83.
19. Yanaga H, Imai K, Koga M, Yanaga K. Cell-engineered human elastic chondrocytes regenerate natural scaffold in vitro and neocartilage with neoperichondrium in the human body post-transplantation. Tissue Eng Part A. 2012;18(19–20):2020–9.
20. Kobayashi S, Takebe T, Inui M, et al. Reconstruction of human elastic cartilage by a CD44 + CD90 + stem cell in the ear perichondrium. Proc Natl Acad Sci USA. 2011;108(35): 14479–84.
21. Bichara D, O'Sullivan N, Pomerantseva I, et al. The tissue-engineered auricle: past, present, and future. Tissue Eng Part B Rev. 2012;18(1):51–61.
22. Langer R, Vacanti J. Tissue engineering. Science. 1993;260(5110): 920–6.
23. Vacanti C. The history of tissue engineering. J Cell Mol. 2006;10(3):569–76.
24. Bourell D, Beaman J, Leu M, Rosen D. A brief history of additive manufacturing and the 2009 roadmap for additive manufacturing: looking back and looking ahead. Proceedings of RapidTech. 2009:24–5.
25. ••Ventola C. Medical applications for 3D printing: current and projected uses. Pharm Ther. 2014;39(10):704–11.
26. Melchels F, Domingos M, Klein T, et al. Additive manufacturing of tissues and organs. Prog Polym Sci. 2012;37(8):1079–104.
27. •Bogue R. 3D printing: the dawn of a new era in manufacturing? Assem Autom. 2013;33(4):307–11.
28. ••Michalski M, Ross J. The shape of things to come. JAMA. 2014;312(21):2213–4.
29. ••Bauermeister A, Zuriarrain A, Newman M. Three-dimensional printing in plastic and reconstructive surgery: a systematic review. Ann Plast Surg. 2015;77(5):569–76.
30. ••Zhang Y, Oklu R, Dokmeci M, Khademhosseini A. Three-dimensional bioprinting strategies for tissue engineering. Cold Spring Harb Perspect Med. 2017:pii: a025718.
31. ••Reiffel A, Kafka C, Hernandez K, et al. High fidelity tissue engineering of patient specific auricles for reconstruction of pediatric microtia and other auricular deformities. PLoS ONE. 2013;8(2):e56506.
32. ••Jeon B, Lee C, Kim M, et al. Fabrication of three-dimensional scan-to-print ear model for microtia reconstruction. J Surg Res. 2016;206(2):490–7.
33. ••Zhou J, Pan B, Yang Q, et al. Three-dimensional autologous cartilage framework fabrication assisted by new additive manufactured ear-shaped templates for microtia reconstruction. J Plast Reconstr Aesthet Surg. 2016;69(10):1436–44.
34. ••Chen H, NG L, Chang C, et al. Pursuing mirror image reconstruction in unilateral microtia: customizing auricular framework by application of three-dimensional imaging and three-dimensional printing. Plast Reconstr Surg. 2017;139(6):1433–43.
35. Wellisz T. Reconstruction of the burned external ear using a Medpor porous polyethylene pivoting helix framework. Plast Reconstr Surg. 1993;91(5):811–8.
36. ••Constantine K, Gilmore J, Lee K, et al. Comparison of microtia reconstruction outcomes using rib cartilage vs porous polyethylene implant. JAMA Facial Plast Surg. 2014;16(4):240–4.
37. Ciocca L, Scotti R. CAD-CAM generated ear cast by means of a laser scanner and rapid prototyping machine. J Prosthet Dent. 2004;92(6):591–5.
38. ••Im D, Paskhover B, Staffenberg D, Jarrahy R. Current management of microtia: a national survey. Aesthetic Plast Surg. 2013;37(2):402–8.
39. ••Otto I, van Doremalen R, Melchels F, et al. Accurate measurements of the skin surface area of the healthy auricle and skin deficiency in microtia patients. Plast Reconstr Surg Glob Open. 2016;4(12):e1146.
40. Jiao T, Zhang F, Huang X, Wang C. Design and fabrication of auricular prostheses by CAD/CAM system. Int J Prosthodont. 2004;17(4):460–3.
41. Park G, Wiseman J, Clark W. Correction of congenital microtia using stereolithography for surgical planning. Plast Reconstr Surg. 2000;105(4):1444.
42. Walsh W, Reisberg D. A new device for creating and positioning an autogenous cartilage framework during microtia reconstruction. Laryngoscope. 2005;115(11):2068–71.
43. Danter J, Siegert R, Weerda H. 20-MHz ultrasonographical measurement of skin and cartilage thickness in patients with normal and reconstructed external ears. In: Weerda H, Siegert R, editors. Auricular and middle ear malformations, ear defects and their reconstruction. New York: Kugler Publications; 1998.
44. •Shokri T, White D. The Nagata technique for microtia reconstruction. Op Tech Otolaryngol Head Neck Surg. 2017;28(2): 84–9.
