PERSPECTIVE

Opportunities and challenges in modeling human brain disorders in transgenic primates

Charles Jennings[1,2], Rogier Landman[1–3], Yang Zhou[1,2], Jitendra Sharma[2,4], Julia Hyman[1,2], J Anthony Movshon[5], Zilong Qiu[6], Angela Roberts[7], Anna Wang Roe[8], Xiaoqin Wang[9], Huihui Zhou[10], Liping Wang[11], Feng Zhang[1,2,12], Robert Desimone[1,2] & Guoping Feng[1–3]

Molecular genetic tools have had a profound impact on neuroscience, but until recently their application has largely been confined to a few model species, most notably mouse, zebrafish, *Drosophila melanogaster* and *Caenorhabditis elegans*. With the development of new genome engineering technologies such as CRISPR, it is becoming increasingly feasible to apply these molecular tools in a wider range of species, including nonhuman primates. This will lead to many opportunities for brain research, but it will also pose challenges. Here we identify some of these opportunities and challenges in light of recent and foreseeable technological advances and offer some suggestions. Our main focus is on the creation of new primate disease models for understanding the pathological mechanisms of brain disorders and for developing new approaches to effective treatment. However, we also emphasize that primate genetic models have great potential to address many fundamental questions about brain function, providing an essential foundation for future progress in disease research.

Brain disorders: an unmet need

Brain disorders are among the largest causes of disease burden world-wide[1], affecting millions of people and imposing enormous societal and economic costs[2]. Many of these disorders are chronic and incurable conditions, for which existing treatment options are inadequate and in some cases almost completely ineffective. Yet despite the urgent clinical need, there has been little recent progress in the development of new treatments for most common brain disorders, and many currently prescribed drugs are based on decades-old science[3]. This may seem surprising given the rapid rate of progress in fundamental neuroscience, but it has proven extraordinarily difficult to translate advances in basic science into the development of new and better clinical treatments[4]. The failure rates for experimental CNS drugs are very high[5,6], leading many pharmaceutical companies to disinvest in brain disorders research and focus their research and development efforts elsewhere[7].

Many reasons have been identified for this disappointingly slow progress, but one of the most important is the lack of good animal models[4,8–12]. Extrapolation from animal models to human patients is always uncertain, but this is especially true for brain disorders given

the profound differences in brain and behavior between humans and the rodent species that are commonly used as preclinical disease models. Among brain disorders, psychiatric diseases present a particular challenge given that they are diagnosed purely through behavioral symptoms that are difficult or impossible to model in rodents. The lack of good animal models for these complex diseases poses a challenge for understanding fundamental pathological mechanisms, for discovering potential drug targets, for identifying biomarkers of disease progression or treatment response, and for development and preclinical testing of new treatments.

The emergence of new transgenic technologies has led to growing interest in the use of nonhuman primates to study diseases that are difficult to model in rodents. Primates (mainly macaques and marmosets) are already widely used in pharmaceutical research, mainly for pharmacokinetic and toxicology studies that precede human clinical trials[13]. However, they are rarely used for preclinical studies of efficacy, mainly because there are few validated primate models of CNS disorders. The ability to create targeted genomic alterations in primates, combined with advances in human disease genetics, now promises to change this picture and, as we argue below, could greatly improve our ability to develop new treatments for these previously intractable conditions.

New technologies for primate research

Transgenic macaques were first reported 15 years ago[14], and an overexpression model of Huntington's disease was described in 2008 (ref. 15). These pioneering efforts, followed by the demonstration of germline transmission in transgenic marmosets[16], generated considerable interest in the possibility of modeling genetic diseases in nonhuman primates[17] and helped stimulate major investments in primate research by Japan[18] and China[19]. Despite these advances, however, the widespread adoption of transgenic primate models appeared impractical until recently, given the paucity of methods for making precise genetic changes in primate embryos[17]. The situation changed with the development of TALEN- and CRISPR-mediated genome editing in eukaryotic cells[20–22] and with subsequent demonstrations that these programmable nucleases could be used to make genetic changes in embryos from many species[23,24], including nonhuman primates[25–29] and nonviable human embryos[30]. Genome editing methods are now advancing rapidly and promise to revolutionize many areas of biomedical research, including the generation of animal models for human genetic disease.

Other technical advances are also converging to create new opportunities for the creation and analysis of transgenic primate models.

Perhaps most importantly, advances in human genomic research are now enabling the rapid discovery of genetic risk factors for common brain disorders, including autism, schizophrenia, bipolar disorder, Alzheimer's disease and Parkinson's disease, among others. Thanks to large-scale consortium efforts involving tens of thousands of samples, many of these associations can be assigned with high confidence[31–33], providing an essential foundation for the creation of genetic models that reflect human disease mechanisms.

Other important advances include new viral vectors with potential applications in primate research[34,35], as well as human gene therapy[36]; development of genetic activity reporters and optogenetic effectors, some of which have already been applied in primates[37,38]; scalable single-cell transcriptional profiling[39,40], including methods applicable to human post-mortem brain tissue[41], which will enable cross-species comparisons of neural populations at single-cell resolution; advances in automated behavioral analysis[42]; new methods for high-field magnetic resonance imaging of primate brains[43,44]; and development of chronic implantable electrode arrays[45] and telemetric methods for wireless electrophysiology[46–48]. Taken together, this confluence of technological advances will lead to new opportunities to produce genetically modified primates and to analyze the resulting phenotypes at multiple levels from molecules to cells, circuits and behavior[49].

Despite these advances, however, the creation and analysis of transgenic primate models represents a major technical challenge, requiring large investments of time and funding and raising important ethical questions around the justification of such work. It is therefore important to carefully consider when, why and how such projects should be attempted.

When to use primates

Basic and translational neuroscience has made, and will continue to make, great progress by studying mice and other simpler organisms. Much can also be learned by studying humans directly, and technological advances in areas such as neuroimaging, genomics and induced pluripotent stem (iPS) cells are allowing human disease researchers to address questions that were previously restricted to experimental animals. Nonhuman primate research will in no way replace these approaches, and, for both ethical and practical reasons, primate genetic models should only be considered where other alternatives are not available.

Several recent reviews have discussed the potential impact of genetically modified primates in neuroscience[17,28,29,49–52], and in particular there is growing interest in the marmoset as a genetic model for disease research[53] and for basic research in areas such as audition[54,55], vision[56], emotion regulation[57] and social neuroscience[58]. Other primate-related species under consideration include mouse lemurs and tree shrews (*Tupaia glis*, not to be confused with the unrelated Etruscan shrew). Our goal here is not to review the pros and cons of these different species, but rather to highlight some applications for which primate models may be critical for the advancement of the field. The advantages for basic neuroscience have been highlighted by others, and here we focus primarily on the potential applications for diseases of the CNS, including developmental, neurological and psychiatric disorders.

