Understanding and Treating Niemann–Pick Type C Disease: Models Matter

Valentina Pallottini 1,* and Frank W. Pfrieger 2,*

1 Biomedical and Technology Science Section, Department of Science, Roma Tre University, Viale Marconi 446, 00146 Rome, Italy
2 Centre National de la Recherche Scientifique, Institut des Neurosciences Cellulaires et Intégratives, Université de Strasbourg, 8 Allée General Rouvillois, 67000 Strasbourg, France
* Correspondence: valentina.pallottini@uniroma3.it (V.P.); fw-pfrieger@gmx.de (F.W.P.)

Received: 6 October 2020; Accepted: 23 November 2020; Published: 26 November 2020

Abstract: Biomedical research aims to understand the molecular mechanisms causing human diseases and to develop curative therapies. So far, these goals have been achieved for a small fraction of diseases, limiting factors being the availability, validity, and use of experimental models. Niemann–Pick type C (NPC) is a prime example for a disease that lacks a curative therapy despite substantial breakthroughs. This rare, fatal, and autosomal-recessive disorder is caused by defects in NPC1 or NPC2. These ubiquitously expressed proteins help cholesterol exit from the endosomal–lysosomal system. The dysfunction of either causes an aberrant accumulation of lipids with patients presenting a large range of disease onset, neurovisceral symptoms, and life span. Here, we note general aspects of experimental models, we describe the line-up used for NPC-related research and therapy development, and we provide an outlook on future topics.

Keywords: lysosomal disorder; cholesterol; transgenic; cell culture; induced pluripotent stem cells; neurodegeneration; Drosophila; zebrafish; C. elegans; feline

1. Niemann–Pick Type C Disease

The prime purpose of biomedical research is to understand the molecular underpinnings of human diseases enabling the development of curative therapies. Unfortunately, these goals have been reached merely for a minuscule fraction of diseases. The large majority of ailments—afflicting from just a handful of patients to millions worldwide—awaits a treatment [1–3]. There are numerous reasons for the slow progress such as rare occurrence, molecular complexity, and variability of symptoms. However, a decisive factor is the availability, quality, and use of experimental models [4–13].

NPC is a prime example for a disease that lacks a curative therapy despite impressive breakthroughs within the last decades [14–17] and rapidly growing publication counts (Appendix A, Figure 1). At first sight, the disease seems relatively easy to study and to understand: previous research showed that it is monogenic with autosomal-recessive inheritance and caused by mutations in either of two genes, NPC1 (OMIM #257220) [18] or NPC2 (OMIM #607625) [19,20]. The structures of the corresponding proteins [21–25] together with a wealth of cell-based data indicate that this duo collaborates to pilot unesterified cholesterol out of the endosomal–lysosomal system [26–28]. If the activity of the membrane-resident NPC1 or its intraluminal partner NPC2 is diminished or absent, unesterified cholesterol accumulates in compartments of the endosome-lysosome [29,30] together with other molecules [31,32]. How can this—at first sight well-defined—cellular problem cause havoc in humans presenting an enormous variability in disease onset, symptoms, and life span? In fact, NPC disease comprises several forms based on the age at which patients present neurological symptoms [14,20,33–38]. Rare peri- and neonatal cases present hepatosplenomegaly, jaundice, and fetal hydrops with rapid death.
often due to hepatic and respiratory failure [39–44]. Most patients show infantile forms presenting hypotonia and delayed motor development (early: < 2 years) as well as clumsiness, speech delay, and cataplexy (late: 2–6 years) reaching life spans of several years [20,44,45]. The second largest group of patients shows the juvenile form (6–15 years) presenting cognitive impairment, ataxia, and dystonia [20,38,40,41,44]. The adolescent/adult form (>15 years) is characterized by cognitive impairment and psychiatric symptoms such as hallucinations and schizophrenia; the number of these patients is probably underestimated [14,20,46–50]. Notably, there is considerable overlap between the groups with respect to symptoms; many patients present common signs such as ataxia, dysphagia, and vertical supranuclear gaze palsy [20,38,41,44]. However, siblings bearing the same mutations can show distinct forms of the disease [20,33].

Figure 1. Growth of the NPC research field. Cumulative counts of publications obtained by Boolean queries in PubMed using the keywords “Niemann–Pick type c OR Niemann–Pick type c1 OR Niemann–Pick type c2 OR npc1 OR npc2” (Appendix A). Black and orange lines indicate original articles “(... NOT review [pt]” and reviews [pt] “(...) AND review [pt]”, respectively. To retrieve publications more specifically related to NPC, we restricted the query to titles [ti] or abstracts [ab] by adding the corresponding field tags to each keyword “Niemann–Pick type c [tiab] OR Niemann–Pick type c1 [tiab] OR Niemann–Pick type c2 [tiab] OR npc1 [tiab] OR npc2 [tiab]”. Sky blue and green lines indicate original articles and reviews of this subset, respectively.

The diagnosis of NPC disease is complicated by the heterogeneous clinical presentation and therefore depends on laboratory tests. This includes the so-called filipin test, the detection of plasma biomarkers [51–55], and genetic analyses [56–58]. For decades, the filipin test represented the sine qua non to diagnose NPC. It requires primary cultures of fibroblasts from patient-derived skin biopsies followed by the staining of chemically fixed cells with filipin. This bacteria-derived, fluorescent complex of molecules binds unesterified cholesterol, thus allowing the visualization of its intracellular distribution [59]. Therapeutic options are limited to symptomatic treatment [14]. The only disease-modifying drug approved for NPC in many countries, except for the USA, is Miglustat/Zavesca (N-butyldeoxynojirimycin), which decelerates disease progression in some patients [38,60,61]. The drug also serves as FDA-approved substrate reduction therapy for Gaucher disease [62,63].

Understanding the somewhat mysterious links between cellular damage and the unpredictable outcome in patients, and the development of diagnostic tests and of efficient therapies require appropriate experimental approaches and models. In the following, we will make some general remarks, we will present currently available models for NPC research, and we will highlight crucial points.

2. The Use of Experimental Models in Biomedicine

The use of experimental models in research has a long history. The first “publication” dates to the 17th century, when William Harvey described physiologic experiments with animals such as shrimp, eel, chick, and pigeon to understand blood circulation [64]. For centuries, it was believed that animals
are unable to feel pain and that they resemble machines [65]. These views changed during the age of enlightenment: In 1789, the philosopher and jurist Jeremy Bentham was one of the first to raise the issue of animal protection by stating: “The question is not, Can they reason? or Can they talk? but, Can they suffer?” [66]. In 1876, the parliament of the United Kingdom passed the “Cruelty to Animals/Anti-Vivisection Act” that updated previous legislation and imposed rules on experiments with animals. The 20th century saw the establishment of three rules, named replacement, reduction, and refinement (the 3Rs) to match “the intimate relationship between humanity and efficiency in experimentation” [67]. These rules have become a key element of laws regulating the scientific use of animals worldwide [68].

Today, biomedical research on human diseases depends entirely on experimental models that range from single cells to non-human primates. Disease models may emerge from spontaneous changes. A famous example is the nude mouse (Mus musculus) introduced by Flanagan [69] and used extensively to create models requiring immunodeficiency, for example for patient-derived xenografts [70]. Models can also be based on healthy animals, in which a disease-like state is induced experimentally. Examples include pharmacologically-induced diabetes in rodents and rabbits [71,72], Parkinson-like symptoms in non-human primates [73] and autism-like behavior in rats (Rattus norvegicus) [74]. Other pathologic conditions such as stroke and retinal ischemia can be provoked by an artificial interruption of blood supply [75] and increase of intraocular pressure [76,77], respectively. Loss of bone mass mimicking osteoporosis occurs after tail immobilization in rats [78]. Meanwhile, most experimental disease models are generated by powerful genetic tools. Not surprisingly, oncology was the first area profiting from genetically modified mice with transgenic expression of oncogenes [79]. Mice are not the only species used to mimic human pathologies. The nematode Caenorhabditis elegans has been genetically modified to generate models of Parkinson’s [80], Alzheimer’s disease [81], PolyQ disease [82], and lysosomal disorders [83]. The fruit fly Drosophila melanogaster serves as disease model for different organs including the brain [84–86], kidney [87], and pancreas [88].

The usefulness of a model depends on the specific question. Ideally, the model accurately recapitulates key aspects of the disease of interest, for example pathologic changes in cells or symptoms of patients. Furthermore, it should allow extrapolating results to the target organism. Interestingly, history teaches that extrapolability does not necessarily scale with evolutionary kinship: closer may not be better. For example, thalidomide and aspirin are well tolerated by mammalian species but not by pregnant women [89], and chimpanzee have proven inappropriate for studies on AIDS [90]. Similarly, body size and metabolic rates do not always scale with disease processes. For example, some drugs can be effective at different dosages in different animal models [91,92] and humans [93]. In addition, some animals simply do not show specific symptoms: rats cannot really cough [94], rabbits (Oryctolagus cuniculus) and rats do not show some symptoms of cystic fibrosis [95], and no animal except for non-human primates displays endometriosis symptoms [96]. On the other hand, exotic species can serve as important models for human diseases. Examples are the armadillo Dasypus novemcinctus for research on leprosy [97], the turtle Trachemys scripta to study brain hypoxia and anoxia [98], and the pet Chinchilla lanigera to investigate hearing loss [99]. Diurnal rodents represent unique models of cone-related retinal diseases [100].

Disease models based on cultured cells have seen a remarkable renaissance due to the possibility of generating specific human cells from patient-derived induced pluripotent stem cells [101]. A recent article exemplifies this new approach going from in vitro data to retrospective analysis of clinical data exposing a possible treatment [102].

3. Experimental Models for Niemann–Pick Type C1 and C2 Disease

Numerous experimental models are available to study NPC disease [103], probably because the disease is monogenic, the transmission is recessive, and orthologues of the causative genes are present in many phyla ranging from plants to mammals [104] (Table 1). The models have driven the enormous progress in the field during the last decades. Most of them concern NPC1, which is mutated in 95%
of patients. Only a few experimental models are available to study mutant NPC2. The presence of multiple isoforms in specific phyla suggests important and so far undiscovered functions of these proteins. Figure 2 indicates the use of the different models based on the number of publications (Appendix A). Clearly, the mouse has become the preferred workhorse in the NPC disease field.

![Figure 2](image_url)

**Figure 2.** Use of experimental models in NPC research. Cumulative counts (log10 values) of publications obtained by respective Boolean queries in PubMed [e.g., for mouse: (Niemann–Pick type c [tiab] OR Niemann–Pick type c1 [tiab] OR Niemann–Pick type c2 [tiab] OR npc1 [tiab] OR npc2 [tiab]) AND (mouse [tiab] OR mouse [tiab] OR mus musculus [tiab]) NOT review]. Inset, the histogram shows that most publications relate to one animal model and that only a small fraction of articles contributes to multiple cumulative counts.

### 3.1. Non-Mammalian Models

The knock-out of NPC1 orthologues in plants (*Arabidopsis thaliana*) [105] and yeast (*Saccharomyces cerevisiae*) [106] have been generated. Because both showed changes in sphingolipid, but not sterol metabolism, and because NPC2 orthologues were not known [107], it was assumed that the proteins have distinct roles across phyla. However, yeast cells bear a homologue of NPC2, which can replace the human version [108]. Moreover, yeast cells have been used to screen for pathways influencing the outcome of NPC1 deficiency [109–111] and to explore the molecular mechanism of sterol transfer based on structural data [25]. The knock-down of NPC1 and NPC2 homologues in the sterol auxotroph pathogen *Entamoeba histolytica* revealed their contribution to cholesterol uptake (*Ehnpc1*, *Ehnpc2*) [112]. The genome of *Caenorhabditis elegans* contains two homologues of mammalian NPC1 (ncr-1, ncr-2). Elimination of both forms stalls a specific phase of larval development, which is probably due to defects in the intracellular transport of cholesterol and the production of essential steroid hormones [113–115] (Table 1). The defects can be rescued by human NPC1L1 and NPC1 proteins [116] and by specific glycosphingolipids and endocannabinoids [117,118].