45. ••Nuseir A, Hatamleh M, Watson J, et al. Improved construction of auricular prosthesis by digital technologies. J Craniofac Surg. 2015;26(6):e502–5.
46. Federspil P. Ear epistheses as an alternative to autogenous reconstruction. Facial Plast Surg. 2009;25(3):190–203.
47. Tanner P, Mobley S. External auricular and facial prosthetics: a collaborative effort of the reconstructive surgeon and anaplastologist. Facial Plast Surg Clin N Am. 2006;14(2):137–45.
48. Bencharit S. Challenges and prospective applications of extra-oral implants for maxillofacial rehabilitation. Anaplastology. 2012;1:e103.
49. De crescenzio F, Fantini M, Ciocca L, et al. Design and manufacturing of ear prosthesis by means of rapid prototyping technology. Proc Inst Mech Eng H. 2011;225(3):296–302.
50. Cheah C, Chua C, Tan K, Teo C. Integration of laser surface digitizing with CAD/CAM techniques for developing facial prostheses. Part 1: design and fabrication of prosthesis replicas. Int J Prosthodont. 2003;16(4):435–41.
51. Cheah C, Chua C, Tan K. Integration of laser surface digitizing with CAD/CAM techniques for developing facial prostheses. Part 2: development of molding techniques for casting prosthetic parts. Int J Prosthodont. 2003;16(5):543–8.
52. Subburaj K, Nair C, Rajesh S, et al. Rapid development of auricular prosthesis using CAD and rapid prototyping technologies. Int J Oral Maxillofac Surg. 2007;36(10):938–43.
53. Liacouras P, Garnes J, Roman N, et al. Designing and manufacturing an auricular prosthesis using computed tomography, 3-dimensional photographic imaging, and additive manufacturing: a clinical report. J Prosthet Dent. 2011;105(2):78–82.
54. 3-D printed facial prosthesis offers new hope for eye cancer patients follow-ing surgery. http://www.aao.org/news-%20room/news-releases/detail/3d-printed-facial-prosthesis-offers-new-hope-eye-c.
55. •Schubert C vLMDL. Innovations in 3D printing: a 3D overview from optics to organs. Br J Ophthalmol. 2014;98:159–61.
56. ••He Y, Xue G, Fu J. Fabrication of low cost soft tissue prostheses with the desktop 3D printer. Sci Rep. 2014;4:6973.
57. •Watson J, Hatamleh M. Complete integration of technology for improved reproduction of auricular prostheses. J Prosthet Dent. 2014;111(5):430–6.
58. Cao Y, Vacanti J, Paige K, et al. Transplantation of chondrocytes utilizing a polymer-cell construct to produce tissue-engineered cartilage in the shape of a human ear. Plast Reconstr Surg. 1997;100(2):297–302.
59. Chu C, Coutts R, Yoshioka M, et al. Articular cartilage repair using allogeneic perichondrocyte-seeded biodegradable porous

polylactic acid (PLA): a tissue-engineering study. J Biomed Mater Res Part A. 1995;29(9):1147–54.
60. Wayne J, McDowell C, Sheilds K, Tuan R. In vivo response of polylactic acid-alginate scaffolds and bone marrow-derived cells for cartilage tissue engineering. Tissue Eng. 2005;11(5–6):953–63.
61. Moran J, Pazzano D, Bonassar L. Characterization of polylactic acid-polyglycolic acid composites for cartilage tissue engineering. Tissue Eng. 2004;9(1):63–70.
62. Jeon Y, Choi J, Sung J, et al. Different effects of PLGA and chitosan scaffolds on human cartilage tissue engineering. J Craniofac Surg. 2007;18(6):1249–58.
63. ••Zopf D, Mitsak A, Flanagan C, et al. Computer aided-designed, 3-dimensionally printed porous tissue bioscaffolds for craniofacial soft tissue reconstruction. Otolaryngol Head Neck Surg. 2015;152(1):57–2.
64. ••Xu T, Binder K, Albanna M, et al. Hybrid printing of mechanically and biologically improved constructs for cartilage tissue engineering applications. Biofabrication. 2013;5(1):015001.
65. ••Zheng R, Duan H, Xue J, et al. The influence of Gelatin/PCL ratio and 3-D construct shape of electrospun membranes on cartilage regeneration. Biomaterials. 2014;35(1):152–64.
66. Nayyer L, Patel K, Esmaeili A, et al. Tissue engineering: revolution and challenge in auricular cartilage reconstruction. Plast Reconstr Surg. 2012;129(5):1123–37.
67. Nayyer L, Birchall M, Seifalian A, Jell G. Design and development of nanocomposite scaffolds for auricular reconstruction. Nanomedicine. 2014;10(1):235–46.
68. ••Nayyer L, Jell G, Esmaeili A, et al. A biodesigned nanocomposite biomaterial for auricular cartilage reconstruction. Adv Healthc Mater. 2016;5(10):1203–12.
69. •Bos E, Doerga P, Breugem C, Van zuijlen P. The burned ear; possibilities and challenges in framework reconstruction and coverage. Burns. 2016;42(7):1387–95.