One specific challenge in modeling brain disorders is that many of the clinical symptoms involve higher cognitive functions that are controlled by the prefrontal cortex (PFC), which is much less developed in rodents than in primates[59] and which also shows many differences in gene expression between the two taxa[60]. Thus, although cognitive processes such as executive function, decision making, attentional control or working memory can be studied in rodents, the organization and integration of these processes within the PFC may differ from primates. For example, although rodents can perform working memory tasks, it is unclear whether they show the type of delay activity that is believed to underlie working memory in the primate PFC (ref. 61). Such differences may contribute to the difficulty of modeling deficits of working memory in conditions such as fragile X syndrome, in which mutant mice have failed to reproduce key cognitive features of the human disorder despite the well-established genetic etiology[62].

Another challenge arises with modeling social behavior, which is disrupted in many psychiatric diseases and which differs profoundly between primates and rodents[58]. The differences may have deep evolutionary roots, since it has been argued that group living is an ancestral trait that emerged early in primate evolution[63], before the divergence of Old and New World primates but millions of years after the divergence of primates and rodents. Assuming that many primate social behaviors reflect adaptations to group living, we would predict that these behaviors depend on neural systems that are conserved among primates but are likely to diverge from those of rodents. This would limit the utility of rodent models for disorders such as autism, in which disruption of social behavior is a primary feature.

The neural substrates of social behavior are not well defined, but one important modulator is believed to be the neuropeptide oxytocin. While a role of oxytocin in affiliative and reproductive behavior is probably common to all vertebrates[64] (and may even be shared with invertebrates such as nematodes[65]), the detailed organization of the primate oxytocin system differs substantially from that of rodents[66]. There has been much interest in oxytocin signaling as a possible drug target for the treatment of autism and other psychiatric disorders, and primate models would seem inherently preferable to rodent models for preclinical evaluation of such approaches.

Many of the drugs used to treat CNS disorders exert their effects via neurotransmitter and neuromodulatory systems, and, as with the oxytocin example, many of these systems may differ between primates and rodents. Another prominent example is the α7-nicotinic acetylcholine receptor, widely studied as a target for treating the cognitive symptoms of Alzheimer's disease, attention deficit–hyperactivity disorder and schizophrenia[67]. Again, the distribution of this receptor differs between primates and rodents[68], and, while the significance of these differences is unknown, the importance of understanding the role of α7 signaling in the primate brain is underlined by the recent clinical failure in two large phase III trials of encenicline, an α7 agonist that was under development by the now-defunct Forum Pharmaceuticals as a treatment for schizophrenia[69].

In addition to specific receptor systems, there are also many differences in connectivity between primate and rodent brains. To cite one well-known example, rodents lack the direct cortico–motor neuron projections that support fine control of forelimb movement[70], a characteristic ability of primates that is impaired in many neurological conditions. There are also differences in cell types between primate and rodent brains; notably, rodents appear to lack von Economo neurons, a specialized cell type present in PFC that is implicated in social cognition and subject to deterioration in dementia[71]. There are also substantial differences between rodents and primates with respect to the scaling relationships between brain size and cell number[72] and connectivity[73]. Whether these factors are significant for disease models is not known, but it seems plausible that neurodegenerative conditions such as Alzheimer's disease (which appears to spread through the brain via mechanisms that are not yet well understood[74]) will be substantially affected by parameters such as neuronal density and connectivity. In this context it is noteworthy that it has been difficult to generate realistic models of Alzheimer's disease in mice[75] and that experimental

drugs for Alzheimer's disease have had extraordinarily high failure rates in human clinical trials[76]. It seems plausible that genetic primate models may provide a more realistic model for understanding and preventing the development of Alzheimer-like pathology, a possibility that is supported by a recent report that injection of amyloid-β oligomers induces an Alzheimer-like pathology in macaque but not rat brains[77].

One advantage of primate disease models will be the ability to study developmental phenotypes and prodromal disease stages, which are difficult to study in human patients yet critical for understanding the mechanism of disease onset and for identifying opportunities for early intervention. Rodent models are not ideal for this purpose given that primate brain development is much slower than that of rodents and shows a number of distinctive features. Prenatally, primate neurogenesis is characterized by an expanded subventricular zone with a complex population of progenitor cells[78], which give rise over a prolonged period to the large primate cortex[79]. Postnatally, primate development involves a long juvenile phase during which the brain is extensively modified through experience-dependent plasticity and learning, including strong effects of social learning via parent–offspring interactions. Human developmental disorders are therefore likely to involve complex cascade effects in which early deficits impair the brain's ability to undergo subsequent experience-dependent changes[80]. Primate models seem better suited than rodent models for understanding the abnormal developmental processes that lead to the clinical manifestations of these disorders and for identifying potential therapeutic interventions.

As a final point, primate models may be essential to understanding the many risk variants for psychiatric and other disorders that occur within noncoding regions of the genome. These variants can affect cell-type-specific promoters, enhancers, introns, noncoding RNAs or intragenic regions[81,82], which, unlike coding sequences, are often highly divergent between rodents and primates[83]. These divergences presumably explain the different patterns of gene expression that have been described in the neocortex of primates and rodents[84] and whose significance for disease remains to be explored.

Ethical issues

The scientific case for studying nonhuman primates is based on their similarities to humans, but it is widely recognized that these similarities also raise ethical issues that go beyond the general '3R' imperative to replace, reduce and refine the use of animals in research. A recent report commissioned by several major UK funding agencies[85] argued that evaluation of any proposed primate research project should consider four factors: quality and importance of the science, likelihood of medical or other public benefit, likelihood of animal suffering, and availability of alternatives. Although not addressed in the report, these criteria seem equally applicable to studies involving transgenic or mutant animals.

When considering these ethical issues, it is useful to distinguish between different types of transgenic studies. In mice, and potentially in primates, many questions about brain function can be addressed in transgenic knock-in lines expressing genetically encoded reporters (for example, calcium indicators or trans-synaptic tracers) or effectors (for example, channelrhodopsin or Cre recombinase) under the control of an endogenous promoter, allowing monitoring and manipulation of activity in specific subsets of cells. Such studies, while they will need to be carefully evaluated, do not appear to raise new ethical issues beyond those that arise with all primate neuroscience research.