The elimination of *Npc1a*, one of two NPC1 homologues from *Drosophila melanogaster*, causes larval lethality (Table 1), which can be rescued by dietary supply of the steroid hormone ecdysone or by local expression of *Npc1a* in the ring gland [119,120]. The elimination of *Npc1b* also causes larval lethality due to defects in sterol absorption in the midgut (Table 1), which cannot be rescued by ecdysone [121]. The fruit fly bears a family of eight genes resembling NPC2. The simultaneous elimination of two of these genes, *Npc2a* and *Npc2b*, causes larval lethality and neurodegeneration, which again can be rescued by dietary cholesterol or ecdysone [122,123]. A genetic screen for pathways mediating cholesterol trafficking and steroidogenesis in *Drosophila* revealed that the activation of autophagy can overcome cholesterol accumulation due to NPC1 deficiency [124].
Table 1. Animal models available to study NPC disease.

| Species, Gene, Animal model | Symptom Onset | Life Span | Visceral Symptoms | Neurologic Symptoms | Lipid Accumulation in Tissues | References |
|-----------------------------|---------------|-----------|-------------------|---------------------|-----------------------------|------------|
| *Caenorhabditis elegans*    |               |           |                   |                     |                             | [113-115] |
| ncr-1                       | ND            | Dauer formation | ND               | Modified trafficking or release of synaptic vesicles | Nerve ring, spermatheca and oocytes: DHE accumulation |           |
| ncr-1(nr2022)               |               |           |                   |                     |                             |            |
| ncr-2                       |               |           |                   |                     |                             |            |
| ncr-2(nr2023)               |               |           |                   |                     |                             |            |
| *Drosophila melanogaster*   |               |           |                   |                     |                             | [119,120] |
| *Npc1a*                     | ND            | Larval lethality |                   |                     | Malpighian tubules and midgut: Sterol accumulation. Brain and retina: Chol aggregates |            |
| *Npc1b*                     | ND            | Larval lethality |                   |                     |                             | [121]      |
| *Npc2a*                     | ND            | Larval lethality |                   |                     |                             | [122,123] |
| *Npc2b*                     | ND            | Larval lethality |                   |                     | Apoptotic cell death in the nervous system | No sterol distribution abnormality |            |
| *Danio rerio*               | 99% animals die within the first MFP; 1% die before 8 months of age | Larval stage | Hepatomegaly, splenomegaly | Disturbed balance and motor control, loss of Purkinje cells | Liver: accumulation of Chol, CER, DG, LPA, PA, PC, PE, PS, TG, SL | [125-128] |
| *Mus musculus*              | 6 wks (N)     | 9–11 wks  | Hepatomegaly, splenomegaly | Disturbed motor coordination, tremor, ataxia, loss of Purkinje cells | Spleen, liver, lung, lymph nodes, thymus, bone marrow, brain: accumulation of FA, CER, Chol, SL | [18,129]  |
| *BALB/cNctr-Npc1m1Mbjg*     | 4 wks (V)     | 11–15 wks | Hepatomegaly, splenomegaly, decreased weight gain | Disturbed motor coordination, tremor, ataxia, loss of Purkinje cells | Liver: accumulation of FA, CER, Chol, SL. Brain: Chol accumulation | [130]     |
| *Mus musculus*              | 7 wks (N)     | 12 wks    | Hepatomegaly, splenomegaly, decreased weight gain | Disturbed motor coordination, tremor, ataxia, loss of Purkinje cells | Spleen, liver, lung, lymph nodes, thymus, bone marrow, brain: accumulation of FA, CER, Chol, SL | [131]     |
| *Mus musculus*              | 4 wks (N)     | 16 wks    | Hepatomegaly, splenomegaly, decreased weight gain | Disturbed motor coordination, tremor, ataxia, loss of Purkinje cells | Brain, kidney, liver, lung, and spleen: Chol accumulation. Brain and liver: GM accumulation. | [132]     |
| Species, Gene, Animal model | Symptom Onset | Life Span | Visceral Symptoms | Neurologic Symptoms | Lipid Accumulation in Tissues | References |
|----------------------------|--------------|-----------|-------------------|---------------------|-----------------------------|------------|
| *Mus musculus* Npc1<sup>tm1/I1061T</sup> Dso | 8 wks (N) | 17–18 wks | ND | Decreased motor coordination, tremor, loss of Purkinje cells | Liver and brain: Chol accumulation | [133] |
| *Mus musculus* Npc1<sup>tm1Tacf/J</sup> Npc1<sup>Imagine/J</sup> | 7 wks (N) | 9 wks | ND | Decreased motor coordination, tremor, ataxia, age-dependent hyperactivity, reduced anxiety, cortico-hippocampal defects, higher pain threshold | ND | [134] |
| *Mus musculus* Npc1<sup>tm2Tacf/J</sup> Pioneer/J | ND | Only 2% live births | ND | ND | ND | [134] |
| *Mus musculus* Npc1<sup>Imagine/Pioneer</sup> | 7 wks (N) | 9 wks | ND | Decreased motor coordination, tremor, ataxia, age-dependent hyperactivity, reduced anxiety, higher pain threshold | Liver: Chol and CER accumulation Brain: Chol, CER, GM accumulation | [134] |
| *Mus musculus* Npc1<sup>em1Pav</sup> | 4 wks (VN) | 10–12 wks, strain-dependent | ND | Loss of Purkinje cells, decreased motor coordination | Liver, brain, spleen: GM accumulation | [135] |
| *Mus musculus* Npc1<sup>1.1P</sup> Cell-specific knock-out based on Cre/loxP | | | | Depends on target cells/tissues | | [136] |
| *Mus musculus* Npc1(ASO) Knock-down of NPC1 in liver and lung by antisense oligonucleotides (ASOs) | | | Hepatomegaly; foamy, vacuolated macrophages and increased apoptosis/proliferation in liver | No neurologic symptoms | Liver: Chol accumulation | [137] |
| *Mus musculus* Tg(Gfap-Npc1) Rescue of Npc1<sup>+</sup> expression in Gfap-expressing cells | Delayed onset with respect to NPC1<sup>+</sup> (N) | 24 wks | Weight gain with respect to NPC1<sup>+</sup> | Reduced numbers of axonal spheroids and reactive astrocytes, restoration of myelin, loss of Purkinje cells, decreased neurodegeneration with respect to NPC1<sup>+</sup> | Reduced Chol accumulation in some brain areas with respect to NPC1<sup>+</sup> | [138] |
| *Mus musculus* Tg(tetO-Npc1/YFP)1Mps Cell-specific over-expression based on Tet-On/Off | | | | Depends on target cells/tissues | | [139] |
| *Mus musculus* Npc1<sup>fl/fl</sup> Cell-specific reversal of Npc1 knock-out based on Cre/loxP | | | | Depends on target cells/tissue | | [140] |
| Species, Gene, Animal model | Symptom Onset | Life Span | Visceral Symptoms | Neurologic Symptoms | Lipid Accumulation in Tissues | References |
|-----------------------------|---------------|-----------|-------------------|---------------------|-----------------------------|------------|
| *Felis catus* *NPC1*        | 6 wks (N)     | 20 wks    | Hepatomegaly, spleen and lung with multifocal histiocytosis | Tremor, ataxia, loss of Purkinje cells, astrogliosis, myelin abnormalities in peripheral nervous system | Pyramidal neurons: GM2 accumulation | [141-143] |
| *Bos taurus* *NPC1*         | 3 months (N)  | before 8 months (N = 1) | Marked hypertrophy of Purkinje cells in heart, foamy macrophages in lymph nodes | Limb weakness, dysmetria, incoordination, a wide based stance, walking sideways or falling over and recumbency, vacuolation of Purkinje cells, astrocytosis, microgliosis | Fibroblasts: Chol, GM, SL accumulation | [144] |
| *Mus musculus* *Npc2*tm1Plob | 4 wks (V)     | 18 wks    | Decreased weight gain | Tremor, motor defects, ataxia, loss of Purkinje cells | Liver: Chol accumulation, neocortex, dentate gyrus, hippocampus, and cerebellum: Chol accumulation | [145] |
| *Mus musculus* *Npc2*tm1Lst1601Beg | 8 wks (N) | ND | Decreased weight gain | Tremor, ataxia, loss of Purkinje cell, astrocytosis | Liver, spleen, kidney, lung: Chol accumulation. | [146,147] |
| *Mus musculus* Tg(ApoE-Npc2) Overexpression of NPC2 in liver | ND | ND | | | | [148] |

Abbreviations: not determined (ND); weeks (wks); neurological symptoms (N); visceral symptoms (V); cholesterol (Chol); ceramide (CER); dehydroergosterol (DHE); diacylglycerol (DG); gangliosides (GM); lysophosphatidic acid (LPA); months post fertilization (MPF); phosphatidic acid (PA); phosphatidyl-choline (PC); phosphatidyl-ethanolamine (PE); phosphatidyl-serine (PS); sphingolipids (SL); triglycerides (TG)
Induced models of NPC were created in the zebrafish Danio rerio (Table 1) using anti-oligonucleotide-based knock-down of npc1 [125,126]. These manipulations interfered with gastrulation and led to the premature death of embryos, which could be rescued by mouse Npc1 and in part by steroids [125]. Moreover, morpholino-based knock-down mimicked thrombopenia observed in human patients possibly due to defects in myeloid development [126]. CRISPR-CAS-induced null alleles of npc1 caused premature death with only a few animals surviving into adulthood. Mutant animals showed massive cholesterol accumulation and defects in the liver, cerebellum, and lateral line organ causing disturbed balance and motor control [127,128].

The non-mammalian models clearly matter, as they reveal how functions of NPC-related proteins evolved, they enable screens to identify NPC1- or NPC2-related pathways and processes, and they help to explore new therapeutic approaches. Up to now, their publication counts are lower than those of mammalian models (Figure 2).

3.2. Mammalian Models

Of all mammalian species serving biomedical research, only mice and cats (Felis catus) are currently used to study NPC (Table 1). No rat or large animal model has been established for this disease. A single case report described NPC-like symptoms in a Boxer dog [149], and a recent study described calves (Bos taurus) with progressive neurologic symptoms due to mutant NPC1, suggesting the possibility of a bovine model [144] (Table 1).

The first mouse strains to study NPC carried spontaneous mutations in Npc1, namely the insertion of a retroposon (Nih allele, further referred to as Npc1Nih) [18,129] and of a 43 base-pair (spm allele, Npc1Spm) [130,132] in BALB/c and C57BLKS/J colonies, respectively each causing a de facto Npc1 knock-out. These mice present a relatively early onset of the disease, which is characterized by hepatomegaly, weight loss, disturbed motor coordination, tremor, and ataxia. The mice die prematurely between 11 and 13 weeks of age (Table 1). Cells show an accumulation of unesterified cholesterol, gangliosides, and other lipids in different organs and tissues [132,150–154]. A similar phenotype was observed in a genetically modified mouse from the Goldstein/Brown lab. In this line (Npc1pf), a double mutation (P202A/F203A) abolishes cholesterol binding by NPC1 and invalidates its function, but it leaves its level and localization unaffected [131] (Table 1). The mice discussed so far represent one end of the model spectrum as they lack the NPC1 function completely and irreversibly. The complete absence of NPC1 occurs only in a small fraction of patients [45]. Nevertheless, these models mattered, as they enabled important discoveries including the gene responsible for the disease [18], the progressive neurodegeneration in the cerebellum [155–157], and links to autophagy [158–160] and Alzheimer’s disease [161]. Moreover, they were used extensively to explore new therapies (Table 2).

| Table 2. Summary of therapeutic approaches for NPC explored with animal models. |
|---------------------------------|-----------------|------|------|
| Treatment                        | Model           | Effect | Reference |
| Cholesterol lowering drugs       | Npc1<sup>nih</sup> | No    | [162]  |
| Apoptosis, inhibition            | Npc1<sup>nih</sup> | No    | [163]  |
| Mitogen-activated protein kinase, inhibition | Npc1<sup>nih</sup> | No    | [164]  |
| Dietary restriction              | Cat             | No    | [165]  |
| Implantation of neural stem cells | Npc1<sup>nih</sup> | No    | [166]  |
| Transplantation of mesenchymal stem cell | Npc1<sup>nih</sup> | Small | [167–169] |
| Vitamin C                        | Npc1<sup>nih</sup> | No    | [170]  |
| Vitamin E                        | Npc1<sup>nih</sup> | Yes   | [171,172] |
| Liver X receptor, activation     | Npc1<sup>nih</sup> | Yes   | [173]  |
| Pregnan X receptor, activation   | Npc1<sup>nih</sup> | Yes   | [174]  |
| Estradiol                        | Npc1<sup>nih</sup> | Small | [175]  |
| C-Abl inhibition (Imatinib)      | Npc1<sup>nih</sup> | Yes   | [176]  |
| 2-hydroxypropyl-beta-cyclodextrin| Npc1<sup>nih</sup>, Cat | Yes  | [177–181] |
| Cyclin-dependent kinase-5, inhibition | Npc1<sup>nih</sup> | Small | [182]  |
| Non-steroidal anti-inflammatory drugs | Npc1<sup>nih</sup> | Yes   | [170]  |
More common NPC1 mutations in humans induce errors in the structure of the protein leading to its degradation but leave its function more or less intact. Mouse models mimicking these changes have appeared on the scene within the last ten years (Table 1). Maue and colleagues described a mouse line with a D1005G variant that was generated by ethyl nitrosourea mutagenesis (Npc1D1005G) [132]. Praggastis and colleagues presented a knock-in of the human I1061T version of NPC1 (Npc1I1061T) [133]. This model matters as it represents approximately 20% of all NPC cases [211,212]. The mouse strains bear misfolded NPC1, causing a partial loss of function. The onset of the disease is delayed, its progress is less severe, and the life span is extended to 17 weeks compared to the complete loss-of-function mutants [37] (Table 1). In 2017, two mouse strains bearing specific human mutations were presented (Table 1). Imagine mice and compound heterozygous animals displayed different disease onsets, progress, and life spans [213,214]. In mice, the outcome of a given mutation varies with the genetic background of strains [37,135,215–218]. Numerous double mutant mice have been created to test whether and how specific candidate genes impact the disease [163,173,177,190,191,219–235]. Sex-dependent differences in behavior [236], life span [37,134], and responses to immune activation [237] and to potential therapies [171,238] were reported in some NPC1 mutant mice, raising the question of whether sex is a modifying factor in NPC disease [37] as in other cholesterol-related pathologies [239–242] and normal cholesterol homeostasis [243,244].