70. ••Cohen B, Hooper R, Puetzer J, et al. Long term morphological and microarchitectural stability of tissue engineered, patient-specific auricles. Tissue Eng Part A. 2016;22(5–6):461–8.
71. ••Zhao X, Bichara D, Zhou L, et al. Conditions for seeding and promoting neo-auricular cartilage formation in a fibrous collagen scaffold. J Cranio-Maxillofac Surg. 2015;43(3):382–9.
72. ••Rhee S, Puetzer J, Mason B, et al. 3D Bioprinting of spatially heterogeneous collagen constructs for cartilage tissue engineering. ACS Biomater Sci Eng. 2016;2(10):1800–5.
73. Mannoor M, Jiang Z, James T, et al. 3D printed bionic ears. Nano Lett. 2013;13(6):2634–9.
74. Fragonas E, Valente M, Pozzi-Mucelli M, et al. Articular cartilage repair in rabbits by using suspensions of allogenic chondrocytes in alginate. Biomaterials. 2000;21(8):795–801.
75. •Rami M, Aditya K, Gemma P, et al. Chondrocyte culture in three dimensional alginate sulfate hydrogels promotes proliferation while maintaining expression of chondrogenic markers. Tissue Eng Part A. 2014;20(9–10):1454–64.
76. Lin Y, Yen C, Wu Y, et al. Chondrocytes culture in three-dimensional porous alginate scaffolds enhanced cell proliferation, matrix synthesis and gene expression. J Biomed Mater Res Part A. 2008;88A(1):23–33.
77. Lahiji A, Sohrabi A, Hungerford D, Frondoza D. Chitosan supports the expression of extracellular matrix proteins in human osteoblasts and chondrocytes. J Biomed Mater Res. 2000;51(4):586–95.
78. Xia W, Liu W, Cui L, et al. Tissue engineering of cartilage with the use of chitosan-gelatin complex scaffolds. J Biomed Mater Res B. 2004;71B(2):373–80.
79. Bittencourt R, Pereira H, Felisbino S, et al. Chondrocyte cultures in tridimensional scaffold: alginate hydrogel. Acta Ortopedica Brasileira. 2009;17(4):242–5.
80. Zhang Z, Ortiz O. Biomaterials in tissue engineering: biodegradable polymers. In: Lanza R, Langer R, Vacanti J, editors. Principles of tissue engineering. 4th ed. Amsterdam: Elsevier Inc; 2014.
81. ••Gasperini L, Mano J, Reis R. Natural polymers for the microencapsulation of cells. J R Soc Interface. 2014;11(100):20140817.
82. ••Cohen B, Bernstein J, Morrison K, et al. Tissue engineering the human auricle by auricular chondrocyte-mesenchymal stem cell co-implantation. Sci Transl Med. 2017; Submitted Nov 10, 2017.
83. Khan I, Francis L, Theobald P, et al. In vitro growth factor-induced bio engineering of mature articular cartilage. Biomaterials. 2013;34(5):1478–87.
84. Khan I, Evans S, Young R, et al. Fibroblast growth factor 2 and transforming growth factor β1 induce precocious maturation of articular cartilage. Arthritis Rheum. 2011;63(11):3417–27.
85. Shim J, Kim J, Park M, et al. Development of a hybrid scaffold with synthetic biomaterials and hydrogel using solid freeform fabrication technology. Biofabrication. 2011;3(3):034102.
86. Visser J, Peters B, Burger T, et al. Biofabrication of multi-material anatomically shaped tissue constructs. Biofabrication. 2013;5(3):035007.
87. Lee H, Ahn S, Bonassar L, et al. Cell(MC3T3-E1)-printed poly (ε-caprolactone)/alginate hybrid scaffolds for tissue regeneration. Macromol Rapid Commun. 2013;34(2):142–9.
88. ••Bernstein J, Cohen B, Harper A, et al. Abstract: scaffolding the scaffold: 3D-printed external biodegradable cage mitigates contraction during maturation of elastic cartilage constructs. Plast Reconstr Surg Glob Open. 2017;5(9 Suppl).
89. ••Markstedt K, Mantas A, Tournier I, et al. 3D bioprinting human chondrocytes with nanocellulose-alginate bioink for cartilage tissue engineering applications. Biomacromolecules. 2015;16(5):1489–96.
90. ••Park J, Choi Y, Shim J, et al. Development of a 3D cell printed structure as an alternative to autologs cartilage for auricular reconstruction. J Biomed Mater Res Part B. 2016;105(5):1016–28.
91. ••Lee J, Hong J, Jung J, et al. 3D printing of composite tissue with complex shape applied to ear regeneration. Biofabrication. 2014;6(2):024103.
92. ••Morris V, Nimbalkar S, Younesi M, et al. Stereolithography: mechanical properties, cytocompatibility and manufacturability of chitosan: PEGDA hybrid-gel scaffolds by stereolithography. Ann Biomed Eng. 2017;45(1):286–96.