Additional considerations arise, however, with studies that involve animals in which normal gene function has been disrupted to address questions about basic brain function or to model specific human diseases.

First, such studies, which involve comparisons between groups, will require larger numbers of animals than studies of normal brain function. Projects will need to be designed carefully to avoid using more animals than necessary (and to minimize the production of superfluous animals with unwanted genotypes), but they must also avoid using too few animals, since (as with human clinical trials[86]) it is also unethical to use sample sizes that are inadequate to provide statistically valid conclusions.

Second, disease models (genetic or otherwise) necessarily involve a risk of suffering. Animal created for this purpose will require careful veterinary oversight, including evaluation of behavioral as well as physical well-being, along with the capacity for skilled veterinary intervention where needed. Researchers and veterinarians will need to work together to develop strict and standardized guidelines for when and how to intervene in such cases.

Third, the development of primate genetic models will be an international effort, involving many countries with different cultural traditions and public attitudes toward animal research. It will be important to establish shared standards and regulations and to assure all stakeholders that work is performed to the highest standards of animal welfare regardless of where it is conducted.

Finally, there is an ethical obligation to use animal resources wisely, minimizing unnecessary duplication of effort and maximizing the benefit obtained from each animal by sharing data and (where applicable) cell and tissue samples. This of course applies to all animal research, but it is especially so for primate research given the high costs, the long lead times and the need to minimize the numbers of animals used. Achieving this will require coordination at national and international levels, as we discuss in the final section of this article.

Methods for creating transgenic primates

Early efforts to model genetic disorders in primates have typically relied on overexpression of randomly inserted transgenes, but the development of programmable nucleases, including ZFN, TALEN, Cas9 and other CRISPR-associated endonucleases has enabled the insertion of mutations at specific genomic target sites. These groundbreaking advances have been widely reviewed elsewhere, and their application to the nonhuman primate is summarized in **Box 1**.

Two key challenges stand out. First, all genetically modified primates reported to date have been produced through direct manipulation of embryos, which can lead to mosaicism if mutations arise after replication of the zygotic genome[27,87]. Possible solutions are noted in **Box 1**, but until this problem is overcome it seems likely that preliminary analysis will need to be performed on founder animals whose mosaicism is not fully characterized (especially in the brain, where biopsies are difficult) and that fuller characterization will rely on breeding from the founders. This is routine in mouse research, but for primates, with longer generation times, it will impose significant delays. It will therefore be desirable to accelerate the production of offspring using established and emerging reproductive technologies (for example, testis xenografting[88] or cultured spermatogonial stem cells[89–91]). Breeding strategies will also need to be carefully designed to avoid problems with inbreeding, given that lab primates (unlike rodents) are not inbred and are therefore likely to carry deleterious recessive mutations.

A second challenge is the creation of animals with multiple parallel mutations. In mice this is accomplished by crossing individual lines but in primates this would seem impractical for time and cost reasons. Solving this challenge will require significant technical advances, perhaps including the production of cloned animals by nuclear transfer from cultured cell lines[92].

Box 1 Methods for creating genetically modified primates

Viral vectors

Mechanism. A transgene is delivered via a retroviral vector, which is injected into the perivitelline space of the oocyte or early embryo and integrates randomly into the host genome.

Examples. Genes encoding huntingtin in Huntington's disease[15,113] or α-synuclein in Parkinson's disease[114]; *MECP2* duplication syndrome[87].

Advantages. Ease of delivery; this technique is the first method to be used successfully in nonhuman primates[14].

Limitations. The amount and distribution of transgene expression are not controlled; resulting phenotypes may not reflect disease mechanisms; and multiple random insertions are possible and can segregate differently in offspring[87].

Programmable nucleases

Mechanism. A nuclease is targeted to a specific DNA sequence by engineering either the protein or, for CRISPR, an associated guide RNA. Reagents are injected into the early embryo, causing cleavage of the targeted genomic sequence. Mutations can arise at the cleavage site by either nonhomologous end-joining (NHEJ) or homology-directed repair (HDR) as described below (reviewed in ref. 115).

Examples. ZFN, TALEN, Cas9 and other CRISPR-associated endonucleases.

Advantages. This technique can target specific locations, is fast and flexible (especially CRISPR) and can target multiple gene loci in parallel[24,116]. The rapid pace of progress means key technical obstacles may soon be solved.

Limitations. Off-target effects remain a concern, although methods to improve specificity are being developed[117,118]. Another concern is mosaicism; this might be reduced by injecting Cas9 protein instead of mRNA, eliminating the translation delay and increasing the chance of mutations before genome replication, but this approach has not yet been validated.

NHEJ pathway

Mechanism. Following targeted DNA cleavage, double-stranded breaks are rejoined, usually with random insertions or deletions that can lead to frameshifts or other functional disruptions.

Advantages. Can produce loss-of-function mutations with high efficiency; proven to work in a variety of species

Limitations. Mutations are random and effects not always predictable; it may be difficult or impossible to create gain-of-function mutations.

HDR pathway

Mechanism. Following targeted DNA cleavage, the cut ends recombine with an exogenously supplied DNA template, replacing the original sequence with that of the template.

Advantages. This technique can introduce precise changes and can mimic human risk variants or introduce reporter and/or effector genes under control of endogenous promoters (knock-in).

Limitations. Efficiency of transgene delivery is lower than via the NHEJ pathway, requiring larger numbers of donor embryos and surrogate mothers. It has been reported in cynomolgus embryos[119] but no live births have yet been reported in primates.

Future possibilities

Preimplantation genetic screening. The ability to identify embryos with the desired genotype before implantation would reduce both the number of surrogate mothers needed and the production of superfluous offspring. Genetic testing of individual blastomeres is an established method in human *in vitro* fertilization, and similar methods should be feasible for nonhuman primate embryos.

ES or iPS cells. It may be possible to create defined mutations in cultured embryonic stem or iPS cells that would then be incorporated into a chimeric embryo, including (ideally) germline. The method is standard in mice but has not yet been successful in primates[120].

Nuclear transfer. It may be possible to establish genotypes in cultured cells before creation of animals. Unlike other methods, nuclear transfer could enable single-step generation of cloned animals with identical genotypes including multiple parallel mutations. Live births have been obtained in other mammalian species but not yet in primates.

Somatic cell gene targeting. Cas9 can be delivered to the brain using a viral vector[121], or it can be expressed ubiquitously via a germline insertion and locally targeted to a desired gene using virally delivered guide RNA[122]. This has been done in mice and may be possible in primates using methods already available.