Several mouse models were established to study the relevance of NPC1 in specific cell types or tissues (Table 1). Using morula aggregation, so-called chimeric mouse lines were generated, in which distinct ratios of cells harbor the wild-type or the mutant allele [158]. Mice for the cell-specific

| Treatment | Model | Effect | Reference |
|-----------|-------|--------|-----------|
| Protein replacement, NPC2 | 129P2/OlaHsd-Npc1Glu1061StyBby/Nya | Small | [147] |
| Curcumin | Npc1I1061T | No | [183] |
| Glucosylceramide synthase, inhibition | Npc1I1061T, cat | Yes | [184,185] |
| N-acetylcyesteine | Npc1I1061T, Npc1(ASO) | Small | [186] |
| Copper chelation | Npc1I1061T | Yes, not CNS | [187] |
| Acetylcholinesterase, inhibition | Npc1I1061T | Small | [188] |
| Combination miglustat, curcumin, ibuprofen | Npc1I1061T | Yes | [189] |
| Glucocerebrosidase, inhibition | Npc1I1061T | Yes | [190] |
| Necroptosis, inhibition | Npc1I1061T | Yes | [191,192] |
| Heat shock protein, activation (Azimolomol) | Npc1I1061T, Npc1I1061T | Yes, not CNS | [194] |
| Histone-deacetylases, inhibition (Vorinostat) | Npc1I1061T, Npc1I1061T | Yes | [195–197] |
| Gene therapy, AAV9-NPC1 | Npc1I1061T | Yes | [198] |
| Gene therapy, AAV rh.10-NPC2 | Npc2I1061T | Yes | [199] |
| Glutathion | Npc1I1061T | Yes | [200] |
| Adenosine A2A receptor, activation | Npc1I1061T | Yes | [201] |
| Polymeric beta-cyclodextrin | Npc1I1061T, Small | Yes | [202] |
| Pneumococcal immunization | Npc1I1061T | Yes | [203] |
| Histamine H3 receptor, activation | Npc1I1061T | Yes | [204] |
| 6-O-alpha-maltosyl-beta-cyclodextrin | Npc1I1061T | Yes | [205] |
| Implantation of VEGF-overexpressing neural stem cells | Npc1I1061T, Npc1I1061T | Yes | [206] |
| CYP46A1, activation | Npc1I1061T, Npc1I1061T | Yes | [207] |
| High-density lipoprotein nanoparticles | Npc1I1061T | Small | [208] |
| Gene therapy, AAV-mediated base editing | Npc1I1061T, small | No | [209] |
| Iron chelation | Npc1I1061T | No | [210] |
| Gene therapy, Trojan horse liposomes | Npc1I1061T | No | [211] |

Table 2. Cont.
elimination of \( Npc1 \) were based on the Cre\( / \)loxP technique (\( Npc1^{tm1.1Apl} \)) [136,245,246] (Table 1). A first study showed that the elimination of \( Npc1 \) from Purkinje cells induces their degeneration but leaves the life span of mice unaffected [136]. A mouse model to study NPC1 deficiency in the liver forgoing neurologic complications was established by intra-peritoneal injections of antisense oligonucleotides in healthy BALB/c mice [137,186]. The over-expression of \( Npc1 \) in specific cell types has been accomplished using classic transgenic mice to target GFAP-expressing cells [138], the inducible TetOn/Off system, which was used to target neurons [139], and the Cre\( / \)loxP system allowing the cell-specific reversal of a \( Npc1 \) knock-out [140] (Table 1). These mice enable a cell- or tissue-specific rescue of NPC1 deficiency [218,247,248]. For example, the re-establishment of \( Npc1 \) expression in the liver rescued liver disease, but it did not prevent progressive neurodegeneration and premature death [140]. The use of cell-specific promoters requires a thorough validation of their expression patterns [249,250]. Moreover, the observation that NPC1 deficiency in neurons is sufficient to induce their death [158,245] does not exclude a demise-provoking contribution by non-neuronal cells such as microglia or astrocytes [251–253], serving potentially as therapeutic targets.

Compared to \( Npc1 \), the line-up of mouse models targeting \( Npc2 \) is much smaller. The first mouse line was created by gene targeting, resulting in 4% of normal protein levels. These animals showed a similar phenotype as NPC1-deficient mice and as mice lacking both proteins. The latter finding provided first evidence for the functional cooperation between NPC1 and NPC2 in vivo [145]. Additional lines targeting \( Npc2 \) have been generated using the gene trap approach [146,147] (Table 1). The over-expression of \( Npc2 \) in the liver was accomplished using transgenic mice and specific promoter elements [148]. More mutant alleles of mouse \( Npc1 \) and \( Npc2 \) are listed on the MGI website.

NPC-like symptoms in a domestic cat (\( Felis catus \)) were first reported by Lowenthal and collaborators [141] (Table 1). A colony was subsequently established, and the cats were further characterized. They develop neurologic symptoms such as ataxia and vestibular defects at juvenile age similar to humans, and they show neuroaxonal dystrophy [141,142,254–258]. In 2003, the genetic defect was uncovered: a single base substitution (2864G-C) in \( NPC1 \) causes an amino acid change (C955S) [143]. Two case reports described cats with distinct mutant alleles of \( NPC1 \) [259] and \( NPC2 \) [260], indicating that more feline NPC models could be established.

### 3.3. In Vitro Models

Cultured cells are instrumental to uncover basic protein functions and molecular disease mechanisms and to test potential therapeutic approaches at the cellular level [12]. The use of cell cultures to study NPC disease dates back to the 1960s, when the Fredrickson group prepared primary fibroblasts from skin and bone marrow of patients with different forms of Niemann–Pick disease, including type C [261]. This pioneering publication initiated a decades-long series of studies based on patient-derived fibroblasts (Figure 3), enabling ground-breaking discoveries. Examples are the defect in cholesterol esterification and the accumulation of unesterified cholesterol [262–264], the functional validation of \( NPC1 \)-encoding cDNA [265] and of secreted NPC2 [19], and the degradation of the misfolded p.I1061T NPC1 variant [266].

An alternative method to induce the cellular hallmark of NPC, an accumulation of unesterified cholesterol, relies on hydrophobic amines such as U18666A [267–270]. Originally, this molecule was developed as an inhibitor of cholesterol synthesis [271], and it was later shown to inhibit NPC1 activity directly [272].

#### 3.3.1. Cell-Lines

The first cell lines to study NPC disease were established from patient-derived blood lymphocytes, which were immortalized through transformation by the Epstein–Barr virus [273]. A similar approach was used to immortalize lymphoid cells from NPC2 patients [153]. A fibroblast cell line based on the \( Npc1^{ppm} \) mouse was generated using a spontaneous immortalization (3T3) protocol [274,275]. Immortalized mouse embryonic fibroblasts from NPC1-deficient mice were transduced with different constructs to monitor autophagy [276]. A mouse embryonic fibroblast cell line from NPC2-deficient
mice expressing a NPC2-crmCherry fusion protein was established to track the intracellular distribution of the protein [277]. A line of NPC2-deficient patient human fibroblasts showed a down-regulation of NPC1 upon infection with HIV [278]. Several models were derived from Chinese hamster ovary (CHO) cells, the workhorse of cell biology: NPC1-deficient CHO cells were generated using chemical or gene trap mutagenesis and assays to detect cholesterol transport-deficiency [279–281]. Other CHO lines stably over-express NPC1 [282,283], myc-tagged NPC2 [284], as well as NPC1-EGFP or -RFP fusion proteins [285–287], allowing for example to track the movement of NPC1-containing organelles [285]. CRISPR-Cas technology [288] or transfection with short interfering RNA constructs were used to generate NPC1- and NPC2-deficient HeLa [289–292] and Hek-293T cells [293]. The knock-down of NPC1 in a neuroblastoma cell line (SH-SY5Y) was achieved by stable transfection with short hairpin RNA [294]. Immortalized human hepatocytes and hepatic stellate cells with stable knock-down of NPC1 or NPC2 were obtained by transduction with lentivirus and short hairpin RNAs [295,296]. The artificial expression of NPC1 in Escherichia coli has been used to study its transport function [297]. In the context of Alzheimer disease research, NPC1 was stably down-regulated in a neuron-like Neuro-2a line that over-expresses a specific form of the amyloid precursor protein [298]. Schwann cell lines were derived using dorsal root ganglia and peripheral nerves of the Npc1<sup>erm</sup> mouse [299]. Knock-down in an oligodendroglial cell line was accomplished using short interfering RNA [200]. The first NPC model based on a haploid human cell line has been introduced recently [300].

**Figure 3.** NPC research on specific types of cells. Cumulative counts of publications obtained by respective Boolean queries in PubMed [e.g., for fibroblasts: (Niemann–Pick type c[tiab] OR Niemann–Pick type c1[tiab] OR Niemann–Pick type c2[tiab] OR npc1[tiab] OR npc2[tiab]) AND (fibroblast[tiab] OR fibroblasts[tiab]) NOT review]. Inset, the histogram shows that most publications relate to one cell model and that only a small fraction of articles contributes to multiple cumulative counts.

Cell line-based models matter to uncover basic molecular functions of NPC1 [21] or NPC2 [301] and NPC1-dependent signaling pathways [293], to perform comparative studies at the cellular level [302], and to identify disease-relevant genes [289]. Cell lines helped to identify NPC1 as a receptor mediating Ebola virus infection [303,304] and to investigate its involvement in hepatitis C virus replication [305]. However, they cannot inform about cell-type specific dependency on NPC1 and consequences of its dysfunction. Moreover, it is not clear whether NPC1- or NPC2-related cellular processes observed in cell lines occur also in specialized cells in vivo. Another caveat derives from the fact that cell lines are per definitionem mitotic, whereas most differentiated cells in the body are post-mitotic. Cell division may modify how NPC1- or NPC2-deficiency affects cells.

### 3.3.2. Primary Cultures of Brain Cells

An alternative to cell lines are primary cultures, where cells are isolated from the organism and used after different periods of culture without immortalization. Cultured cells retain their in vivo properties to degrees that depend on the cell type and the culture conditions, namely the artificial exposure to chemically undefined serum [306–310].
Most NPC patients suffer from debilitating neurologic symptoms and therefore, it appears imperative to study the impact of dysfunctional NPC1 or NPC2 on cells in the brain (Figure 3). The first studies using primary cultures of central nervous system (CNS) cells investigated the expression and distribution of NPC1 in cerebellar neurons and glial cells [311] and reported defects in cholesterol metabolism and neurotrophin signaling in striatal neurons [312]. Thereafter, sympathetic [313], cortical [314], hippocampal [315,316] and retinal neurons [317] (Figure 4) as well as purified cerebellar Purkinje cells [318] have been studied in vitro. These models matter, as they revealed neuron-specific defects caused by NPC1 deficiency such as impaired synaptic function [316,318,319], depletion of cholesterol from axons, and an accumulation of cholesterol independently from lipoprotein uptake [313, 317]. They also helped to identify lamellar inclusions as the site of cholesterol accumulation [317]. Cultured astrocytes [320], oligodendrocytes [321–323], and microglial cells [324,325] have rarely been studied, despite the potential glial involvement in neurodegeneration [251], evidence for myelination defects [246], and signs of neuroinflammation in NPC disease [326]. Organotypic cultures represent a more integrated preparation to study neurons, but they have been used only sporadically in this field [325,327].

3.3.3. Primary Cultures of Other Cells

Predominant in vitro models of NPC research are the above-mentioned patient-derived skin fibroblasts, which are mitotic primary cells, but not cell lines unless they have been immortalized. Only very few differentiated cell types are studied in the field (Figure 3). Liver and spleen are affected in many NPC patients, but few reports used primary hepatocytes [199,328,329] and hepatic stellate (Ito) cells [330] from NPC1-deficient mice, splenocytes from NPC2-deficient mice [146], and NPC1-deficient splenic B cells [331]. Acutely isolated Kupffer cells were examined in chimeric mice following bone marrow transplantation [332]. With respect to lung defects, one report studied primary type 2 pneumocytes treated with U18666A [333]. With respect to immune cells, studies used NPC1-deficient macrophages [328,332,334], invariant Natural Killer T cells and human B cell lines [335], lymphoblasts [275], monocyte-derived dendritic cells [336], and T cells [146,337,338]. To date, no studies on cultured leukocytes or granulocytes have been published. Among other cells, the effects of NPC2 knock-down on adipocyte differentiation and function were studied using primary cultures [339], and spermatozoa from NPC2-deficient mice were isolated and analyzed [340].

3.3.4. Stem Cell-Derived Models

The differentiation of specific cell types from embryonic or induced stem cells has become popular, because this technology allows studying cells from patients and producing them in large quantities. Consequently, the number of publications related to these models in the NPC field is increasing (Figure 3). A first report showed the impaired self-renewal and differentiation of neural stem cells from embryonic brains of NPC1-deficient mice [341]. Ordonez and colleagues created a short hairpin RNA-based knock-down of NPC1 in human embryonic stem cells and differentiated these cells to neurons [342]. These neurons recapitulated the pathologic hallmark of NPC, the accumulation of unesterified cholesterol, and showed impaired mitochondrial function and defective autophagy. Multipotent adult stem cells were isolated from skin biopsies of NPC patients and control subjects and differentiated to neurons showing an accumulation of cholesterol [343]. These cells were selected by specific culture conditions. An alternative and meanwhile standard approach is the reprogramming of cells from adult tissues to create induced pluripotent stem cells and their subsequent differentiation to specialized, often postmitotic cells. Several studies used this approach to generate neurons from NPC patients and healthy donors [344–349]. Maetzel and colleagues also generated stem cell-derived hepatic cells and isogenic control lines to avoid confounding effects by distinct genetic backgrounds of patients and donors [346]. The stem cell-derived models matter: they enable studying the impact of NPC1 or NPC2 deficiency on differentiated human cells, notably neurons, and to explore new
therapeutic strategies [347,350,351]. However, the protocols for reprogramming and differentiation need to be standardized to allow for comparison of results.

Figure 4. Models to study the impact of NPC1 deficiency on selected neurons in the retina. (a) Fluorescence micrographs of retinal neurons from one-week-old wild-type (WT) and NPC1-deficient (NPC) mice in vivo. NPC1 deficiency causes an intracellular accumulation of unesterified cholesterol in neurons of the ganglion cell layer in vivo (arrowheads). (b) Phase-contrast (left) and fluorescence micrographs (middle, right) of retinal neurons acutely isolated from one-week-old wild-type (WT: left, middle) and NPC1-deficient mice (NPC). In this ex vivo model, NPC1-deficient neurons maintain the increased levels of cholesterol as shown by filipin staining. (c) Phase-contrast (left) and fluorescence micrographs (middle, right) of neurons purified from the retina of one-week-old rats, cultured for 48 h and stained with filipin. Treatment with the NPC1-inhibiting drug U18666A induced an accumulation of unesterified cholesterol. Scale bars: 20 µm. In (a–c), the distribution of unesterified cholesterol was shown by the staining of chemically fixed material with filipin (a,b): Barthélémy, Pfrieger, unpublished; (c): modified from [317].

4. Models Mattering for Therapy Development

Experimental models are indispensable for the preclinical exploration of therapeutic approaches. In the NPC field, cell-based screens for targets and drugs used yeast [109], immortalized embryonic fibroblasts [276] or ovarian granulosa cells from mutant mice [352], human stem cell-derived neurons [347,348,350,351,353,354], mutant CHO lines [355], and patient-derived fibroblasts [356,357]. Numerous therapeutic approaches were tested in vivo using NPC mice and cats. Table 2 lists studies where the impact of treatments on disease progression was assessed with proper controls.

Few studies have delivered an approved drug or treatments reaching clinical development. The disease-modifying N-butyldeoxyojirimycin inhibits glucosylceramide synthase [358]. Curiously, a first in vitro study on CHO cells showed that the compound does not revert cholesterol accumulation in NPC1-deficient cells [359]. This was also observed in stem cell-derived neurons in vitro [347],
arguing against a therapeutic effect. However, in vivo studies showed that the drug slows down neurologic disease progression and prolongs the life span of NPC1-deficient BALB/c mice and NPC1 mutant cats [184,360], providing preclinical evidence for its therapeutic use (Table 2).

A potential treatment is based on 2-hydroxypropyl-beta-cyclodextrin (CD) that chelates cholesterol and other components [361] (Table 2). Curiously, the exploration of this compound started with in vivo experiments—again with discouraging results. A first study using intra-peritoneal or intra-thecal injection in NPC1-deficient mice failed to show a positive effect [362]. However, subsequent reports revealed that CD prolongs the life span, slows down neurologic disease progression, and halts the degeneration of Purkinje cells in the mouse and cat model [177–181,363,364]. Intra-thecal injections were required, as CD cannot pass the blood–brain barrier [365]. NPC1-deficient mice were also used to study ototoxicity of CD [366,367] and its effects on microglial cells [368] and the liver [369]. Effects of CD on NPC1-deficient cells were explored in lateral line neuromast cells in vivo [128], siRNA-treated HeLa cells [370], liver-derived cell lines [371], cultured fibroblasts [372–374], and primary [317,375,376] or stem cell-derived neurons [342,353,377]. First clinical data showed that CD decelerates disease progression in patients [378,379].

Histone deacetylases (HDACs) emerged as a possible therapeutic target for NPC from a genetic screen in yeast [109] and from in vitro studies of NPC1-deficient neuronal stem cells [380], patient- and mutant mouse-derived fibroblasts [133,357,374,381–384], cell lines [384,385], and U18666A-treated hippocampal neurons [386]. A first in vivo study using Npc1 mutant mice claimed that repeated intra-peritoneal injections of vorinostat, an HDAC inhibitor, together with polyetheylene–glycol and CD slow down neurologic disease progression, but some controls were missing [387]. A subsequent report on mice attributed the effects on neurologic symptoms to CD [363]. Repeated intra-peritoneal injections of vorinostat in NPC1 mutant mice improved liver function but did not slow down weight loss or increase life span [194] probably because the drug cannot enter the central nervous system [363]. A comparison of drug effects using different mouse models revealed that drug effects on liver function were not mediated by proteostatic effects on NPC1 [194] (Table 2).

Evidence from Npc1 mutant mice that heat-shock proteins protect Purkinje cells from degeneration suggested these components as new drug targets in NPC [157,227]. The idea was supported by in vitro studies on patient-derived fibroblasts [193,227,388] and U18666A-treated neurons [227] and in vivo studies exploring the over-expression or knock-down of heat shock protein beta-1 in NPC1-deficient mice [227]. A corresponding disease-modifying therapy may be based on arimoclomol, a small molecule enhancer of heat shock proteins, whose effects were explored in patient-derived fibroblasts and NPC1-deficient mice [193] (Table 2).

Within the last years, NPC1-deficient mice also helped to explore gene therapy for NPC (Table 2). First support for this approach came from two observations. The over-expression of NPC1 in brain cells was achieved following the intra-cerebral injection of an adenoviral construct in vivo [389]. The cell-specific over-expression of NPC1 in transgenic mice rescued pathologic changes due to NPC1 deficiency [139,390]. Within the last few years, a series of studies showed that the progress of neurologic disease in NPC1-deficient mice is slowed down by intra-cardiac [195], intra-cisternal [196], and intra-cerebroventricular [197] injection of vectors based on adeno-associated virus 9 (AAV9). Similar improvements were found in mice lacking NPC2 following intra-cisternal injections of AAVrh.10 carrying NPC2 [198].

5. Conclusions and Outlook

The diversity and validity of experimental models and their pertinence to topics of interest are key to advance biomedical research. Over the last decades, the NPC field has developed a gang of models that matter as they revealed the origin of the disease, provided important insight in disease mechanisms, and helped to explore new diagnostic and therapeutic approaches. Moreover, these models are used extensively outside the NPC field to understand fundamental aspects of
cholesterol homeostasis [391] in different organs, notably the brain [392], and mechanisms of other cholesterol-related diseases [393–395].

The publication record indicates a clear preference for NPC1, mice, and fibroblasts as gene, animal, and cell of choice, respectively. A few points should be considered with respect to future developments and advances. The focus on NPC1 is understandable given that most patients bear mutations in this gene. However, new models targeting NPC2 are of high interest, as they can help for example to discern NPC1- and NPC2-dependent genetic, epigenetic, and sex-dependent disease modifiers. The identification of modifiers remains a top priority in the field. The predominance of mouse models in NPC research is readily explained by the increasing ease of genetic manipulations and the relative cost efficacy. However, mice impose several limitations, notably with respect to their small size and their limited behavioral repertoire [396]. Therefore, new models based on larger mammals including rats are highly desirable last but not least to enable the successful translation of therapeutic approaches into the clinic [6]. There is also a clear demand for inducible/reversible pharmacologic models based on highly selective small molecule inhibitors of NPC1 or NPC2. These approaches would allow for before/after studies and thereby help to discern within-subject variability. The surprising discovery that NPC1 serves as receptor for filovirus entry into cells [303,304] will help to develop such inhibitors and new models.

The focus on fibroblasts originates from their availability through skin biopsies, their ease of maintenance, and their long-standing use as a diagnostic tool. However, studies of patient-derived fibroblasts cannot inform about the outcome of NPC1 dysfunction in highly specialized postmitotic cells such as neurons. Therefore, it is imperative to elucidate how specific cells, namely the most vulnerable, react to defects in NPC1 and NPC2. This will require a combination of preparations allowing to study the same type of cells in vivo, ex vivo, and in vitro (Figure 4) as well as new approaches to analyze mRNA, protein, and lipid content of defined cell types replacing transcriptomic, proteomic, and lipidomic studies of entire organs or tissues. As an example, acutely isolated cells combined with single cell transcriptomics [231] represent a first step that needs to be refined and extended with a focus on vulnerable cells in most affected organs, including the brain, liver, and lung. Cells differentiated from induced pluripotent stem cells represent an alternative although with caveats [397]. Whatever the source of cells, advanced culture systems preserving their three-dimensional arrangement should be considered as well [398,399]. The development of therapeutic approaches for neurologic and psychiatric symptoms faces fundamental hurdles with respect to diagnosis and model validity that are not specific to NPC [400,401].

Clearly, the establishment of new models requires substantial investments and bears risks, but ultimately, all that matters are the models: they are indispensable to expose molecular mechanisms underlying the disease and to develop efficient therapies.

**Author Contributions:** Conceptualization, V.P. and F.W.P.; methodology, F.W.P.; software, F.W.P.; visualization, F.W.P.; writing—original draft preparation, V.P. and F.W.P.; writing—review and editing, V.P. and F.W.P. All authors have read and agreed to the published version of the manuscript.

**Funding:** The APC for this review was funded by the Niemann-Pick Selbsthilfegruppe e.V. (Germany). The authors’ research is funded by Together Strong NPC Foundation (V.P.), Niemann-Pick Selbsthilfegruppe e.V., Ara Parseghian Medical Research Foundation and Bild hilft e.V. “Ein Herz für Kinder” (F.W.P).

**Conflicts of Interest:** The authors declare no conflict of interest.

**Appendix A**

To obtain quantitative data on the publication output in the field, Boolean queries were performed in PubMed and records were downloaded in csv format. Data analysis and visualization were accomplished using the open source software R [402] and selected R packages (data.table [403], ggplot2 [404], readr [405].
References

1. Boycott, K.M.; Vanstone, M.R.; Bulman, D.E.; MacKenzie, A.E. Rare-disease genetics in the era of next-generation sequencing: Discovery to translation. *Nat. Rev. Genet.* 2013, 14, 681–691. [CrossRef]

2. Ruz, C.; Alcantud, J.L.; Vives Montero, F.; Duran, R.; Bandres-Ciga, S. Proteotoxicity and Neurodegenerative Diseases. *Int. J. Mol. Sci.* 2020, 21, 5646. [CrossRef]

3. Tanner, L.; Single, A.B. Animal Models Reflecting Chronic Obstructive Pulmonary Disease and Related Respiratory Disorders: Translating Pre-Clinical Data into Clinical Relevance. *J. Innate Immun.* 2020, 12, 203–225. [CrossRef]

4. Justice, M.J.; Dhillon, P. Using the mouse to model human disease: Increasing validity and reproducibility. *Dis. Model. Mech.* 2016, 9, 101–103. [CrossRef]

5. Cacheiro, P.; Haendel, M.A.; Smedley, D. New models for human disease from the International Mouse Phenotyping Consortium. *Mamm. Genome* 2019, 30, 143–150. [CrossRef]

6. Gurda, B.L.; Vite, C.H. Large animal models contribute to the development of therapies for central and peripheral nervous system dysfunction in patients with lysosomal storage diseases. *Hum. Mol. Genet.* 2019, 28, R119–R131. [CrossRef]

7. Madeja, Z.E.; Pawlak, P.; Pliszek, A. Beyond the mouse: Non-rodent animal models for study of early mammalian development and biomedical research. *Int. J. Dev. Biol.* 2019, 63, 187–201. [CrossRef] [PubMed]

8. Slanzi, A.; Iannoto, G.; Rossi, B.; Zenaro, E.; Constantin, G. In vitro Models of Neurodegenerative Diseases. *Front. Cell Dev. Biol.* 2020, 8, 328. [CrossRef] [PubMed]

9. Bräuer, A.U.; Kuhla, A.; Holzmann, C.; Wree, A.; Witt, M. Current Challenges in Understanding the Cellular and Molecular Mechanisms in Niemann-Pick Disease Type C1. *Int. J. Mol. Sci.* 2019, 20, 4392. [CrossRef] [PubMed]

10. Braverman, T.; Song, C.; Li, J.; Gu, J.Z.; Yin, Z.; Huang, X.; Zhou, L.; Guo, N.; Zhu, N.Y.; Wen, Q.; et al. Identification of HE1 as the Second Gene of Niemann-Pick C Disease. *Mol. Chem. Neuropathol.* 2016, 74, 71–82. [CrossRef] [PubMed]
22. Gong, X.; Qian, H.; Zhou, X.; Wu, J.; Wan, T.; Cao, P.; Huang, W.; Zhao, X.; Wang, X.; Wang, P.; et al. Structural Insights into the Niemann-Pick C1 (NPC1)-Mediated Cholesterol Transfer and Ebola Infection. *Cell* 2016, 165, 1467–1478. [CrossRef] [PubMed]

23. Li, X.; Wang, J.; Coutavas, E.; Shi, H.; Hao, Q.; Blobel, G. Structure of human Niemann-Pick C1 protein. *Proc. Natl. Acad. Sci. USA* 2016, 113, 8212–8217. [CrossRef] [PubMed]

24. Li, X.; Lu, F.; Trinh, M.N.; Schmiege, P.; Seemann, J.; Wang, J.; Blobel, G. 3.3 Å structure of Niemann-Pick C1 protein reveals insights into the function of the C-terminal luminal domain in cholesterol transport. *Proc. Natl. Acad. Sci. USA* 2017, 114, 9116–9121. [CrossRef] [PubMed]

25. Winkler, M.B.L.; Kidmose, R.T.; Szomek, M.; Thaysen, K.; Rawson, S.; Muench, S.P.; Wüstner, D.; Pedersen, B.P. Structural Insight into Eukaryotic Sterol Transport through Niemann-Pick Type C Proteins. *Cell* 2019, 179, 485–497.e418. [CrossRef] [PubMed]

26. Pfisterer, S.G.; Peränen, J.; Ikonen, E. LDL-cholesterol transport to the endoplasmic reticulum: Current concepts. *Curr. Opin. Lipidol.* 2016, 27, 282–287. [CrossRef] [PubMed]

27. Pfiffer, S.R. NPC intracellular cholesterol transporter 1 (NPC1)-mediated cholesterol export from lysosomes. *J. Biol. Chem.* 2019, 294, 1706–1709. [CrossRef] [PubMed]

28. Meng, Y.; Heybrock, S.; Neculai, D.; Saftig, P. Cholesterol Handling in Lysosomes and Beyond. *Trends Cell Biol.* 2020, 30, 452–466. [CrossRef] [PubMed]

29. Sokol, J.; Blanchette-Mackie, J.; Kruth, H.S.; Dwyer, N.K.; Amende, L.M.; Butler, J.D.; Robinson, E.; Patel, S.; Brady, R.O.; Comly, M.E.; et al. Type C Niemann-Pick disease. Lysosomal accumulation and defective intracellular mobilization of low density lipoprotein cholesterol. *J. Biol. Chem.* 1988, 263, 3411–3417. [PubMed]

30. Liscum, L.; Ruggiero, R.M.; Faust, J.R. The intracellular transport of low density lipoprotein-derived cholesterol is defective in Niemann-Pick type C fibroblasts. *J. Cell Biol.* 1989, 108, 1625–1636. [CrossRef] [PubMed]

31. Breiden, B.; Sandhoff, K. Mechanism of Secondary Ganglioside and Lipid Accumulation in Lysosomal Disease. *Int. J. Mol. Sci.* 2020, 21, 2566. [CrossRef]

32. Lloyd-Evans, E.; Morgan, A.J.; He, X.; Smith, D.A.; Elliot-Smith, E.; Sillence, D.J.; Churchill, G.C.; Schuchman, E.H.; Gallone, A.; Platt, F.M. Niemann-Pick disease type C1 is a sphingosine storage disease that causes deregulation of lysosomal calcium. *Nat. Med.* 2008, 14, 1247–1255. [CrossRef]

33. Patterson, M.C.; Hendriksz, C.J.; Walterfang, M.; Sedel, F.; Vanier, M.T.; Wijburg, F. Recommendations for the diagnosis and management of Niemann-Pick disease type C: An update. *Mol. Genet. Metab.* 2012, 106, 330–344. [CrossRef]

34. Patterson, M.C.; Mengel, E.; Wijburg, F.A.; Muller, A.; Schwierin, B.; Drevon, H.; Vanier, M.T.; Pineda, M. Disease and patient characteristics in NP-C patients: Findings from an international disease registry. *Orphanet J. Rare Dis.* 2013, 8, 12. [CrossRef]

35. Mengel, E.; Klinemann, H.H.; Lourenço, C.M.; Hendriksz, C.J.; Sedel, F.; Walterfang, M.; Kolb, S.A. Niemann-Pick disease type C symptomatology: An expert-based clinical description. *Orphanet J. Rare Dis.* 2013, 8, 166. [CrossRef]

36. Mengel, E.; Pineda, M.; Hendriksz, C.J.; Walterfang, M.; Torres, J.V.; Kolb, S.A. Differences in Niemann-Pick disease Type C symptomatology observed in patients of different ages. *Mol. Genet. Metab.* 2017, 120, 180–189. [CrossRef]

37. Bianconi, S.E.; Hammond, D.I.; Farhat, N.Y.; Dang Do, A.; Jenkins, K.; Cougnoux, A.; Martin, K.; Porter, F.D. Evaluation of age of death in Niemann-Pick disease, type C: Utility of disease support group websites to understand natural history. *Mol. Genet. Metab.* 2019, 126, 466–469. [CrossRef]

38. Patterson, M.C.; Garver, W.S.; Giugliani, R.; Imrie, J.; Jahnova, H.; Meaney, F.J.; Nadjar, Y.; Vanier, M.T.; Moneuse, P.; Morand, O.; et al. Long-term survival outcomes of patients with Niemann-Pick disease type C receiving miglustat treatment: A large retrospective observational study. *J. Inherit. Metab. Dis.* 2020, 43, 1060–1069. [CrossRef]

39. Spiegel, R.; Raas-Rothschild, A.; Reish, O.; Regev, M.; Meiner, V.; Bargal, R.; Sury, V.; Meir, K.; Nadjari, M.; Hermann, G.; et al. The clinical spectrum of fetal Niemann-Pick type C. *Am. J. Med. Genet. A* 2009, 149, 446–450. [CrossRef]
40. Jahnova, H.; Dvorakova, L.; Vlaskova, H.; Hulko, H.; Poupetova, H.; Hrebicek, M.; Jesina, P. Observational, retrospective study of a large cohort of patients with Niemann-Pick disease type C in the Czech Republic: A surprisingly stable diagnostic rate spanning almost 40 years. *Orphanet J. Rare Dis.* 2014, 9, 140. [CrossRef]

41. Imrie, J.; Heptinstall, L.; Knight, S.; Strong, K. Observational cohort study of the natural history of Niemann-Pick disease type C in the UK: A 5-year update from the UK clinical database. *BMC Neurol.* 2015, 15, 257. [CrossRef] [PubMed]

42. Gumus, E.; Haliloglu, G.; Karhan, A.N.; Demir, H.; Gurakan, F.; Topcu, M.; Yuce, A. Niemann-Pick disease type C in the newborn period: A single-center experience. *Eur. J. Pediatr.* 2017, 176, 1669–1676. [CrossRef] [PubMed]

43. Staretz-Chacham, O.; Aviram, M.; Morag, I.; Goldbart, A.; Hershkovitz, E. Pulmonary involvement in Niemann-Pick C type 1. *Eur. J. Pediatric* 2018, 177, 1609–1615. [CrossRef] [PubMed]

44. Pineda, M.; Juríčková, K.; Karimzadeh, P.; Kohnikova, M.; Malinová, V.; Insúa, J.L.; Velen, C.; Kolb, S.A. Disease characteristics, prognosis and miglustat treatment effects on disease progression in patients with Niemann-Pick disease Type C: An international, multicenter, retrospective chart review. *Orphanet J. Rare Dis.* 2019, 14, 32. [CrossRef]

45. Yilmaz, B.S.; Baruteau, J.; Rahim, A.A.; Gissen, P. Clinical and Molecular Features of Early Infantile Niemann Pick Type C disease. *Int. J. Mol. Sci.* 2020, 21, 5059. [CrossRef]

46. Di Lazzaro, V.; Marano, M.; Florio, L.; De Santis, S. Niemann-Pick type C: Focus on the adolescent/adult onset form. *Int. J. Neurosci.* 2016, 126, 963–971. [CrossRef]

47. Wassif, C.A.; Cross, J.L.; Iben, J.; Sanchez-Pulido, L.; Cougnoux, A.; Platt, F.M.; Ory, D.S.; Ponting, C.P.; Bailey-Wilson, J.E.; Biesecker, L.G.; et al. High incidence of unrecognized visceral/neurological late-onset Niemann-Pick disease, type C1, predicted by analysis of massively parallel sequencing data sets. *Genet. Med.* 2016, 18, 41–48. [CrossRef]

48. Bonnot, O.; Klünnemann, H.H.; Velen, C.; Torres Martin, J.V.; Walterfang, M. Systematic review of psychiatric signs in Niemann-Pick disease type C. *World J. Biol. Psychiatry* 2019, 20, 320–332. [CrossRef]

49. Nadjar, Y.; Hütter-Moncada, A.L.; Latour, P.; Ayrignac, X.; Kaphan, E.; Tranchant, C.; Cintas, P.; Degardin, A.; Goizet, C.; Laurenccin, C.; et al. Adult Niemann-Pick disease type C in France: Clinical phenotypes and long-term miglustat treatment effect. *Orphanet J. Rare Dis.* 2018, 13, 175. [CrossRef]

50. Rego, T.; Farrand, S.; Goh, A.M.Y.; Eratne, D.; Kelso, W.; Mangelsdorf, S.; Velakoulis, D.; Walterfang, M. Psychiatric and Cognitive Symptoms Associated with Niemann-Pick Type C Disease: Neurobiology and Management. *CNS Drugs* 2019, 33, 125–142. [CrossRef]

51. Porter, F.D.; Scherrer, D.E.; Lanier, M.H.; Langmade, S.J.; Molugu, V.; Gale, S.E.; Olzeski, D.; Sidhu, R.; Dietzen, D.J.; Fu, R.; et al. Cholesterol oxidation products are sensitive and specific blood-based biomarkers for Niemann-Pick C1 disease. *Sci. Transl. Med.* 2010, 2, 56ra81. [CrossRef] [PubMed]

52. Giese, A.K.; Mascher, H.; Grittner, U.; Eichler, S.; Kramp, G.; Lukas, J.; Te Vruchte, D.; Al Eisa, N.; Cortina-Borja, M.; Porter, F.D.; et al. A novel, highly sensitive and specific biomarker for Niemann-Pick type C1 disease. *Orphanet J. Rare Dis.* 2015, 10, 78. [CrossRef] [PubMed]

53. Jiang, X.; Sidhu, R.; Mydock-McGrane, L.; Hsu, F.F.; Covey, D.F.; Scherrer, D.E.; Earley, B.; Gale, S.E.; Farhat, N.Y.; Porter, F.D.; et al. Development of a bile acid-based newborn screen for Niemann-Pick disease type C. *Sci. Transl. Med.* 2016, 8, 337ra363. [CrossRef]

54. Maekawa, M.; Jinnoh, I.; Matsumoto, Y.; Narita, A.; Mashima, R.; Takahashi, H.; Iwahori, A.; Saigusa, D.; Fujii, K.; Abe, A.; et al. Structural Determination of Lysoosphingomyelin-509 and Discovery of Novel Class Lipids from Patients with Niemann-Pick Disease Type C. *Int. J. Mol. Sci.* 2019, 20, 5018. [CrossRef]

55. Sidhu, R.; Kell, P.; Dietzen, D.J.; Farhat, N.Y.; Do, A.N.D.; Porter, F.D.; Berry-Kravis, E.; Vite, C.H.; Reunert, J.; Marquardt, T.; et al. Application of N-palmitoyl-O-phosphocholine for diagnosis and assessment of response to treatment in Niemann-Pick type C disease. *Mol. Genet. Metab.* 2020, 129, 292–302. [CrossRef]

56. Vanier, M.T.; Gissen, P.; Bauer, P.; Coll, M.J.; Burlina, A.; Hendriksz, C.J.; Latour, P.; Goizet, C.; Welford, R.W.; Marquardt, T.; et al. Diagnostic tests for Niemann-Pick disease type C (NP-C): A critical review. *Mol. Genet. Metab.* 2016, 118, 244–254. [CrossRef]

57. Patterson, M.C.; Clayton, P.; Gissen, P.; Anheim, M.; Bauer, P.; Bonnot, O.; Dardis, A.; Dionisi-Vici, C.; Klünnemann, H.H.; Latour, P.; et al. Recommendations for the detection and diagnosis of Niemann-Pick disease type C: An update. *Neurol. Clin. Pract.* 2017, 7, 499–511. [CrossRef] [PubMed]
58. Sitarska, D.; Ługowska, A. Laboratory diagnosis of the Niemann-Pick type C disease: An inherited neurodegenerative disorder of cholesterol metabolism. *Metab. Brain Dis.* 2019, 34, 1253–1260. [CrossRef] [PubMed]

59. Vanier, M.T.; Latour, P. Laboratory diagnosis of Niemann-Pick disease type C: The filipin staining test. *Methods Cell Biol.* 2015, 126, 357–375.

60. Patterson, M.C.; Vecchio, D.; Prady, H.; Abel, L.; Wraith, J.E. Miglustat for treatment of Niemann-Pick C disease: A randomised controlled study. *Lancet Neurol.* 2007, 6, 765–772. [CrossRef]

61. Pineda, M.; Walterfang, M.; Patterson, M.C. Miglustat in Niemann-Pick disease type C patients: A review. *Orphanet J. Rare Dis.* 2018, 13, 140. [CrossRef]

62. Stirnemann, J.; Belmatoug, N.; Camou, F.; Serratrice, C.; Froissart, R.; Caillaud, C.; Levade, T.; Astudillo, L.; Serratrice, J.; Brassier, A.; et al. A Review of Gaucher Disease Pathophysiology, Clinical Presentation and Treatments. *Int. J. Mol. Sci.* 2017, 18, 441. [CrossRef]

63. Platt, F.M. Emptying the stores: Lysosomal diseases and therapeutic strategies. *Nat. Rev. Drug Discov.* 2018, 17, 133–150. [CrossRef]

64. Harvey, W.; Leake, C.D. Exercitatio anatomica de motu cordis et sanguinis in animalibus. *Am. J. Med. Sci.* 1929, 177, 578. [CrossRef]

65. Descartes, R. *Discours de la Methode Pour Bien Conduire la Raison et Chercher la Verite Dans les Sciences*; De l’imprimerie de Ian Maire: Ian Maire, Leyden, 1637.

66. Bentham, J. *An Introduction to the Principles of Morals and Legislation*; T. Payne and Sons: London, UK, 1789.

67. Russell, W.M.S.B. *The Principles of Humane Experimental Technique*; Universities Federation for Animal Welfare: Wheathampstead, UK, 1959.

68. Tannenbaum, J.; Bennett, B.T. Russell and Burch’s 3Rs then and now: The need for clarity in definition and purpose. *J. Am. Assoc. Lab. Anim. Sci.* 2015, 54, 120–132.

69. Flanagan, S.P. ‘Nude’, a new hairless gene with pleiotropic effects in the mouse. *Genet. Res.* 1966, 8, 295–309. [CrossRef] [PubMed]

70. Lai, Y.; Wei, X.; Lin, S.; Qin, L.; Cheng, L.; Li, P. Current status and perspectives of patient-derived xenograft models in cancer research. *J. Hematol. Oncol.* 2017, 10, 106. [CrossRef]

71. Arison, R.N.; Ciaccio, E.I.; Glitzer, M.S.; Cassaro, J.A.; Pruss, M.P. Light and Electron Microscopy of Lesions in Rats Rendered Diabetic with Streptozotocin. *Diabetes* 1967, 16, 51–56. [CrossRef]

72. Lazarus, S.S.; Shapiro, S.H. Serial morphologic changes in rabbit pancreatic islet cells after streptozotocin. *Lab. Investig.* 1972, 27, 174–183.

73. Burns, R.S.; Chiueh, C.C.; Markey, S.P.; Ebert, M.H.; Jacobowitz, D.M.; Kopin, I.J. A primate model of parkinsonism: Selective destruction of dopaminergic neurons in the pars compacta of the substantia nigra by N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine. *Proc. Natl. Acad. Sci. USA* 1983, 80, 4546–4550. [CrossRef] [PubMed]

74. Servadio, M.; Vanderschuren, L.J.; Trezza, V. Modeling autism-relevant behavioral phenotypes in rats and mice. *Behav. Pharmacol.* 2015, 26, 522–540. [CrossRef] [PubMed]

75. Lunardi Baccetto, S.; Lehmann, C. Microcirculatory Changes in Experimental Models of Stroke and CNS-Injury Induced Immunodepression. *Int. J. Mol. Sci.* 2019, 20, 5184. [CrossRef] [PubMed]

76. Wagner, L.; Pannicke, T.; Rupprecht, V.; Frommherz, I.; Volz, C.; Illes, P.; Hirrlinger, J.; Jägle, H.; Egger, V.; Haydon, P.G.; et al. Suppression of SNARE-dependent exocytosis in retinal glial cells and its effect on ischemia-induced neurodegeneration. *Glia* 2017, 65, 1059–1071. [CrossRef] [PubMed]

77. Karamian, P.; Burford, J.; Farzad, S.; Blair, N.P.; Shahidi, M. Alterations in Retinal Oxygen Delivery, Metabolism, and Extraction Fraction During Bilateral Common Carotid Artery Occlusion in Rats. *Investig. Ophthalmol. Vis. Sci.* 2019, 60, 3247–3253. [CrossRef]

78. Fiorentino, S.; Melillo, G.; Fedele, G.; Clavenna, G.; D’Agostino, C.; Mainetti, E.; Caselli, G.F. Ketoprofen lysine salt inhibits disuse-induced osteopenia in a new non-traumatic immobilization model in the rat. *Pharmacol. Res.* 1996, 33, 277–281. [CrossRef]

79. Hanahan, D.; Wagner, E.F.; Palmer, R.D. The origins of oncomice: A history of the first transgenic mice genetically engineered to develop cancer. *Genes Dev.* 2007, 21, 2258–2270. [CrossRef]

80. Cooper, J.F.; Van Raamsdonk, J.M. Modeling Parkinson’s Disease in C. elegans. *J. Park. Dis.* 2018, 8, 17–32. [CrossRef]
81. Fang, E.F.; Hou, Y.; Palikaras, K.; Adriaanse, B.A.; Kerr, J.S.; Yang, B.; Lautrup, S.; Hasan-Olive, M.M.; Caponio, D.; Dan, X.; et al. Mitophagy inhibits amyloid-β and tau pathology and reverses cognitive deficits in models of Alzheimer’s disease. *Nat. Neurosci.* 2019, 22, 401–412. [CrossRef]

82. Morley, J.F.; Brignull, H.R.; Weyers, J.J.; Morimoto, R.I. The threshold for polyglutamine-expansion protein aggregation and cellular toxicity is dynamic and influenced by aging in Caenorhabditis elegans. *Proc. Natl. Acad. Sci. USA* 2002, 99, 10417–10422. [CrossRef]

83. de Voer, G.; Peters, D.; Taschner, P.E. Caenorhabditis elegans as a model for lysosomal storage disorders. *Biochim. Biophys. Acta* 2008, 1782, 433–446. [CrossRef]

84. Jeibmann, A.; Paulus, W. *Drosophila melanogaster* as a Model Organism of Brain Diseases. In *Int. J. Mol. Sci.* 2009, 10, 407–440. [CrossRef] [PubMed]

85. Prüfing, K.; Voigt, A.; Schulz, J.B. *Drosophila melanogaster* as a model organism for Alzheimer’s disease. *Mol. Neurodegener.* 2013, 8, 35. [CrossRef] [PubMed]

86. Smith, P.; Arias, R.; Sorti, S.; Odgerel, Z.; Santa-Maria, I.; McCabe, B.D.; Tsaneva-Atanasova, K.; Louis, E.D.; Hodge, J.J.L.; Clark, L.N. A *Drosophila* Model of Essential Tremor. *Sci. Rep.* 2018, 8, 7664. [CrossRef] [PubMed]

87. Banderali, G.; Riva, E.; Fiocchi, A.; Cordaro, C.I.; Giovannini, M. *Drosophila melanogaster* as a model organism for Alzheimer’s disease. *PLoS ONE* 2011, 6, e20022. [CrossRef]

88. King, N.W. *Simian Models of Acquired Immunodeficiency Syndrome (AIDS): A Review.* In *Vet. Pathol.* 2004, 41, S7. [CrossRef] [PubMed]

89. Mann, R.D. Modern Drug Use: An Enquiry on Historical Principles; MTP Press: Lancaster, UK, 1984; p. 769.

90. Luo, Y.L.; Li, P.B.; Zhang, C.C.; Zheng, Y.F.; Wang, S.; Nie, Y.C.; Zhang, K.J.; Su, W.W. *Caenorhabditis elegans* as a model for lysosomal storage disorders. *Biochim. Biophys. Acta* 2007, 1740, 338–348. [CrossRef] [PubMed]

91. Yamawaki, I.; Geppetti, P.; Bertrand, C.; Huber, O.; Da Prüfing, K.; Voigt, A.; Schulz, J.B. *Drosophila melanogaster* as a model organism for Alzheimer’s disease. *Mol. Neurodegener.* 2013, 8, 35. [CrossRef] [PubMed]

92. Milton, S.L.; Prentice, H.M. Beyond anoxia: The physiology of metabolic downregulation and recovery in the anoxia-tolerant turtle. *Comp. Biochem. Physiol. A Mol. Integr. Physiol.* 2004, 147, 383–388. [CrossRef]

93. Rosen, B.H.; Chanson, M.; Gawenis, L.R.; Liu, J.; Sofoluwe, A.; Zoso, A.; Engelhardt, J.F. Animal and model systems for studying cystic fibrosis. *J. Cyst. Fibros.* 2018, 17, S28–S34. [CrossRef] [PubMed]

94. Balamayooran, G.; Pena, M.; Sharma, R.; Truman, R.W. The armadillo as an animal model and reservoir host for Mycobacterium leprae. *Clin. Dermatol.* 2015, 33, 108–115. [CrossRef] [PubMed]

95. Balamayooran, G.; Pena, M.; Sharma, R.; Truman, R.W. The armadillo as an animal model and reservoir host for Mycobacterium leprae. *Clin. Dermatol.* 2015, 33, 108–115. [CrossRef] [PubMed]

96. Hastings, J.M.; Fazleabas, A.T. A baboon model for endometriosis: Implications for fertility. *Reprod. Biol. Endocrinol.* 2006, 4, S7. [CrossRef] [PubMed]

97. Balamayooran, G.; Pena, M.; Sharma, R.; Truman, R.W. The armadillo as an animal model and reservoir host for Mycobacterium leprae. *Clin. Dermatol.* 2015, 33, 108–115. [CrossRef] [PubMed]

98. Milton, S.L.; Prentice, H.M. Beyond anoxia: The physiology of metabolic downregulation and recovery in the anoxia-tolerant turtle. *Comp. Biochem. Physiol. A Mol. Integr. Physiol.* 2007, 147, 277–290. [CrossRef] [PubMed]

99. Trevino, M.; Lobarinias, E.; Maulden, A.C.; Heinz, M.G. The chinchilla animal model for hearing science and noise-induced hearing loss. *J. Acoust. Soc. Am.* 2019, 146, 3710. [CrossRef]

100. Verra, D.M.; Saletta, A.; Albertini, A.; Boschetti, A.; D’Arminio Monforte, A.; Haddad, G. *Drosophila melanogaster* as a model system for studies of islet amyloid polypeptide aggregation. *PLoS ONE* 2011, 6, e20022. [CrossRef]

101. Caponio, D.; Dan, X.; et al. Mitophagy inhibits amyloid-β and tau pathology and reverses cognitive deficits in models of Alzheimer’s disease. *Nat. Neurosci.* 2019, 22, 401–412. [CrossRef]

102. Sellgren, C.M.; Gracias, J.; Watmuff, B.; Biag, J.D.; Thanos, J.M.; Whittredge, P.B.; Fu, T.; Worringer, K.; Brown, H.E.; Wang, J.; et al. Increased synapse elimination by microglia in schizophrenia patient-derived models of synaptic pruning. *Nat. Neurosci.* 2019, 22, 374–385. [CrossRef]

103. Fog, C.K.; Kirkegaard, T. Animal models for Niemann-Pick type C: Implications for drug discovery & development. *Expert Opin. Drug Discov.* 2019, 14, 499–509.
104. Higaki, K.; Almanzar-Paramio, D.; Sturley, S.L. Metazoan and microbial models of Niemann-Pick Type C disease. *Biochim. Biophys. Acta* 2004, 1685, 38–47. [CrossRef]  
105. Feldman, M.J.; Poirier, B.C.; Lange, B.M. Misexpression of the Niemann-Pick disease type C1 (NPC1)-like protein in Arabidopsis causes sphingolipid accumulation and reproductive defects. *Planta* 2015, 242, 921–933. [CrossRef] [PubMed]  
106. Malathi, K.; Higaki, K.; Tinkelenberg, A.H.; Balderes, D.A.; Almanzar-Paramio, D.; Wilcox, L.J.; Erdeniz, N.; Redican, F.; Padamsee, M.; Liu, Y.; et al. Mutagenesis of the putative sterol-sensing domain of yeast Niemann-Pick C-related protein reveals a primordial role in subcellular sphingolipid distribution. *J. Cell Biol.* 2004, 164, 547–556. [CrossRef] [PubMed]  
107. Zhu, J.; Wang, G.; Pelosi, P. Plant transcriptomes reveal hidden guests. *Biochim. Biophys. Res. Commun.* 2016, 474, 497–502. [CrossRef] [PubMed]  
108. Berger, A.C.; Vanderford, T.H.; Gernert, K.M.; Nichols, J.W.; Faundez, V.; Corbett, A.H. *Saccharomyces cerevisiae* Npc2p is a functionally conserved homologue of the human Niemann-Pick disease type C 2 protein, hNPC2. *Eukaryot. Cell* 2005, 4, 1851–1862. [CrossRef] [PubMed]  
109. Munkaci, A.B.; Chen, F.W.; Brinkman, M.A.; Higaki, K.; Gutiérrez, G.D.; Chaudhari, J.; Layer, J.V.; Tong, A.; Bard, M.; Boone, C.; et al. An “exacerbate-reverse” strategy in yeast identifies histone deacetylase inhibition as a correction for cholesterol and sphingolipid transport defects in human Niemann-Pick type C disease. *J. Biol. Chem.* 2011, 286, 23842–23851. [CrossRef]  
110. Tsuji, T.; Fujimoto, M.; Tatematsu, T.; Cheng, J.; Orii, M.; Takatori, S.; Fujimoto, T. Niemann-Pick type C proteins promote microautophagy by expanding raft-like membrane domains in the yeast vacuole. *eLife* 2017, 6, e25960. [CrossRef]  
111. Colaco, A.; Fernández-Suárez, M.E.; Shepherd, D.; Gal, L.; Bibi, C.; Chuetrazman, S.; Diet, A.; Morten, K.; Eden, E.; Porter, F.D.; et al. Unbiased yeast screens identify cellular pathways affected in Niemann-Pick disease type C. *Life Sci. Alliance* 2020, 3, e201800253. [CrossRef]  
112. Bolaños, J.; Betanzos, A.; Javier-Reyna, R.; García-Rivera, G.; Huerta, M.; Pais-Morales, J.; González-Robles, A.; Rodríguez, M.A.; Schnoor, M.; Orozco, E. EhNPC1 and EhNPC2 Proteins Participate in Trafficking of Exogenous Cholesterol in Entamoeba histolytica Trophozoites: Relevance for Phagocytosis. *PLoS Pathog.* 2016, 12, e1006089. [CrossRef]  
113. Sym, M.; Basson, M.; Johnson, C. A model for Niemann–Pick type C disease in the nematode Caenorhabditis elegans. *Curr. Biol.* 2000, 10, 527–530. [CrossRef]  
114. Li, J.; Brown, G.; Allion, M.; Lee, S.; Thomas, J.H. NCR-1 and NCR-2, the *C. elegans* homologs of the human Niemann-Pick disease 1 and 2 protein, function upstream of DAF-9 in the dauer formation pathways. *Development* 2004, 131, 5741–5752. [CrossRef]  
115. Wüstner, D.; Landt Larsen, A.; Faergeman, N.J.; Brewer, J.R.; Sage, D. Selective visualization of fluorescent proteins promote microautophagy by expanding raft-like membrane domains in the yeast vacuole. *eLife* 2017, 6, e25960. [CrossRef]  
116. Smith, M.M.; Levitan, D.J. Human NPC1L1 and NPC1 can functionally substitute for the ncr genes to promote reproductive development in *C. elegans*. *Biochim. Biophys. Acta* 2007, 1770, 1345–1351. [CrossRef] [PubMed]  
117. Boland, S.; Schmidt, U.; Zagarov, V.; Sampaio, J.L.; Czerwonka, R.; Lübben, T.; Reimann, J.; Penkov, S.; Knöller, H.J.; et al. Phosphorylated glycosphingolipids essential for cholesterol mobilization in Caenorhabditis elegans. *Nat. Chem. Biol.* 2017, 13, 647–654. [CrossRef] [PubMed]  
118. Galles, C.; Prez, G.M.; Penkov, S.; Boland, S.; Porta, E.O.J.; Altabe, S.G.; Labadie, G.R.; Schmidt, U.; Knöller, H.J.; Kurzchalia, T.V.; et al. Endocannabinoids in Caenorhabditis elegans are essential for the mobilization of cholesterol from internal reserves. *Sci. Rep.* 2018, 8, 6398. [CrossRef]  
119. Huang, X.; Suyama, K.; Buchanan, J.; Zhu, A.J.; Scott, M.P. A *Drosophila* model of the Niemann-Pick type C lysosome storage disease: Dnpc1a is required for molting and sterol homeostasis. *Development* 2005, 132, 5115–5124. [CrossRef]  
120. Fluegel, M.L.; Parker, T.J.; Pallanck, L.J. Mutations of a *Drosophila* NPC1 gene confer sterol and ecdysone metabolic defects. *Genetics* 2006, 172, 185–196. [CrossRef]  
121. Voght, S.P.; Fluegel, M.L.; Andrews, L.A.; Pallanck, L.J. *Drosophila* NPC1b promotes an early step in sterol absorption from the midgut epithelium. *Cell Metab.* 2007, 5, 195–205. [CrossRef]
122. Huang, X.; Warren, J.T.; Buchanan, J.; Gilbert, L.I.; Scott, M.P. Drosophila Niemann-Pick Type C-2 genes control sterol homeostasis and sterol biosynthesis: A model of human neurodegenerative disease. Development 2007, 134, 3733–3742. [CrossRef]

123. Phillips, S.E.; Woodruff, E.A., III; Liang, P.; Patten, M.; Broadie, K. Neuronal loss of Drosophila NPC1a causes cholesterol aggregation and age-progressive neurodegeneration. J. Neurosci. 2008, 28, 6569–6582. [CrossRef]

124. Danielsen, E.T.; Moeller, M.E.; Yamanaka, N.; Ou, Q.; Laursen, J.M.; Soenderholm, C.; Zhuo, R.; Phelps, B.; Tang, K.; Zeng, J.; et al. A Drosophila Genome-Wide Screen Identifies Regulators of Steroid Hormone Production and Developmental Timing. Dev. Cell 2016, 37, 558–570. [CrossRef]

125. Schwend, T.; Loucks, E.J.; Snyder, D.; Ahlgren, S.C. Requirement of Npc1 and availability of cholesterol for early embryonic cell movements in zebrafish. J. Lipid Res. 2011, 52, 1328–1344. [CrossRef] [PubMed]

126. Lin, Y.; Cai, X.; Wang, G.; Ouyang, G.; Cao, H. Model construction of Niemann-Pick type C disease in zebrafish. Biol. Chem. 2018, 399, 903–910. [CrossRef] [PubMed]

127. Tseng, W.-C.; Loeb, H.E.; Pei, W.; Tsai-Morris, C.-H.; Xu, L.; Cluzeau, C.V.; Wassif, C.A.; Feldman, B.; Burgess, S.M.; Pavan, W.J.; et al. Modeling Niemann-Pick disease type C1 in zebrafish: A robust platform for in vivo screening of candidate therapeutic compounds. Dis. Model. Mech. 2018, 11, dmm034165. [CrossRef] [PubMed]

128. Pacheco, C.D.; Yu, T.; Dadgar, N.; Shakkottai, V.G.; Ware, C.; Paulson, H.L.; Lieberman, A.P. Conditional Niemann-Pick C mice demonstrate cell autonomous Purkinje cell neurodegeneration. Hum. Mol. Genet. 2010, 19, 837–847. [CrossRef] [PubMed]

129. Rinkunas, V.M.; Graham, M.J.; Crooke, R.M.; Liscum, L. In vivo antisense oligonucleotide reduction of NPC1 expression as a novel mouse model for Niemann-Pick Type C-associated liver disease. Hepatology 2008, 47, 1504–1512. [CrossRef] [PubMed]

130. Zhang, M.; Stratnaka, D.; Donohue, C.; Hallows, J.L.; Vincent, I.; Erickson, R.P. Astrocyte-only Npc1 reduces neuronal cholesterol and triples life span of Npc1−/− mice. J. Neurosci. Res. 2008, 86, 2848–2856. [CrossRef] [PubMed]

131. Elrick, M.J.; Pacheco, C.D.; Yu, T.; Dadgar, N.; Shakkottai, V.G.; Ware, C.; Paulson, H.L.; Lieberman, A.P. Conditional Niemann-Pick C mice demonstrate cell autonomous Purkinje cell neurodegeneration. Hum. Mol. Genet. 2010, 19, 837–847. [CrossRef] [PubMed]

132. Praggastis, M.; Tortelli, B.; Zhang, J.; Fujiwara, H.; Sidhu, R.; Chacko, A.; Chen, Z.; Chung, C.; Lieberman, A.P.; Pentchev, P.G.; Gal, A.E.; Booth, A.D.; Omodeo-Sale, F.; Fours, J.; Neumeyer, B.A.; Quirk, J.M.; Dawson, G.; Tseng, W.-C.; Loeb, H.E.; Pei, W.; Tsai-Morris, C.-H.; Xu, L.; Cluzeau, C.V.; Wassif, C.A.; Feldman, B.; Burgess, S.M.; Pavan, W.J.; et al. Modeling Niemann-Pick disease type C1 in zebrafish: A robust platform for in vivo screening of candidate therapeutic compounds. Dis. Model. Mech. 2018, 11, dmm034165. [CrossRef] [PubMed]

133. Xie, X.; Brown, M.S.; Shelton, J.M.; Richardson, J.A.; Goldstein, J.L.; Liang, G. Amino acid substitution in NPC1 that abolishes cholesterol binding reproduces phenotype of complete NPC1 deficiency in mice. Proc. Natl. Acad. Sci. USA 2011, 108, 15330–15335. [CrossRef]

134. Miyawaki, S.; Mitsuoka, S.; Sakiyama, T.; Kitagawa, T. Sphingomyelinosis, a new mutation in the mouse: A model of Niemann-Pick disease in humans. J. Hered. 1982, 73, 257–263. [CrossRef]

135. Rodriguez-Gil, J.L.; Watkins-Chow, D.E.; Baxter, L.L.; Elliot, G.; Harper, U.L.; Wincovitch, S.M.; Wedel, J.C.; Incao, A.A.; Huebecker, M.; Boehm, F.; et al. Genetic background modifies phenotypic severity and longevity in a mouse model of Niemann-Pick disease type C1. Dis. Model. Mech. 2020, 13, dmm042614. [CrossRef] [PubMed]

136. Elrick, M.J.; Pacheco, C.D.; Yu, T.; Dadgar, N.; Shakkottai, V.G.; Ware, C.; Paulson, H.L.; Lieberman, A.P. Conditional Niemann-Pick C mice demonstrate cell autonomous Purkinje cell neurodegeneration. Hum. Mol. Genet. 2010, 19, 837–847. [CrossRef] [PubMed]

137. Sikora, J.; et al. A murine Niemann-Pick C1 I1061T knock-in model recapitulates the pathological features of the most prevalent human disease allele. J. Neurosci. 2015, 35, 8091–8106. [CrossRef]

138. Gómez-Grau, M.; Albaigés, J.; Casas, J.; Auladell, C.; Dierssen, M.; Vilageliu, L.; Grinberg, D. New murine Niemann-Pick type C models bearing a pseudoexon-generating mutation recapitulates the pathological features of the most prevalent human disease allele. J. Neurosci. 2015, 35, 8091–8106. [CrossRef]

139. Rodriguez-Gil, J.L.; Watkins-Chow, D.E.; Baxter, L.L.; Elliot, G.; Harper, U.L.; Wincovitch, S.M.; Wedel, J.C.; Incao, A.A.; Huebecker, M.; Boehm, F.; et al. Genetic background modifies phenotypic severity and longevity in a mouse model of Niemann-Pick disease type C1. Dis. Model. Mech. 2020, 13, dmm042614. [CrossRef] [PubMed]

140. Elrick, M.J.; Pacheco, C.D.; Yu, T.; Dadgar, N.; Shakkottai, V.G.; Ware, C.; Paulson, H.L.; Lieberman, A.P. Conditional Niemann-Pick C mice demonstrate cell autonomous Purkinje cell neurodegeneration. Hum. Mol. Genet. 2010, 19, 837–847. [CrossRef] [PubMed]

141. Rinkunas, V.M.; Graham, M.J.; Crooke, R.M.; Liscum, L. In vivo antisense oligonucleotide reduction of NPC1 expression as a novel mouse model for Niemann-Pick Type C-associated liver disease. Hepatology 2008, 47, 1504–1512. [CrossRef] [PubMed]

142. Zhang, M.; Stratnaka, D.; Donohue, C.; Hallows, J.L.; Vincent, I.; Erickson, R.P. Astrocyte-only Npc1 reduces neuronal cholesterol and triples life span of Npc1−/− mice. J. Neurosci. Res. 2008, 86, 2848–2856. [CrossRef] [PubMed]

143. Elrick, M.J.; Pacheco, C.D.; Yu, T.; Dadgar, N.; Shakkottai, V.G.; Ware, C.; Paulson, H.L.; Lieberman, A.P. Conditional Niemann-Pick C mice demonstrate cell autonomous Purkinje cell neurodegeneration. Hum. Mol. Genet. 2010, 19, 837–847. [CrossRef] [PubMed]

144. Rinkunas, V.M.; Graham, M.J.; Crooke, R.M.; Liscum, L. In vivo antisense oligonucleotide reduction of NPC1 expression as a novel mouse model for Niemann-Pick Type C-associated liver disease. Hepatology 2008, 47, 1504–1512. [CrossRef] [PubMed]

145. Zhang, M.; Stratnaka, D.; Donohue, C.; Hallows, J.L.; Vincent, I.; Erickson, R.P. Astrocyte-only Npc1 reduces neuronal cholesterol and triples life span of Npc1−/− mice. J. Neurosci. Res. 2008, 86, 2848–2856. [CrossRef] [PubMed]

146. Lopez, M.E.; Klein, A.D.; Dimbil, U.J.; Scott, M.P. Anatomically defined neuron-based rescue of neurodegenerative Niemann-Pick Type C disorder. J. Neurosci. 2011, 31, 4367–4378. [CrossRef]
140. Beltroy, E.P.; Richardson, J.A.; Horton, J.D.; Turley, S.D.; Dietschy, J.M. Cholesterol accumulation and liver cell death in mice with Niemann-Pick type C disease. *Hepatology* **2005**, *42*, 886–893. [CrossRef]

141. Li, H.; Repa, J.J.; Valasek, M.A.; Beltroy, E.P.; Turley, S.D.; German, D.C.; Dietschy, J.M. Molecular, anatomical, and biochemical events associated with neurodegeneration in mice with Niemann-Pick type C disease. *J. Neuropathol. Exp. Neurol.* **2005**, *64*, 323–333. [CrossRef]

142. Claudepierre, T.; Paques, M.; Simonutti, M.; Buard, I.; Sahel, J.; Maue, A.; Pichaud, S.; Pfierrer, F.W. Lack of Niemann-Pick type C1 induces age-related degeneration in the mouse retina. *Mol. Cell Neurosci.* **2010**, *43*, 164–176. [CrossRef]

143. Zervas, M.; Dobrenis, K.; Walkley, S.U. Neurons in Niemann-Pick disease type C accumulate gangliosides as well as unesterified cholesterol and undergo dendritic and axonal alterations. *J. Neuropathol. Exp. Neurol.* **2007**, *66*, 49–64. [CrossRef]

144. Reid, P.C.; Sakashita, N.; Sugii, S.; Ohno-Iwashita, Y.; Shimada, Y.; Hickey, W.F.; Chang, T.Y. A novel cholesterol stain reveals early neuronal cholesterol accumulation in the Niemann-Pick type C1 mouse brain. *J. Lipid Res.* **2004**, *45*, 582–591. [CrossRef]

145. Miquel, J.F.; Zanlungo, S. Transgenic overexpression of Niemann-Pick C2 protein promotes cholesterol gallstone formation in mice. *J. Hepatol.* **2001**, *34*, 588–591. [CrossRef] [PubMed]

146. Németh, P.; Tóth, T.; Lóczy, B.; Fülöp, Z.; Solymosi, J. Effect of cholesterol on PC12 cells. *Int. J. Mol. Sci.* **2020**, *21*, 8979. [CrossRef] [PubMed]

147. Ko, D.C.; Milenkovic, L.; Beier, S.M.; Manuel, H.; Buchanan, J.; Scott, M.P. Cell-autonomous death of cerebellar Purkinje neurons with autophagy in Niemann-Pick type C disease. *PLoS Genet.* **2011**, *7*, e10023897. [CrossRef] [PubMed]

148. Tanaka, J.; Nakamura, H.; Miyawaki, S. Cerebellar involvement in murine sphingomyelinosis: A new model of Niemann-Pick disease. *J. Neuropathol. Exp. Neurol.* **1988**, *47*, 291–300. [CrossRef]

149. Higashi, Y.; Murayama, S.; Pentechev, P.G.; Suzuki, K. Cerebellar degeneration in the Niemann-Pick type C mouse. *Acta Neuropathol.* **1993**, *85*, 175–184. [CrossRef]

150. Sarna, J.R.; Larouche, M.; Marzban, H.; Sillito, R.V.; Rancourt, D.E.; Hawkes, R. Patterned Purkinje cell degeneration in mouse models of Niemann-Pick type C disease. *J. Comp. Neurol.* **2003**, *456*, 279–291. [CrossRef] [PubMed]

151. Sarna, J.R.; Larouche, M.; Marzban, H.; Sillito, R.V.; Rancourt, D.E.; Hawkes, R. Patterned Purkinje cell degeneration in mouse models of Niemann-Pick type C disease. *J. Comp. Neurol.* **2003**, *456*, 279–291. [CrossRef] [PubMed]

152. Li, H.; Repa, J.J.; Valasek, M.A.; Beltroy, E.P.; Turley, S.D.; German, D.C.; Dietschy, J.M. Molecular, anatomical, and biochemical events associated with neurodegeneration in mice with Niemann-Pick type C disease. *J. Neuropathol. Exp. Neurol.* **2005**, *64*, 323–333. [CrossRef]

153. Li, H.; Repa, J.J.; Valasek, M.A.; Beltroy, E.P.; Turley, S.D.; German, D.C.; Dietschy, J.M. Molecular, anatomical, and biochemical events associated with neurodegeneration in mice with Niemann-Pick type C disease. *J. Neuropathol. Exp. Neurol.* **2005**, *64*, 323–333. [CrossRef]

154. Clausen, B.; de Lahunta, A. Feline Niemann-Pick disease in a boxer dog. *Am. J. Pathol.* **1988**, *131*, 189–197. [CrossRef]

155. Kaya, E.; Hoerner, C.; et al. Molecular basis for a new bovine model of Niemann-Pick type C disease. *PLoS ONE* **2020**, *15*, e0238697. [CrossRef]

156. Bosch, M.; Fajardo, A.; Alcalá-Valdés, R.; Fernández-Vidal, A.; Tebar, F.; Enrich, C.; Cardellach, F.; Pérez-Navarro, E.; Pol, A. Hepatic Primary and Secondary Cholesterol Deposition and Damage in Niemann-Pick Disease. *Am. J. Pathol.* **2016**, *186*, 517–523. [CrossRef]

157. Muñana, K.R.; Luttgen, P.J.; Thrall, M.A.; Wenger, D.A.; Wood, P.A.; de Lahunta, A. Feline sphingolipidosis resembling Niemann-Pick disease type C. *Acta Neuropathol.* **1990**, *81*, 189–197. [CrossRef]

158. Muñana, K.R.; Luttgen, P.J.; Thrall, M.A.; Wenger, D.A.; Wood, P.A.; de Lahunta, A. Feline sphingolipidosis resembling Niemann-Pick disease type C. *Acta Neuropathol.* **1990**, *81*, 189–197. [CrossRef]
160. Pacheco, C.D.; Kunkel, R.; Lieberman, A.P. Autophagy in Niemann-Pick C disease is dependent upon Beclin-1 and responsive to lipid trafficking defects. *Hum. Mol. Genet.* 2007, 16, 1495–1503. [CrossRef] [PubMed]

161. Burns, M.; Gaynor, K.; Olm, V.; Mercken, M.; LaFrancois, J.; Wang, L.; Mathews, P.M.; Noble, W.; Matsuoka, Y.; Duff, K. Presenilin redistribution associated with aberrant cholesterol transport enhances beta-amyloid production in vivo. *J. Neurosci.* 2003, 23, 5645–5649. [CrossRef]

162. Erickson, R.P.; Garver, W.S.; Camargo, F.; Hessain, G.S.; Heidenreich, R.A. Pharmacological and genetic modifications of somatic cholesterol do not substantially alter the course of CNS disease in Niemann-Pick C mice. *J. Inherit. Metab. Dis.* 2000, 23, 54–62. [CrossRef]

163. Erickson, R.P.; Bernard, O. Studies on neuronal death in the mouse model of Niemann-Pick C disease. *J. Neurosci. Res.* 2002, 68, 738–744. [CrossRef]

164. Zhang, M.; Hallows, J.L.; Wang, X.; Bu, B.; Wang, W.; Vincent, I. Mitogen-activated protein kinase activity inhibits autophagy in Niemann-Pick C disease. *Cell Tissue Res.* 2007, 328, 54–62. [CrossRef] [PubMed]

165. Somers, K.L.; Brown, D.E.; Fulton, R.; Schultheiss, P.C.; Hamar, D.; Smith, M.O.; Allison, R.; Connolly, H.E.; Just, C.; Mitchell, T.W.; et al. Effects of dietary cholesterol restriction in a feline model of Niemann-Pick type C disease. *J. Inherit. Metab. Dis.* 2001, 24, 427–436. [CrossRef] [PubMed]

166. Ahmad, I.; Hunter, R.E.; Flax, J.D.; Snyder, E.Y.; Erickson, R.P. Neural stem cell implantation extends life in Niemann-Pick C1 mice. *J. Appl. Genet.* 2007, 48, 269–272. [CrossRef] [PubMed]

167. Bae, J.S.; Han, H.S.; Youn, D.H.; Carter, J.E.; Modo, M.; Schuchman, E.H.; Jin, H.K. Bone marrow-derived mesenchymal stem cells promote neuronal networks with functional synaptic transmission after transplantation into mice with neurodegeneration. *Stem. Cells* 2007, 25, 1307–1316. [CrossRef]

168. Lee, H.; Bae, J.S.; Jin, H.K. Human umbilical cord blood-derived mesenchymal stem cells improve neurological abnormalities of Niemann-Pick-type C mouse by modulation of neuroinflammatory condition. *J. Vet. Med. Sci.* 2010, 72, 709–717. [CrossRef] [PubMed]

169. Bae, J.S.; Carter, J.E.; Jin, H.K. Adipose tissue-derived stem cells rescue Purkinje neurons and alleviate inflammatory responses in Niemann-Pick disease type C mice. *Cell Tissue Res.* 2010, 340, 357–369. [CrossRef] [PubMed]

170. Smith, D.; Wallom, K.L.; Williams, I.M.; Jeyakumar, M.; Platt, F.M. Beneficial effects of anti-inflammatory therapy in a mouse model of Niemann-Pick disease type C1. *Neurobiol. Dis.* 2009, 36, 242–251. [CrossRef]

171. Bascuñan-Castillo, E.C.; