JPND Consultation on the Use of Non-Human Primates in Neurodegenerative Disease Research

Contents

1. Executive Summary

To contribute to the update of the JPND scientific strategy, a small-scale consultation of academic researchers and industry representatives from 16 countries was conducted on the use of non-human primates (NHPs) in neurodegenerative disease (ND) research. The objective of the consultation was to obtain an understanding of the global application of NHPs in ND research. Findings were as follows:

Respondents identified NHP-ND research in Europe, North America and Asia, with most studies focused on Alzheimer's disease and Parkinson's disease and include a wide range of techniques and models. From a pharmaceutical industry perspective, NHPs were used mainly in advanced preclinical testing to evaluate drug safety, however, some companies adopted strategies that were less dependent on NHPs.

Participants gave scientific justifications for the use of NHPs in ND research for a range of reasons, including improved translation to humans, their suitability for studying the complexity and higher cognitive functions affected in ND and the ability of NHPs to overcome technical limitations of smaller animal models. From an ethical perspective, respondents largely took the view that NHP-ND research could be justified where studies were methodically rigorous, focused on important medical questions and for which no alternatives were available. However, 14% of respondents held the view that there was no justification for NHP-ND research.

Limitations of using NHPs in research, were recognised by respondents to include small sample sizes, high costs, ethical concerns and the inability of NHPs to model all aspects of human disease, although in general these were not specific to ND research.

There was no consensus on whether current techniques that are complementary to the use of animals could reduce or replace the use of NHPs in ND research. Looking further ahead, 23% of respondents anticipated that advances such as the generation of complex organoids and use of sophisticated computer models of disease could potentially reduce or replace future use of NHPs.

Areas identified that would benefit from greater worldwide cooperation were the development of shared international standards for the use of NHPs and the advancement and adoption of alternatives. NHP users had several practical suggestions, which included the development of specialised research centres and sharing of tissues, models and data between researchers to optimise the use of current NHP resources.

Regulatory or legal issues were not considered to constrain necessary and important research involving NHPs, although researchers perceived some countries as having stricter regulations than others.

2. Aim of the consultation

The JPND Scientific Advisory Board (SAB) identified the use of NHPs as an important topic to examine in the context of current ND research. To gain insight into this area, a small-scale questionnaire-based consultation (Annex 1) was conducted across the ND research community from 1st December 2017 to the 29th January 2018.

The objective of the consultation was to obtain a broader understanding of the global application of NHPs used in ND research, encompassing the scientific and regulatory issues, as well as current thinking amongst the scientific community on alternative approaches. In addition, views were sought on which aspects of ND research were considered to necessitate the use of NHPs and on the reasons for NHP research to outsourced to non-EU countries.

The consultation findings will be used by the SAB to inform an updated edition of the JPND Research and Innovation Strategy (*Strategic Research and Innovation Agenda*). This strategy provides a framework for investment in ND research for the next 5-10 years and will be available in early 2019.

To be clear, this report does not represent a 'JPND view' on this topic but aims to summarise the content of the responses that were received to the questionnaire. Responses to each question were analysed irrespective of the participant's previous comments.

3. European Commission 2017 report on NHPs in research

A recent scientific opinion on 'The need for non-human primates in biomedical research, production and testing of products and devices' by the Scientific Committee on Health, Environmental and Emerging Risks (SCHEER) was updated in May 2017. This provided a more general evaluation of the use and requirement of NHPs across the broader field of biomedical research, together with recommendations for advancing the 3Rs (Replacement, Reduction and Refinement) for NHP use and overcoming existing barriers to the implementation of alternatives.

4. Results

The results of the questionnaire are broadly reported in the order of the original questions (see Annex 1).

4.1. ND research involving NHPs[1]

Thirty two percent of participants (14 responses) stated that they performed NHP-ND research (non-human primate, neurodegenerative disease research, Figure 1), while the remaining 68% (30 responses) were non-NHP researchers active in the area of ND (see Annex 2 for a breakdown of consultation responses by research type and country). In addition, nine NHP researchers and five non-NHP researchers were aware of collaborators or colleagues who conducted NHP-ND research.

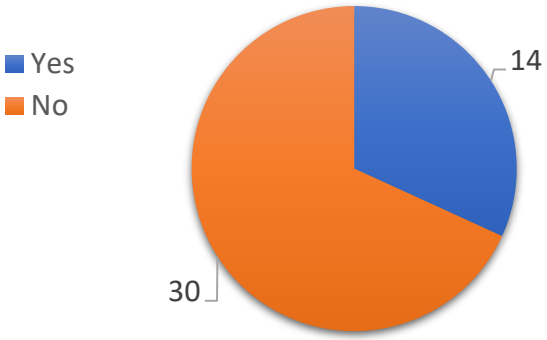

Figure 1. Respondents were asked if they conduct NHP-ND research.

Having first identified areas of NHP-ND research, respondents were asked to detail the disease relevance and type of research conducted. The majority of NHP-ND research reported was conducted in Parkinson's disease and Alzheimer's disease. Other disease areas included studies on dementia with Lewy bodies, Huntington's disease, prion disease and multiple system atrophy.