Other CRISPR-based approaches. In addition to its natural endonuclease activity, Cas9 is a versatile platform that can be engineered to activate or repress transcription[123,124], to induce targeted chromatin modification[125] or to chemically convert DNA sequences[126]. Another CRISPR-based nuclease, C2c2, may allow direct targeting of RNA instead of DNA[127]. These and other CRISPR-based methods are evolving rapidly and may lead to many future applications in primate neuroscience.

Choice of disease models

Methods for producing and analyzing mouse knockout phenotypes have become very efficient, allowing systematic evaluation of large numbers of potential disease models[93]. This will not be possible in primates, given the ethical and logistical constraints, and it will therefore be important to prioritize the creation of relatively few transgenic lines with the greatest potential to advance understanding of brain function and disease mechanisms. In choosing which genes to target, specific criteria might include expectation of a clear and penetrant phenotype, expectation of construct validity (i.e., mechanistic similarity of the mutation to a human variant with a well-established link to disease), and reason to think mouse models will be inadequate (as has been the case, for example, with mouse models of Alzheimer's disease[76], amyotrophic lateral sclerosis[94] and fragile X syndrome[95,96]).

In the near term, the choice of primate models will also be constrained by the efficiency of the available technologies, as described above (**Box 1**) and as also recently discussed by others[52]. The diseases most readily modeled with current methods are those in which a null mutation in a single allele leads to a penetrant phenotype, either because of haploinsufficiency (e.g., Shank3 and autism) or because the gene is X-linked. Examples of the latter include *FMRP* in fragile X syndrome, *MECP2* (although many clinical features of *MECP2* loss and duplication have been successfully modeled in mice[97]), and dystrophin (*DMD*) in Duchenne muscular dystrophy[27]. An additional attraction of these targets is that they lead to early developmental effects in human patients and may therefore provide early proof of concept for the use of primate genetic models.

Gain-of-function mutations and partial loss-of-function mutations are more difficult to model because they will in most cases require the knock-in of a precise sequence change within an endogenous gene, via homology-directed repair. Prominent examples include Huntington's disease, amyotrophic lateral sclerosis and Alzheimer's disease, and

program that will take many years to reach fruition. In addition to the technological challenges identified above, there will be many other logistical obstacles along the way, which we believe can only be overcome through a concerted international effort.

First and foremost, primate research is difficult and labor-intensive, and becoming expert in the necessary techniques represents a major career commitment. Even after a researcher has achieved proficiency, the collection of data from a single animal often requires months or years of work (especially if it involves behavioral training or electrophysiological recording). Genetic research will require larger numbers of animals, and obtaining data at the necessary scale will require new approaches. For example, it will probably be necessary to develop automated methods for behavioral training and monitoring, including methods that can be deployed in the home-cage environment. The development of new chronic electrode recording methods will also be important, especially if it can reduce the effort and risk associated with repeated surgeries for acute recordings.

The production and study of genetically modified primates will require an interdisciplinary combination of expertise that is beyond the capacity of any individual lab. Relevant skills include molecular genetics, reproductive physiology, primate husbandry and veterinary care, behavior, surgery, electrophysiology, neuroanatomy, histology, clinical and translational neuroscience, computational neuroscience and big data analysis. Developing the necessary depth of expertise in all these areas is beyond the means of all but the largest institutions, and collaboration between institutions (including international collaboration) will therefore be essential. This in turn requires infrastructure for distributing materials and data sets, funding structures for sharing costs, and a culture in which researchers (especially younger researchers) can achieve career advancement through contributions to team projects that unfold over long periods compared to the duration of a graduate thesis or postdoctoral fellowship.

To maximize the benefits from valuable transgenic lines it will be important to share them between laboratories. This is routine for transgenic mice, but transporting primates poses several challenges[109]. It is possible in principle to ship frozen sperm or embryos, but this requires local capacity to re-derive animals and would involve long delays. It will probably be necessary to establish an international network of primate centers and commercial vendors, to serve as repositories and distributors for valuable strains.

It is of course easier to transport humans than other primates, and therefore another solution is to create 'hotel space' at major primate research centers, where visiting researchers can spend time carrying out experiments, perhaps along the lines of the summer research programs at Marine Biological Laboratory (MBL) at Woods Hole, Massachusetts. This will probably require specific funding mechanisms for travel and for cost-sharing with home institutions. For long-term studies, it may be necessary to develop telecommuting approaches involving collaboration with expert staff on site, perhaps combined with automated methods for behavioral training and data collection.

In addition to studying live animals, much of the characterization of transgenic lines will involve studies on tissue samples and cell cultures, which can be shipped to laboratories with the necessary expertise (including many which do not have the capacity to house live primates). It would seem desirable to create a centralized tissue and cell bank to distribute these materials.

The field will also depend on shared databases for commonly used species such as macaques and marmosets. These are likely to include standard atlases and areal definitions, connectomic data, genomic and gene expression data, and normative developmental trajectories. They may also include large electrophysiological and behavioral data sets, along

with breeding and rearing histories for individual animals. Curating and distributing these diverse data formats will require the development of appropriate computational platforms, along with community standards for sharing data across multiple collaborating groups.

We suspect that all of these solutions will be needed, along with others yet to be identified. We note that although the challenges of primate genetic research are unusual within the biomedical field, analogous issues arise in other scientific disciplines that depend on multinational collaborations and large core facilities—for example, astronomy or particle physics—and these may hold useful lessons for primate research.

CONCLUSIONS

Advances in genome editing are happening very fast, providing grounds for optimism that techniques that are speculative today will be commonplace in the near future. Even so, primate research will remain expensive and logistically challenging, and efforts to develop new models will take years to come to fruition. But the costs are modest compared to the large sums that have been spent on unsuccessful clinical trials, and they pale into insignificance compared to the vast global economic cost of brain disorders, which is estimated in the trillions of dollars[110–112]. Whether primate models will lead to new treatments is of course unknowable, but given the dismal record of drug development for neurological and psychiatric disease over the past several decades, we believe there is a strong case for trying new approaches, and that the ethical concerns around animal research must be weighed against the ethical obligation toward millions of current and future patients to seek better treatments for conditions that are as yet incurable. We argue that basic neuroscience has failed to deliver substantially new and effective treatments for many brain disorders, partially because the animal modeling was done in species whose brains are too dissimilar from those of humans. Genetic primate models offer new hope that deserves investment if we are serious about doing the best we can to improve human health worldwide.

AUTHOR CONTRIBUTIONS
C.J. and R.L. wrote the first draft of the manuscript. All authors reviewed the manuscript and participated in discussions and development of ideas.

COMPETING FINANCIAL INTERESTS
The authors declare no competing financial interests.

Reprints and permissions information is available online at http://www.nature.com/reprints/index.html.

1. Murray, C.J.L. *et al.* Disability-adjusted life years (DALYs) for 291 diseases and injuries in 21 regions, 1990-2010: a systematic analysis for the Global Burden of Disease Study 2010. *Lancet* **380**, 2197–2223 (2012).

2. Roehrig, C. Mental disorders top the list of the most costly conditions in the United States: $201 billion. *Health Aff. (Millwood)* **35**, 1130–1135 (2016).
3. Hyman, S.E. Revolution stalled. *Sci. Transl. Med.* **4**, 155cm11 (2012).
4. Gribkoff, V.K. & Kaczmarek, L.K. The need for new approaches in CNS drug discovery: why drugs have failed, and what can be done to improve outcomes. *Neuropharmacology* http://dx.doi.org/10.1016/j.neuropharm.2016.03.021 (2016).
5. Kaitin, K.I. & DiMasi, J.A. Pharmaceutical innovation in the 21st century: new drug approvals in the first decade, 2000–2009. *Clin. Pharmacol. Ther.* **89**, 183–188 (2011).
6. Amplion. Clinical Development Success Rates 2006–2015 http://www.amplion.com/clinical-development-success-rates (2016).
7. Fidler, B. After failed schizophrenia trial, Forum Pharma to shutter this week. *Xconomy* http://www.xconomy.com/boston/2016/06/27/after-failed-schizophrenia-trial-forum-pharma-to-shutter-this-week/ 2016).
8. Pangalos, M.N., Schechter, L.E. & Hurko, O. Drug development for CNS disorders: strategies for balancing risk and reducing attrition. *Nat. Rev. Drug Discov.* **6**, 521–532 (2007).
9. Nestler, E.J. & Hyman, S.E. Animal models of neuropsychiatric disorders. *Nat. Neurosci.* **13**, 1161–1169 (2010).
10. Fernando, A.B.P. & Robbins, T.W. Animal models of neuropsychiatric disorders. *Annu. Rev. Clin. Psychol.* **7**, 39–61 (2011).
11. Sarter, M. & Tricklebank, M. Revitalizing psychiatric drug discovery. *Nat. Rev. Drug Discov.* **11**, 423–424 (2012).
12. Institute of Medicine (US) Forum on Neuroscience and Nervous System Disorders. *Improving the Utility and Translation of Animal Models for Nervous System Disorders: Workshop Summary* (National Academies Press, 2013).
13. Bluemel, J., Korte, S., Schenck, E. & Weinbauer, G. (eds.) *The Nonhuman Primate in Nonclinical Drug Development and Safety Assessment* 1st edn. (Elsevier, 2016).
14. Chan, A.W., Chong, K.Y., Martinovich, C., Simerly, C. & Schatten, G. Transgenic monkeys produced by retroviral gene transfer into mature oocytes. *Science* **291**, 309–312 (2001).
15. Yang, S.-H. *et al.* Towards a transgenic model of Huntington's disease in a non-human primate. *Nature* **453**, 921–924 (2008).
16. Sasaki, E. *et al.* Generation of transgenic non-human primates with germline transmission. *Nature* **459**, 523–527 (2009).
17. Chan, A.W.S. Progress and prospects for genetic modification of nonhuman primate models in biomedical research. *ILAR J.* **54**, 211–223 (2013).
18. Okano, H., Miyawaki, A. & Kasai, K. Brain/MINDS: brain-mapping project in Japan. *Phil. Trans. R. Soc. Lond. B* http://dx.doi.org/10.1098/rstb.2014.0310 (2015).
19. Cyranoski, D. Monkey kingdom. *Nature* **532**, 300–302 (2016).
20. Miller, J.C. *et al.* A TALE nuclease architecture for efficient genome editing. *Nat. Biotechnol.* **29**, 143–148 (2011).
21. Cong, L. *et al.* Multiplex genome engineering using CRISPR/Cas systems. *Science* **339**, 819–823 (2013).
22. Mali, P. *et al.* RNA-guided human genome engineering via Cas9. *Science* **339**, 823–826 (2013).
23. Carlson, D.F. *et al.* Efficient TALEN-mediated gene knockout in livestock. *Proc. Natl. Acad. Sci. USA* **109**, 17382–17387 (2012).
24. Wang, H. *et al.* One-step generation of mice carrying mutations in multiple genes by CRISPR/Cas-mediated genome engineering. *Cell* **153**, 910–918 (2013).
25. Niu, Y. *et al.* Generation of gene-modified cynomolgus monkey via Cas9/RNA-mediated gene targeting in one-cell embryos. *Cell* **156**, 836–843 (2014).
26. Liu, H. *et al.* TALEN-mediated gene mutagenesis in rhesus and cynomolgus monkeys. *Cell Stem Cell* **14**, 323–328 (2014).
27. Chen, Y. *et al.* Functional disruption of the dystrophin gene in rhesus monkey using CRISPR/Cas9. *Hum. Mol. Genet.* **24**, 3764–3774 (2015).
28. Sasaki, E. Prospects for genetically modified non-human primate models, including the common marmoset. *Neurosci. Res.* **93**, 110–115 (2015).
29. Whitelaw, C.B.A., Sheets, T.P., Lillico, S.G. & Telugu, B.P. Engineering large animal models of human disease. *J. Pathol.* **238**, 247–256 (2016).
30. Liang, P. *et al.* CRISPR/Cas9-mediated gene editing in human tripronuclear zygotes. *Protein Cell* **6**, 363–372 (2015).
31. Willsey, A.J. & State, M.W. Autism spectrum disorders: from genes to neurobiology. *Curr. Opin. Neurobiol.* **30**, 92–99 (2015).
32. Neale, B.M. & Sklar, P. Genetic analysis of schizophrenia and bipolar disorder reveals polygenicity but also suggests new directions for molecular interrogation. *Curr. Opin. Neurobiol.* **30**, 131–138 (2015).
33. Del-Aguila, J.L. *et al.* Alzheimer's disease: rare variants with large effect sizes. *Curr. Opin. Genet. Dev.* **33**, 49–55 (2015).
34. Watakabe, A. *et al.* Comparative analyses of adeno-associated viral vector serotypes 1, 2, 5, 8 and 9 in marmoset, mouse and macaque cerebral cortex. *Neurosci. Res.* **93**, 144–157 (2015).
35. Deverman, B.E. *et al.* Cre-dependent selection yields AAV variants for widespread gene transfer to the adult brain. *Nat. Biotechnol.* **34**, 204–209 (2016).
36. Choudhury, S.R. *et al.* Viral vectors for therapy of neurologic diseases. *Neuropharmacology* http://dx.doi.org/10.1016/j.neuropharm.2016.02.013 (2016).
37. Han, X. *et al.* Millisecond-timescale optical control of neural dynamics in the nonhuman primate brain. *Neuron* **62**, 191–198 (2009).
38. Diester, I. *et al.* An optogenetic toolbox designed for primates. *Nat. Neurosci.* **14**, 387–397 (2011).
39. Macosko, E.Z. *et al.* Highly parallel genome-wide expression profiling of individual cells using nanoliter droplets. *Cell* **161**, 1202–1214 (2015).
40. Tasic, B. *et al.* Adult mouse cortical cell taxonomy revealed by single cell transcriptomics. *Nat. Neurosci.* **19**, 335–346 (2016).
41. Krishnaswami, S.R. *et al.* Using single nuclei for RNA-seq to capture the transcriptome of postmortem neurons. *Nat. Protoc.* **11**, 499–524 (2016).
42. Anderson, D.J. & Perona, P. Toward a science of computational ethology. *Neuron* **84**, 18–31 (2014).
43. Sadagopan, S., Temiz-Karayol, N.Z. & Voss, H.U. High-field functional magnetic resonance imaging of vocalization processing in marmosets. *Sci. Rep.* **5**, 10950 (2015).
44. Herrmann, T. *et al.* The travelling-wave primate system: a new solution for magnetic resonance imaging of macaque monkeys at 7 Tesla ultra-high field. *PLoS One* **10**, e0129371 (2015).
45. Eliades, S.J. & Wang, X. Chronic multi-electrode neural recording in free-roaming monkeys. *J. Neurosci. Methods* **172**, 201–214 (2008).
46. Roy, S. & Wang, X. Wireless multi-channel single unit recording in freely moving and vocalizing primates. *J. Neurosci. Methods* **203**, 28–40 (2012).
47. Yin, M. *et al.* Wireless neurosensor for full-spectrum electrophysiology recordings during free behavior. *Neuron* **84**, 1170–1182 (2014).
48. Fernandez-Leon, J.A. *et al.* A wireless transmission neural interface system for unconstrained non-human primates. *J. Neural Eng.* **12**, 056005 (2015).
49. Izpisua Belmonte, J.C. *et al.* Brains, genes, and primates. *Neuron* **86**, 617–631 (2015).
50. Vallender, E.J. & Miller, G.M. Nonhuman primate models in the genomic era: a paradigm shift. *ILAR J.* **54**, 154–165 (2013).
51. Kaiser, T. & Feng, G. Modeling psychiatric disorders for developing effective treatments. *Nat. Med.* **21**, 979–988 (2015).
52. Tu, Z., Yang, W., Yan, S., Guo, X. & Li, X.-J. CRISPR/Cas9:a powerful genetic engineering tool for establishing large animal models of neurodegenerative diseases. *Mol. Neurodegener.* **10**, 35 (2015).
53. Okano, H., Hikishima, K., Iriki, A. & Sasaki, E. The common marmoset as a novel animal model system for biomedical and neuroscience research applications. *Semin. Fetal Neonatal Med.* **17**, 336–340 (2012).
54. Bendor, D. & Wang, X. The neuronal representation of pitch in primate auditory cortex. *Nature* **436**, 1161–1165 (2005).
55. Song, X., Osmanski, M.S., Guo, Y. & Wang, X. Complex pitch perception mechanisms are shared by humans and a New World monkey. *Proc. Natl. Acad. Sci. USA* **113**, 781–786 (2016).
56. Mitchell, J.F., Reynolds, J.H. & Miller, C.T. Active vision in marmosets: a model system for visual neuroscience. *J. Neurosci.* **34**, 1183–1194 (2014).
57. Shiba, Y., Santangelo, A.M. & Roberts, A.C. Beyond the medial regions of prefrontal cortex in the regulation of fear and anxiety. *Front. Syst. Neurosci.* **10**, 12 (2016).
58. Miller, C.T. *et al.* Marmosets: a neuroscientific model of human social behavior. *Neuron* **90**, 219–233 (2016).
59. Kaas, J.H. The evolution of brains from early mammals to humans. *Wiley Interdiscip. Rev. Cogn. Sci.* **4**, 33–45 (2013).
60. Mashiko, H. *et al.* Comparative anatomy of marmoset and mouse cortex from genomic expression. *J. Neurosci.* **32**, 5039–5053 (2012).
61. Liu, D. *et al.* Medial prefrontal activity during delay period contributes to learning of a working memory task. *Science* **346**, 458–463 (2014).
62. Leach, P.T., Hayes, J., Pride, M., Silverman, J.L. & Crawley, J.N. Normal performance of Fmr1 mice on a touchscreen delayed nonmatching to position working memory task. *eNeuro* http://dx.doi.org/10.1523/eneuro.0143-15.2016 (2016).
63. Shultz, S., Opie, C. & Atkinson, Q.D. Stepwise evolution of stable sociality in primates. *Nature* **479**, 219–222 (2011).
64. Donaldson, Z.R. & Young, L.J. Oxytocin, vasopressin, and the neurogenetics of sociality. *Science* **322**, 900–904 (2008).
65. Garrison, J.L. *et al.* Oxytocin/vasopressin-related peptides have an ancient role in reproductive behavior. *Science* **338**, 540–543 (2012).
66. Freeman, S.M. & Young, L.J. Comparative perspectives on oxytocin and vasopressin receptor research in rodents and primates: translational implications. *J. Neuroendocrinol.* http://dx.doi.org/10.1111/jne.12382 (2016).
67. Levin, E.D. α7-Nicotinic receptors and cognition. *Curr. Drug Targets* **13**, 602–606 (2012).
68. Quik, M. *et al.* Localization of nicotinic receptor subunit mRNAs in monkey brain by *in situ* hybridization. *J. Comp. Neurol.* **425**, 58–69 (2000).
69. Fidler, B. Restructuring looms for Forum as neuro drug fails key clinical test. *Xconomy* http://www.xconomy.com/boston/2016/03/24/restructuring-looms-for-forum-as-neuro-drug-fails-key-clinical-test/ (2016).
70. Lemon, R.N. Descending pathways in motor control. *Annu. Rev. Neurosci.* **31**, 195–218 (2008).
71. Butti, C., Santos, M., Uppal, N. & Hof, P.R. Von Economo neurons: clinical and evolutionary perspectives. *Cortex* **49**, 312–326 (2013).
72. Herculano-Houzel, S., Collins, C.E., Wong, P. & Kaas, J.H. Cellular scaling rules for primate brains. *Proc. Natl. Acad. Sci. USA* **104**, 3562–3567 (2007).
73. Ventura-Antunes, L., Mota, B. & Herculano-Houzel, S. Different scaling of white matter volume, cortical connectivity, and gyrification across rodent and primate brains. *Front. Neuroanat.* **7**, 3 (2013).
74. Takeda, S. *et al.* Neuronal uptake and propagation of a rare phosphorylated high-molecular-weight tau derived from Alzheimer's disease brain. *Nat. Commun.* **6**, 8490 (2015).
75. LaFerla, F.M. & Green, K.N. Animal models of Alzheimer disease. *Cold Spring Harb. Perspect. Med.* **2**, a006320 (2012).

76. Cummings, J.L., Morstorf, T. & Zhong, K. Alzheimer's disease drug-development pipeline: few candidates, frequent failures. *Alzheimers Res. Ther.* **6**, 37 (2014).

77. Forny-Germano, L. *et al.* Alzheimer's disease-like pathology induced by amyloid-β oligomers in nonhuman primates. *J. Neurosci.* **34**, 13629–13643 (2014).

78. Johnson, M.B. *et al.* Single-cell analysis reveals transcriptional heterogeneity of neural progenitors in human cortex. *Nat. Neurosci.* **18**, 637–646 (2015).

79. Otani, T., Marchetto, M.C., Gage, F.H., Simons, B.D. & Livesey, F.J. 2D and 3D stem cell models of primate cortical development identify species-specific differences in progenitor behavior contributing to brain size. *Cell Stem Cell* **18**, 467–480 (2016).

80. Webb, S.J., Jones, E.J.H., Kelly, J. & Dawson, G. The motivation for very early intervention for infants at high risk for autism spectrum disorders. *Int. J. Speech Lang Pathol.* **16**, 36–42 (2014).

81. Qureshi, I.A. & Mehler, M.F. Emerging roles of non-coding RNAs in brain evolution, development, plasticity and disease. *Nat. Rev. Neurosci.* **13**, 528–541 (2012).

82. Walker, S. & Scherer, S.W. Identification of candidate intergenic risk loci in autism spectrum disorder. *BMC Genomics* **14**, 499 (2013).

83. Batzoglou, S., Pachter, L., Mesirov, J.P., Berger, B. & Lander, E.S. Human and mouse gene structure: comparative analysis and application to exon prediction. *Genome Res.* **10**, 950–958 (2000).

84. Bernard, A. *et al.* Transcriptional architecture of the primate neocortex. *Neuron* **73**, 1083–1099 (2012).

85. Bateson, P. *Review of Research using Non-Human Primates* http://www.bbsrc.ac.uk/documents/review-research-using-nhps-pdf/ (2011).

86. Emanuel, E.J., Wendler, D. & Grady, C. What makes clinical research ethical? *J. Am. Med. Assoc.* **283**, 2701–2711 (2000).

87. Liu, Z. *et al.* Autism-like behaviours and germline transmission in transgenic monkeys overexpressing MeCP2. *Nature* **530**, 98–102 (2016).

88. Liu, Z. *et al.* Generation of macaques with sperm derived from juvenile monkey testicular xenografts. *Cell Res.* **26**, 139–142 (2016).

89. Kanatsu-Shinohara, M. *et al.* Long-term proliferation in culture and germline transmission of mouse male germline stem cells. *Biol. Reprod.* **69**, 612–616 (2003).

90. Sato, T. *et al.* Genome editing in mouse spermatogonial stem cell lines using TALEN and double-nicking CRISPR/Cas9. *Stem Cell Reports* **5**, 75–82 (2015).

91. Zhou, Q. *et al.* Complete meiosis from embryonic stem cell-derived germ cells in vitro. *Cell Stem Cell* **18**, 330–340 (2016).

92. Campbell, K.H., McWhir, J., Ritchie, W.A. & Wilmut, I. Sheep cloned by nuclear transfer from a cultured cell line. *Nature* **380**, 64–66 (1996).

93. Hrabě de Angelis, M. *et al.* Analysis of mammalian gene function through broad-based phenotypic screens across a consortium of mouse clinics. *Nat. Genet.* **47**, 969–978 (2015).

94. Perrin, S. Preclinical research: make mouse studies work. *Nature* **507**, 423–425 (2014).

95. Berry-Kravis, E.M. *et al.* Effects of STX209 (arbaclofen) on neurobehavioral function in children and adults with fragile X syndrome: a randomized, controlled, phase 2 trial. *Sci. Transl. Med.* **4**, 152ra127 (2012).

96. Berry-Kravis, E. *et al.* Mavoglurant in Fragile X syndrome: results of two randomized, double-blind, placebo-controlled trials. *Sci. Transl. Med.* **8**, 321ra5 (2016).

97. Lombardi, L.M., Baker, S.A. & Zoghbi, H.Y. MECP2 disorders: from the clinic to mice and back. *J. Clin. Invest.* **125**, 2914–2923 (2015).

98. Bourgeron, T. Current knowledge on the genetics of autism and propositions for future research. *C. R. Biol.* **339**, 300–307 (2016).

99. Agamaite, J.A., Chang, C.-J., Osmanski, M.S. & Wang, X. A quantitative acoustic analysis of the vocal repertoire of the common marmoset (*Callithrix jacchus*). *J. Acoust. Soc. Am.* **138**, 2906–2928 (2015).

100. Clarke, H.F., Horst, N.K. & Roberts, A.C. Regional inactivations of primate ventral prefrontal cortex reveal two distinct mechanisms underlying negative bias in decision making. *Proc. Natl. Acad. Sci. USA* **112**, 4176–4181 (2015).

101. Kangas, B.D., Bergman, J. & Coyle, J.T. Touchscreen assays of learning, response inhibition, and motivation in the marmoset (*Callithrix jacchus*). *Anim. Cogn.* **19**, 673–677 (2016).

102. Ioannidis, J.P.A. Why most published research findings are false. *PLoS Med.* **2**, e124 (2005).

103. Collins, F.S. & Tabak, L.A. Policy: NIH plans to enhance reproducibility. *Nature* **505**, 612–613 (2014).

104. Button, K.S. *et al.* Power failure: why small sample size undermines the reliability of neuroscience. *Nat. Rev. Neurosci.* **14**, 365–376 (2013).

105. Open Science Collaboration. Psychology. Estimating the reproducibility of psychological science. *Science* **349**, aac4716 (2015).

106. Tsilidis, K.K. *et al.* Evaluation of excess significance bias in animal studies of neurological diseases. *PLoS Biol.* **11**, e1001609 (2013).

107. Bauer, S.A. & Baker, K.C. Persistent effects of peer rearing on abnormal and species-appropriate activities but not social behavior in group-housed rhesus macaques (*Macaca mulatta*). *Comp. Med.* **66**, 129–136 (2016).

108. Committee on Strategies for Small-Number-Participant Clinical Research Trials, Board on Health Sciences Policy & Institute of Medicine. *Small Clinical Trials: Issues and Challenges* (National Academies Press, 2001).

109. National Research Council (US) Institute for Laboratory Animal Research. *Transportation of Primates and the Animal Welfare Act* (National Academies Press, 2003).

110. Bloom, D.E. *et al. The Global Economic Burden of Noncommunicable Diseases* (World Economic Forum, Geneva, 2011).

111. Chisholm, D. *et al.* Scaling-up treatment of depression and anxiety: a global return on investment analysis. *Lancet Psychiatry* **3**, 415–424 (2016).

112. Patel, V. *et al.* Addressing the burden of mental, neurological, and substance use disorders: key messages from Disease Control Priorities, 3rd edition. *Lancet* **387**, 1672–1685 (2016).

113. Chan, A.W.S. *et al.* Progressive cognitive deficit, motor impairment and striatal pathology in a transgenic Huntington disease monkey model from infancy to adulthood. *PLoS One* **10**, e0122335 (2015).

114. Niu, Y. *et al.* Early Parkinson's disease symptoms in α-synuclein transgenic monkeys. *Hum. Mol. Genet.* **24**, 2308–2317 (2015).

115. Cox, D.B.T., Platt, R.J. & Zhang, F. Therapeutic genome editing: prospects and challenges. *Nat. Med.* **21**, 121–131 (2015).

116. Yang, L. *et al.* Genome-wide inactivation of porcine endogenous retroviruses (PERVs). *Science* **350**, 1101–1104 (2015).

117. Slaymaker, I.M. *et al.* Rationally engineered Cas9 nucleases with improved specificity. *Science* **351**, 84–88 (2016).

118. Kleinstiver, B.P. *et al.* High-fidelity CRISPR-Cas9 nucleases with no detectable genome-wide off-target effects. *Nature* **529**, 490–495 (2016).

119. Wan, H. *et al.* One-step generation of p53 gene biallelic mutant cynomolgus monkey via the CRISPR/Cas system. *Cell Res.* **25**, 258–261 (2015).

120. Chen, Y. *et al.* Generation of cynomolgus monkey chimeric fetuses using embryonic stem cells. *Cell Stem Cell* **17**, 116–124 (2015).

121. Swiech, L. *et al. In vivo* interrogation of gene function in the mammalian brain using CRISPR-Cas9. *Nat. Biotechnol.* **33**, 102–106 (2015).

122. Platt, R.J. *et al.* CRISPR-Cas9 knockin mice for genome editing and cancer modeling. *Cell* **159**, 440–455 (2014).

123. Konermann, S. *et al.* Optical control of mammalian endogenous transcription and epigenetic states. *Nature* **500**, 472–476 (2013).

124. Konermann, S. *et al.* Genome-scale transcriptional activation by an engineered CRISPR-Cas9 complex. *Nature* **517**, 583–588 (2015).

125. Dominguez, A.A., Lim, W.A. & Qi, L.S. Beyond editing: repurposing CRISPR-Cas9 for precision genome regulation and interrogation. *Nat. Rev. Mol. Cell Biol.* **17**, 5–15 (2016).

126. Komor, A.C., Kim, Y.B., Packer, M.S., Zuris, J.A. & Liu, D.R. Programmable editing of a target base in genomic DNA without double-stranded DNA cleavage. *Nature* **533**, 420–424 (2016).

127. Abudayyeh, O.O. *et al.* C2c2 is a single-component programmable RNA-guided RNA-targeting CRISPR effector. *Science* http://dx.doi.org/10.1126/science.aaf5573 (2016).

[1]McGovern Institute for Brain Research, Massachusetts Institute of Technology, Cambridge, Massachusetts, USA. [2]Department of Brain and Cognitive Sciences, Massachusetts Institute of Technology, Cambridge, Massachusetts, USA. [3]Stanley Center for Psychiatric Research, Broad Institute of MIT and Harvard, Cambridge, Massachusetts, USA. [4]Picower Institute for Learning and Memory, Massachusetts Institute of Technology, Cambridge, Massachusetts, USA. [5]Center for Neural Science, New York University, New York, New York, USA. [6]Institute of Neuroscience, CAS Key Laboratory of Primate Neurobiology, State Key Laboratory of Neuroscience, CAS Center for Excellence in Brain Science and Intelligence Technology, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai, China. [7]Department of Physiology, Development and Neuroscience, Behavioural and Clinical Neuroscience Institute, University of Cambridge, Cambridge, UK. [8]Interdisciplinary Institute of Neuroscience and Technology, Zhejiang University, Hangzhou, China. [9]Laboratory of Auditory Neurophysiology, Department of Biomedical Engineering, Johns Hopkins University School of Medicine, Baltimore, Maryland, USA. [10]The Brain Cognition and Brain Disease Institute (BCBDI) for Collaboration Research of SIAT at CAS and McGovern Institute at MIT, Shenzhen Institutes of Advanced Technology (SIAT), Chinese Academy of Science, Shenzhen, China. [11]Shenzhen Key Lab of Neuropsychiatric Modulation and Collaborative Innovation Center for Brain Science, CAS Center for Excellence in Brain Science and Intelligence Technology, The Brain Cognition and Brain Disease Institute (BCBDI) for Collaboration Research of SIAT at CAS and McGovern Institute at MIT, Shenzhen Institutes of Advanced Technology (SIAT), Chinese Academy of Science, Shenzhen, China. [12]Broad Institute of MIT and Harvard, Cambridge, Massachusetts, USA. Correspondence should be addressed to G.F. (fengg@mit.edu).