The following types of research were identified by respondents with either an academic or industry background:

Academia

- NHP model development (transgenic, neurotoxin, Aβ oligomer and tau-based models)
- Genome-editing to study gene functions related to neurodegeneration
- Electrophysiology (extracellular recordings) and electrochemistry (*in vivo* voltammetry)
- Therapeutic mechanisms of surgical therapy and pathophysiology of disease
- Transmissibility and mechanisms of prions propagation/toxicity
- Molecular studies in specific NHP brain areas
- Behavioural assessment in NHP models

Industry

- Drug development (delivery, pre-clinical testing, pharmacokinetic/pharmacodynamic modelling, efficacy and toxicology)
- Gene therapy research for rare neurodegenerative diseases
- Vaccine-based studies (immunotherapies)
- Induction of pharmacological deficits (e.g. scopolamine induced memory impairments, levodopa-induced dyskinesia) and lesioning approaches

[1] Section refers to responses received in Q2 of the questionnaire (see Annex 1)

- Studies of memory, motor function, attention systems and eye tracking
- Cognitive assessments in aged NHPs
- Structural and functional imaging (MRI, PET)
- Skull surface electroencephalogram (EEG) and deep brain electrophysiology
- Biological sample collection (e.g. blood, serum, CSF)

4.2. Location of NHP-ND research[2]

Respondents were aware of NHP-ND research activity in the countries detailed in Table 1. Note, the questionnaire was completed by respondents located in all the countries listed below, in addition to others (see Figure 5 in Annex 2 for further details). The consultation involved a relatively small sample size, so this should not be interpreted to mean that NHP-ND research is confined to these countries.

Table 1. Countries identified where NHP-ND research is conducted	
Canada	South Korea
China	Spain
France	UK
Germany	USA
Japan	

The following reasons were given where the NHP-ND studies identified took place outside the European Union (EU):

- the location of the lead investigator's institute
- the location of the necessary research expertise
- the availability of a purpose-built facility in a different region
- improved access to NHP breeding colonies in Asian countries
- the cost of NHP research
- the availability of specific NHP models and assays.

4.3. Justification for the use of NHPs in ND research[3]

Participants were asked for their views on the scientific and ethical justification for the use of NHPs in ND research. Six of the 44 respondents (14%) considered that there were no scientific or ethical reasons to support NHP-ND research and several respondents commented that there were currently insufficient data available to demonstrate the benefits of NHPs over rodent models.

Across the remaining responses that provided justification for NHP-ND research, the following scientific and ethical viewpoints were identified:

Scientific considerations

- The greater proximity of NHP models to human physiology/genetics provides improved translatability of ND mechanisms and therapeutics. e.g. The dynamics of cerebrospinal fluid flow

[2] Section refers to responses received in Q3 of the questionnaire (see Annex 1)
[3] Section refers to responses received in Q4a of the questionnaire (see Annex 1)

between human/NHP brains and those of lower species differs. In addition, neuronal populations containing neuromelanin are only found in NHPs and humans (not in lower species)

- NHP models are required to study complex human disorders such as ND (e.g. involving higher cognitive/motor/sensory function), since they have the necessary neuronal networks and brain structures. Certain brain regions (e.g. neocortex) are only present/more developed in NHPs

- Alternative models in lower species do not replicate all the pathological hallmarks (molecular, cellular and clinical) of ND, which results in poor translation. Several respondents noted that this had contributed to a lack of success with drugs developed initially in rodents. Note, poor translational validity was also viewed as a drawback of NHPs (see Table 2 below)

- Model systems in non-NHP mammals can present technical limitations for certain experiments, e.g. rodent brains are usually too small to evaluate the efficacy of drug delivery

- NHPs are important for advanced preclinical testing of novel therapeutics as they can inform more closely about probable toxicity and side effects in clinical trials[4]. These adverse effects might not be detectable in lower species. This issue was also noted from an ethical perspective, in that experiments performed directly in humans may have unforeseen toxic effects.

Ethical considerations

- the severity and impact of ND is overwhelming society and treatments are currently lacking

- NHP research is justified when there is no alternative approach available to answer a defined scientific question of biomedical importance to humans

- ND-NHP research must be performed in full compliance with relevant legislation and frameworks for more humane animal research e.g. EU council directive (2010/63/EU), 3Rs principles, National Institutes of Health (NIH) Guide for the Care and Use of Laboratory Animals

- NHP research should adopt standards more in line with human clinical trials. For example, evaluation of the research should be carried out by an ethics committee and retrospective assessments should determine research impact

- NHP research is justified if methodologically sound[5], has a clear hypothesis and answers a question of medical importance with future therapeutic potential

[4] Current regulatory guidelines state that safety and tolerability data are required from two species, a rodent (rat or mouse) and non-rodent species (dog, minipig or NHP) before the administration of new pharmaceuticals to humans. The UK NC3Rs and the Association of British Pharmaceutical Industries (ABPI) are reviewing whether in some instances data from a single species may be sufficient to enable safe progression in humans. For further information, please refer to Prior et al. International Journal of Toxicology 37 (2), 121-124

[5] The UK NC3Rs has developed tools and resources for the *in vivo* scientific community to improve the reporting and experimental design of research involving animals. These include the ARRIVE Guidelines and the Experimental Design Assistant.

4.4. Drawbacks of NHPs in ND research[6]

A range of disadvantages or limitations of using NHPs were identified (outlined in Table 2 below), however many of these considerations are not specific to ND research.

Table 2. Drawbacks identified by respondents of using NHPs in ND research	
• Long lifespan increases study length and cost • High cost research is unsustainable for academic groups • Small sample sizes and concerns over validity and significance of findings • Poor translational validity of NHP models to aspects of human disease • Technical difficulties of genetic manipulation in NHPs	• Ethics and emotional considerations* • Political and public pressures and effects on regulations and funding • A lack of trained expertise and research support • Limited availability and proximity to NHP research facilities or resources • Limited number of relevant disease models available

* An in-depth view on ethical issues relating to NHP use in ND research was beyond the scope of this consultation. The fundamental issue is whether it is morally justified to cause highly sentient animals pain, suffering, distress or lasting harm in research aimed at preventing or alleviating human suffering. For further information on the ethics of NHP use in biomedical research see Prescott, 2010 and Arnason, 2018.

4.5. Research advances using NHPs[7]

Research related to Parkinson's disease employing NHPs was identified as the primary area that had created the most advances, particularly in terms of realising approved, clinically efficacious treatments. A number of respondents commented that studies of the motor circuitry of the basal ganglia in rhesus macaques had played an important role in the development and implementation of new techniques for deep brain stimulation in Parkinson's disease patients.

The types of Parkinson's disease research currently considered to be advancing the field included neuroprotection studies, models of the early stages of disease or research where alpha-synuclein was genetically overexpressed or injected into discrete brain areas (e.g. basal ganglia nuclei).

NHP studies were also identified as having made a significant contribution to collective knowledge across ND research. This included work on protein aggregation and the use of genetically modified NHPs to model specific elements of human ND. An example of this was targeted knockdown of Parkin, BACE1 or TDP-43 in NHP brains using CRISPR/Cas9 gene-editing[8]. Other examples of NHP research thought to be progressing the ND field are provided in Table 3.

Table 3. Examples of areas of NHP research thought to be contributing to progress in the ND field	
• Behavioural assessment of motor impairments in dopaminergic depletion models • Radiotracer development for functional imaging studies (e.g. tau PET, TSPO-PET) • Lesion/pharmacological inactivation studies in brain regions involved in task performance • Research on prion disease transmission and infection	• Neural underpinnings of cognition from *in vivo* electrophysiology studies • Multiscale data collection in tasks mirrored in clinical trials to identify non-invasive biomarkers (e.g. EEG, eye-tracking) • Drug delivery studies**,** intranasal delivery of drug to NHP brains • Acute inflammation, tau or amyloid injections in brain structures alongside imaging

[6] Section refers to responses received in Q4b of the questionnaire (see Annex 1)
[7] Section refers to responses received in Q5 of the questionnaire (see Annex 1)
[8] See Yang et al., (2016), CRISPR/Cas9: Implications for Modeling and Therapy of Neurodegenerative Diseases, *Front Mol Neurosci*, 9; 30.

Sixteen percent of respondents stated that they did not have sufficient knowledge of NHP-related research to be able to comment and 11% (5 responses) said that no area of ND research has benefited from NHP use. Specific areas of ND research that were viewed not to have advanced through NHP studies included the identification of novel genes or pathological mechanisms, genetic/epigenetic disease mechanisms and longitudinal studies of molecular and cellular changes[9].

4.6. Legislation and regulatory issues[10]

Respondents commented that legislation and regulatory requirements vary considerably from country to country. In the EU, regulation is under the European Council Directive (2010/63/UE) on the protection of animals used for scientific purposes. Thirteen participants who identified as non-NHP researchers stated that they did not have sufficient knowledge to comment on legislation or regulatory issues involving NHPs.

Views from USA, China[11], Japan[12], South Korea and across the EU in general were that directives do not constrain NHPs-ND research. Furthermore, despite perceptions that the regulatory burden was high, researchers thought that it was possible to make the case to carry out those studies they believed to be necessary and important. The consultation identified France, Germany, Italy and Norway as examples of countries where regulation of NHP research was particularly strict and where there was greater potential for studies (or the use of NHPs) to be constrained.

Industry participants identified the following regulatory or legal issues in relation to the use of NHPs in ND research:

- The rules of the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) can restrict the transfer of NHP samples between countries (particularly to the USA)
- The requirement in Article 10 of Directive 2010/63/EU that NHPs used in scientific procedures should be the offspring of NHPs bred in captivity decreases the availability of animals in Europe
- Regulations may be required to ensure that preclinical studies focusing on ND are carried out in aged model systems involving both male and female animals.

Of the eight participants with an industry background, six indicated that international regulations requiring the testing of pharmaceuticals in a non-rodent species (dog, minipig or NHP) were not currently a barrier to taking ND drugs into clinical application. Two participants indicated that compliance with regulations in some instances had resulted in delays in progressing research through to clinical application.

[9] Participants also identified disease areas where NHP models had not yet been developed. These included models of fronto-temporal dementia, dementia with Lewy bodies, multiple system atrophy, amyotrophic lateral sclerosis, neuropsychiatric/cognitive deficits of ND and genetic models of human mutations.
[10] Section refers to responses received in Q6 and Q10 of the questionnaire (see Annex 1)
[11] In China, it was stated that the National Natural Science Foundation (NSFC) supports NHP-ND research
[12] In Japan, it was stated that NHP-ND research was supported by the National BioResource Project

4.7. Approaches complementary to NHP models in ND research

4.7.1. Viewpoints on current status[13]

There was no clear agreement across participants as to whether currently available complementary approaches[14] could reduce or replace the use of NHPs in ND research (Figure 2). The general opinion across those who answered 'No' was that the status of alternative approaches was not at present adequate or advanced enough to replace NHP models all together. This view was also highlighted in the context of drug development, where direct rodent-to-human translational approaches have had limited success. Participants who answered 'Yes', stated that research involving advanced iPSC models and use of cell and animal models based on human material could reduce the use of NHPs in ND research. Novel approaches to research design in humans and the use of post mortem tissue were also considered to be current, valid NHP alternatives.

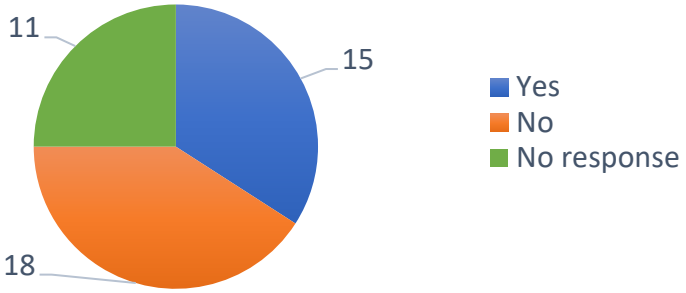

Figure 2. Respondents were asked whether currently available complementary techniques could reduce or replace the use of NHPs in ND research.

The following viewpoints were captured on currently available complementary approaches to the use of NHPs, which include associated strengths and limitations:

Cell based assays (iPSCs, neuronal and microglial cultures)

- Human iPSC-derived models were seen as an alternative approach to reduce the use of NHPs
- Cell models are valuable in drug screening, reducing the number of compounds tested *in vivo*.
- Current cell based assays are unable to replicate the complexity and circuitry of the intact organism or the human brain
- iPSCs may not consistently differentiate into specific cell types and so the generation of homogenous populations can be problematic

Post mortem tissues

- The availability of post mortem tissues from brain banks may help to reduce NHP use
- Post mortem tissues cannot reproduce the *in vivo* responses of a complex organism to a treatment
- Post mortem tissues can only provide a picture of the terminal stages of disease.

Imaging studies in humans

- Human imaging studies do not always have sufficient resolution for studies of specific molecules or molecular pathways

[13] Section refers to responses received in Q7a of the questionnaire (see Annex 1)
[14] Complementary approaches refer to experimental models, methods or tools used to address important scientific questions without the use of animals

- Imaging provides a very indirect measure of brain function in comparison to electrophysiology.

Other comments

- Researchers should adopt a '*systems neuroscience*' approach to understanding the developmental processes leading to ND at the molecular, cellular and circuit level
- A combined approach of human cell models, experimental medicine and human biomarker analysis may be a better way to model disease and evaluate drug efficacy
- Alternative approaches to consider should include sophisticated computer models and devising novel research designs for use in humans.

4.7.2. Viewpoints on future advances[15]

When asked about future directions, almost half of respondents (41%) indicated that they did not envisage any new technologies or advances on the horizon that could reduce or replace the use of NHPs in ND research (Figure 3). Among these responses, there was some anticipation that direct rodent-to-human or other model systems (e.g. iPSCs)-to-human translation might be realised in the future.

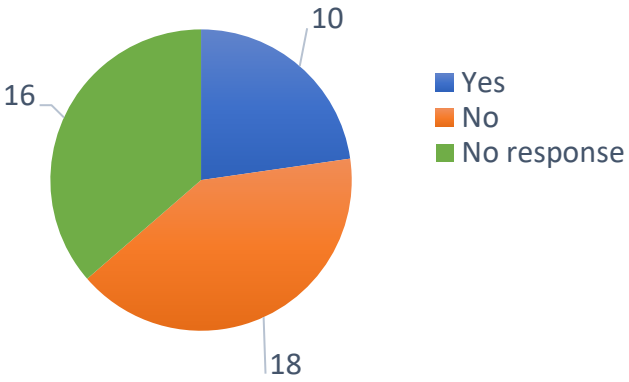

Figure 3. Respondents were asked whether there were any new technologies/advances on the horizon that could result in the reduction or replacement of NHPs in ND research

Approximately a quarter of respondents (23%, 10 responses) stated that the following potential advances could lead to a reduction or replacement of NHPs in ND research:

- Chip-based iPSC models (patient-derived and genetically engineered)
- Complex 3D cultures (e.g. organoids, organs on chips, 3D bioprinting)
- An *ex vivo* living brain with intact blood brain barrier/vessels/lymphatics
- Sophisticated computer models utilising deep learning and artificial intelligence
- Deep brain recording technology in NHPs enabling a single treatment to be profiled across numerous neural systems relevant to multiple diseases within the same subject
- Multiscale data collection and deep-phenotyping approaches e.g. linking non-invasive, clinically viable and translatable techniques with ongoing activity in the brain.

[15] Section refers to responses received in Q7b of the questionnaire (see Annex 1)

4.8. International cooperation[16]

Non-NHP users identified the development of shared international standards and regulations as the area of NHP-ND research that would benefit most from greater international cooperation. Participants also recognised the development and adoption of NHP alternatives as an important issue for international collaboration.

By contrast, those who identified as NHP users, stated that international cooperation was needed to provide additional support to increase funding in medically relevant NHP research. NHP users also had a number of practical suggestions, which included:

- the development of EU core NHP research centres
- building an international NHP ND research initiative
- Minimising barriers to NHP transport and shipping between countries
- Sharing of NHP tissues, models and data between researchers to optimise the use of current resources.

4.9. Industry strategy on NHP-ND research[17]

Most of the companies that participated in the consultation use NHPs predominantly for assessing the safety and efficacy of new disease modifying and symptomatic therapeutics prior to evaluation in humans.

The use of NHPs for ND research in industry typically varies on a case-by-case basis and depends on the disease being researched. Strategies for Parkinson's disease and Alzheimer's disease include drug development and one viewpoint was that dose escalation and pharmacokinetic studies could only be conducted in NHPs, since murine amyloid protein does not aggregate naturally with age, whereas NHP amyloid more closely resembles the protein in humans.

An industry representative stated that their current strategy only minimally relies on the use of NHPs for ND research, based on a view that novel human cell models, studies in human participants and biomarker data may be a more meaningful way to model disease and conduct drug efficacy testing. It was also acknowledged that research involving NHPs presented significant challenges for industry in terms of cost, ethics, validity and time-scales.

5. Additional comments received

At the conclusion of the questionnaire, participants were given the opportunity to provide additional comments on NHP-ND research, whether specific issues, priorities to address or more general comments for JPND to consider. These comments, (provided in full at Annex 3), covered a wide range of topics including NHP alternatives, international collaboration, data sharing, research standards, experimental design and ethical considerations.

[16] Section refers to responses received in Q8 of the questionnaire (see Annex 1)
[17] Section refers to responses received in Q9 of the questionnaire (see Annex 1)

6. Conclusion

In this small-scale consultation, 44 academic researchers and industry representatives from across 16 countries provided a range of views on the use of NHPs for ND research. This report summarises the findings of the questionnaire which details knowledge of NHP-ND research across Europe, North America and Asia, focused primarily on Parkinson's and Alzheimer's disease. Overall, Parkinson's disease research was considered to have benefited the most from NHP studies.

There were differing viewpoints on the scientific justification for the involvement of NHPs in ND research. Rationales in support of NHP use included the requirement to understand the onset of ND in the context of complex higher cognitive function and the perspective that findings from NHPs result in improved translation of ND mechanisms and therapeutics. Inconsistent with this view, several respondents considered that the body of evidence was insufficient to indicate the scientific benefits of NHPs over rodent models. Additionally, poor translational validity was identified in the consultation as a limitation of NHP-ND research. Industry respondents specifically confirmed that NHPs were important for the late, pre-clinical phases of drug discovery research. Fourteen percent of respondents held the view that there was no justification for NHP use in ND research.

Several drawbacks of using NHPs were recognised in the responses received, including ethical concerns, small sample sizes, high costs and an inability to model all aspects of human disease, although these drawbacks were not ND-specific.

There was little consensus as to whether current complementary approaches could reduce or replace the use of NHPs in ND research, although nearly a quarter of respondents anticipated that future scientific advances in alternative approaches would in due course lead to a reduction in NHP use.

Areas identified that would benefit from greater global cooperation were the development of shared international standards for the use of NHPs and the advancement and adoption of alternative approaches. Practical suggestions included the development of specialised research centres and sharing of NHP tissues, models and data between researchers.

Annex 1: Questionnaire

Consultation on the Use of Non-Human Primates in Neurodegenerative Disease Research

The Joint Programme for Neurodegenerative disease (JPND) is the largest global research initiative aimed at tackling the growing societal challenge of age-related neurodegenerative disease. It seeks to increase coordinated investment between participating countries in research directed at finding causes, developing cures, and identifying appropriate ways to care for those with neurodegenerative diseases.

Non-Human Primates in Neurodegenerative Disease Research

The JPND Scientific Advisory Board recently identified the use of non-human primates (NHPs) as an important topical issue in the neurodegenerative disease field, particularly considering the translational limitations of smaller *in vivo* models and the current trend of outsourcing research to non-EU countries.

The objective of this consultation is to obtain an understanding of the global application of NHPs in neurodegenerative disease research. This includes scientific and regulatory issues together with the current thinking and future potential of alternative approaches.

We would like the questionnaire to be primarily answered by the following expert individuals:

- *Academic or industry researchers in the neurodegenerative disease or a related field, who currently or have previously worked with NHPs*
- *Academic or industry researchers in the neurodegenerative disease or a related field, performing non-NHP studies (e.g. clinical studies, work on smaller animal models or cell based studies)*

A recent scientific opinion on '*The need for non-human primates in biomedical research, production and testing of products and devices'* by the Scientific Committee on Health, Environmental and Emerging Risks (SCHEER) was updated in May 2017. This provided a more general evaluation on the use and requirement of NHPs in research together with recommendations for advancing the 3Rs and overcoming current barriers. Link to full report

Statement on confidentiality

The questionnaire responses will be collated and viewed by the JPND Scientific Advisory Board and its Secretariat who will consider this information for a new subsection within the JPND Research Strategy. This Research Strategy is a roadmap for investment in neurodegenerative disease research over the next 5 to 10 years and the updated version is planned for release in early 2018.

Please note that under no circumstances will the individual responses be shared with third parties and all comments will be anonymised in the final summary.

Consultation on the Use of Non-Human Primates in Neurodegenerative Disease Research

1. **Please specify your background and/or experience in relation to neurodegenerative disease research (or related areas)?**

   ☐ Academic research
   ☐ Industry based research
   ☐ Welfare/animal care
   ☐ Policy/regulatory aspects
   ☐ Funding organisation/agency
   ☐ Other please specify_____

2. **a) Do you conduct studies on non-human primates (NHPs) in the context of neurodegenerative disease research?**

   ☐ Yes ☐ No *If no, please proceed to question 2b*

   If yes, what type of research/work is conducted and on what diseases?

   b) Are you aware of any colleagues at your organisation or do you have collaborators who conduct NHP studies in neurodegenerative disease research?

   ☐ Yes ☐ No

   If yes, what type of research/work is conducted and on what diseases?

   If you responded no to both questions 2a and 2b above, please proceed to question 4

3. **a) In which countries are the NHP studies outlined in question 2a or 2b performed?**

b) If these NHP studies are conducted outside the EU, please explain why this was chosen over performing studies within the EU?

4. **a) On what scientific and ethical grounds, should the use of NHPs in neurodegenerative disease research be justified?**

b) What are the drawbacks of using NHPs in neurodegenerative disease research?

5. **a) In your view, is there any current research into neurodegenerative disease involving NHPs that can be considered to be advancing understanding in the field?** Please describe any specific techniques involved.

b) Which areas of neurodegenerative disease research have not benefited from the use of NHPs? Please give details below**.**

6. **In your view, are there legislation or regulatory issues that either constrain or support the use of NHPs in neurodegenerative disease research?** Please explain below indicating the country in which this refers to.

7. **a) In your opinion, in the neurodegenerative disease field, can currently available complementary techniques e.g. iPSCs (neurons, microglia etc.), non-invasive imaging, post mortem tissue reduce or replace the use of NHPs in this area?**

☐ Yes ☐ No

Please explain your response and provide further details below.

b) Do you see any new technologies or potential advances on the horizon (but not necessarily in use) that could result in the reduction or replacement of NHPs in neurodegenerative disease research?

☐ Yes ☐ No

If yes, please explain your response and provide further details below.

8. **Which area of NHP use in neurodegenerative disease research would benefit from greater international cooperation?**

Please select up to three responses and rank your responses in number order of importance in the parentheses below.

☐ Development of shared international standards and regulations ( )
☐ Systematic reviews/meta-analyses of NHP use to minimise duplication ( )
☐ Guidance on NHP training requirements and professional development ( )
☐ Wider dissemination of information on NHP use to the public/medical profession ( )
☐ Further development and adoption of NHP alternatives ( )
☐ Additional support to increase funding in medically relevant NHP research ( )
☐ Other please specify_____ ( )

9. *If you do not come from an industry or regulatory background, please skip to question 11.*

To what extent does the strategy of industry/pharma depend on neurodegenerative disease research involving NHPs? Please provide examples where possible.

10. **International regulations require testing in a non-rodent species (e.g. NHP, dog, minipig) for the safety assessment of pharmaceuticals/vaccines prior to evaluation in humans.**

    Is this currently a barrier to taking drugs for neurodegenerative disease into clinical application? ☐ Yes ☐ No

    If yes, how do you think this could be overcome? Please explain below.

11. **To conclude, what two comments would you like to make to JPND regarding the use of NHPs in neurodegenerative disease research?**

    These could be specific issues or priorities to address or more general comments for JPND to consider in this area.

    1. _____

    2. _____

Annex 2: Numbers and categories of responses

113 academic researchers and industry professionals from the ND research community were invited to participate in the consultation, including experts conducting NHP-based research and specialists performing research in other model systems, (e.g. cell or animal based models) or in human subjects.

Experts in NHP research from the ND field were identified through relevant searches of publications and research funding using PubMed and Uber. Institutional profiles, portfolio data collected in the 2016 JPND Mapping Exercise and contacts obtained through JPND Management Board members were also used to identify participants. Experts working in other model systems were invited to participate if they had previously coordinated a JPND research project funded from 2011 to 2017.

For industry representation, we utilised the networks of the ABPI (Association of the British Pharmaceutical Industry) and EFPIA (European Federation of Pharmaceutical Industries and Associations), who invited their members to respond to the questionnaire. Responses were anonymised and collated by ABPI and EFPIA prior to submission to JPND.

44 responses (*39% response rate*) were received, composed of 38 responses from individual academic researchers and 6 responses representing industry/pharmaceutical companies (Figure 4).

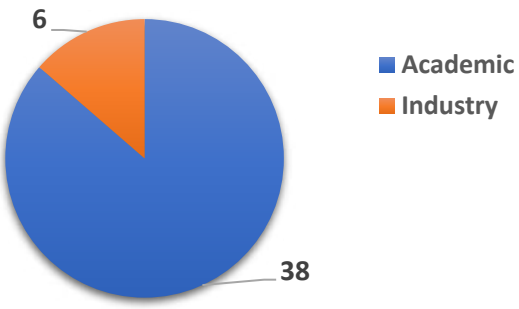

Figure 4. Background of NHP questionnaire respondents. Total number of responses received for from academia and industry are indicated on the chart[18].

[18] *Two academic researchers also had an industrial background, but were not included in the industry total.*

Responses were received from across 16 countries (Figure 5), representing Europe (28), Asia (5) and North America (5).

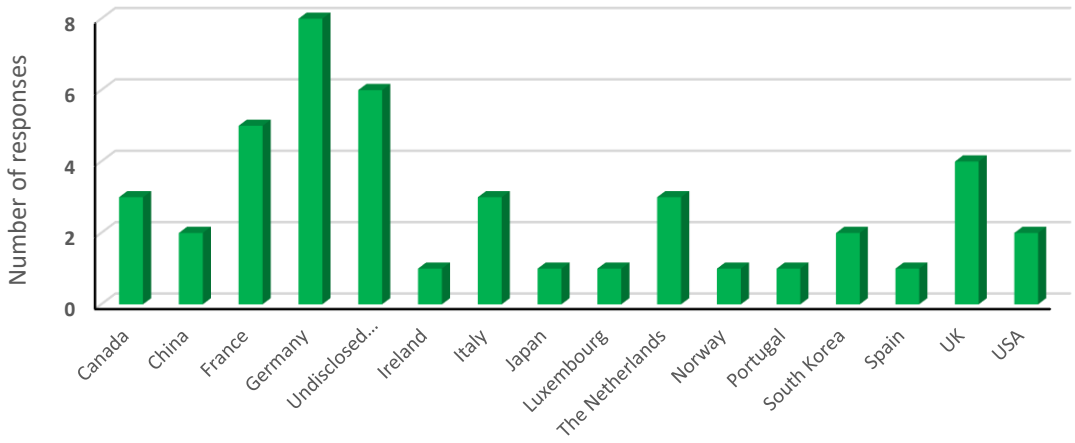

Figure 5. Number of responses shown by the country where the respondent was based. Note, for industry responses (undisclosed), it was not possible to determine the country of origin.

Annex 3: Comments provided to JPND

At the conclusion of the survey, participants were given the opportunity to provide unstructured comments on the use of NHPs in ND research. These could be specific issues or priorities to address or more general comments for JPND to consider in this area.

The table below shows the anonymised comments that JPND received from a total of 32 researchers or industry representatives, with the responses subdivided to show whether they came from NHP researchers or non-NHP researchers.

Comments provided to JPND at the conclusion of the NHP questionnaire	
NHP User/Researcher	**Non-NHP User/Researcher**
Researchers using NHPs should be encouraged, because many mouse to human approaches have failed in the neurodegenerative disease area, and many researchers have little access to NHPs.	Limit use of NHPs/develop alternative in vitro systems Share data on NHP research and increment collaborations
We will NOT cure neurodegenerative disease without NHP models There needs to be much greater international collaboration in using NHP models because it is so expensive and complicated.	The use of NHPs may be justifiable if the results obtained have significant impact on informing public health or advancing our understanding of disease processes to a level that advances treatment discovery. However, the onus must be on the researchers to explain why this research cannot be undertaken in other animal models such as humanised transgenic mice, iPSCs or cell free systems
NHP research protects humans from adverse effects of new experimental drugs NHP research is expensive and difficult to sustain for most labs given its high costs	Humans are the best model for human diseases. We should make more effort to be creative in trial design, routine deep phenotyping of patients, and drug discovery Although databases and population registers are not experiments, they produce vital and invaluable infrastructure for understanding human diseases. There should be an effort to develop and sustain such registers on a large scale
Large-scale research support for international collaborative efforts to develop genetic models Policies, funding and venues to make all models and tissues quickly available to the research community	*Major comment*: I think the discussion on the use of NHPs will sooner or later become obsolete. A more relevant discussion is whether animal research at large (irrespective of species) has any role/future in research. Political, scientific and societal discussions are increasingly voicing concerns about the use of animals in scientific research. *Minor comment*: In case NHPs still have a role to play, do we in Europe want to keep animals that are not in their natural habitat (even outside the lab)?

Humans and NHPs have close homology in both developmental and aging process, monoaminergic distribution, cognitive and behavioural aspects. Therefore, NHP studies will continuously contribute to the understanding of neurodegenerative mechanisms of human brain disease.	The generation of genetically modified NHP disease models would be very valuable for translational research, as an intermediate step between therapeutic effects in mouse models of disease and human clinical trials.
Neurotoxin-based NHP models should be replaced by NHP models showing alpha-synuclein aggregation. Although this was a long, unmet desire, at present the use of viral vectors encoding for mutated human alpha-synuclein is the best available alternative to fulfil such a gap. These strategies are well established in rodents, and need to be upgraded -validated and standardized- to NHPs. JPND should be aligned with this global tendency to maintain competitiveness of EU-based scientific programs. Given the proven success of the US National Primate Research Centres, I guess that JPND should also move into the same direction. A number of EU-based laboratories have already outsourced NHP facilities to China by signing agreements between European institutions and the Chinese Academy of Sciences. In my view, this is not the way we should go, bearing in mind that JPND should make sure that EU-based legislation is strictly maintained at all times. Furthermore and bearing in mind the enormous efforts in terms of budget and personnel that the Chinese government is allocating into this direction, I guess that training Chinese scientists on the use of NHPs sounds like a risky alternative, minimizing further EU competitiveness in what it is already a very competitive scenario.	NHPs are valuable as experimental models. They could be used in preclinical studies with well-defined endpoints or in long-term follow up studies Experiments using NHPs have high costs and large space requirements. The ethical justification of these experiments is extremely difficult
The societal risk of neurodegenerative disorders is potentially paralyzing. We must give research to prevent and treat such disorders the highest priority. The rodent models of AD have been quite disappointing with respect to translational power. The systems affected in AD (e.g., prefrontal cortex) are uniquely well-developed in primates, and thus require primate models	ND research in general requires more funding of basic research in animal models to understand the mechanisms of the diseases. Funding ND research projects for a maximum of 3 years only without the opportunity to get an extension or prolongation (including additional funding) in case of good success within the project is ineffective.

NHPs must be integrated into ALL research programs aiming at (i) understanding the pathophysiology of degenerative conditions and (ii) testing therapeutic strategies. We must ask investigators to use statistically relevant numbers of NHPs. The ethical responsibility stipulates that an experiment must provide a meaningful answer. If a study is underpowered, it is then unethical in my opinion to use NHP as the results would be dubious. Proper power must thus applied to NHP research to enable increased confidence into the results (probably counter-intuitive but true).	Continue to support it when relevant and required Continue to fund reduction and replacement methods
More sharing of data, including negative results, at earlier stages in a project, to avoid duplication of data by other teams. Better describe methods used in publications and favour open/honest discussions between scientists involved in NHP research NHPs remain so far the best translatable model for neurodegenerative diseases.	Collaboration with non-EU consortia already involved in NHP brain projects such as MINDS/ Brain Mapping by Innovative Neurotechnologies for Disease Studies
Ensure activities in frame where models can be used across the scientific field. i.e. detach from university tech transfer mechanisms which delay progress and limit sharing Customer focus, universities and industry, and balance when relevant relative to exploratory clinical studies	Due to the long life span and limited genetic manipulation/cloning technology in NHPs, so far it is still a challenging to generate NHPs animal models to mimic neurodegenerative diseases in clinic. So, to develop the novel strategy to overcome the technological limitation should be the key in future coming years. Ethical issue and international standards/regulations should be another key issue, which should be considered very carefully at the very beginning to develop JPND.
We need to continue to find alternative methods to the use of NHPs, but for the moment we need to continue the use of NHPs, in the best conditions. We need a widespread use and implementation of refinement methods.	JPND should not be involved in this kind of research
NHP research is essential to understanding and treating neurodegenerative disease. Only by investing as fully in NHP research as possible now will we get to a point where we can actively minimize and possibly eliminate the use of NHPs in the future as more predictive cross-species signals are identified. In this regard, NHP research needs to be as supported financially and vocally as possible at all levels of government, academic, industrial, and public discourse.	NHPs use: only when necessary Development of shared international standards and regulations

Significant technological advances have been made that permit NHPs to be utilized in a more ethical and responsible way than ever. Such advances promise to reduce the number of animals necessary for studies and maximize the amount of data gained from each animal during a study. As long as studies are correctly designed they can yield data that can be used to inform multiple diseases or symptoms simultaneously. Now is the time to invest more in cutting edge NHP research, not less.	
	The use of NHPs in neurodegenerative should focus more on drug development International or inter-group cooperation are needed for better NHP models and utilize the benefits of NHP models.
	Consider funding of research on the establishment of alternative large animal models, like in dogs and cats
	Consider intense social criticism It seems a back step regarding clinical investigation, why not develop sophisticated computational models?
	With a dearth of treatments to prevent, slow or cure dementias and with pharmaceutical trials being stopped and research programmes being ended it is key to keep all avenues of research open, including NHP work. I don't think it is currently possible to predict which lines of research will be the most important in treatment discovery. Clinical translational work is vital – cross fertilizing ideas, methods and theories between disciplines with a view to human application could perhaps lead to more therapeutic treatments at disease or symptomatic level.
	Use of NHPs in neurodegenerative disease research should be discouraged

	DO NOT USE NHP! Its not helpful to promote ND research Development of humanised rodent models and real AD models should be proposed. Support alternative ideas that aim on ND but not on Abeta overproduction.
	Carefully evaluate scientific benefits underpinning their use in terms of will they make a paradigm shift in our understanding of disease mechanisms or advance drug efficacy testing or not. Consider alternative options - specifically use of human cellular and experimental models for better understanding of disease mechanisms and testing new drug therapies.
	The use of these models should be restricted to well-developed studies, nevertheless, they are very necessary.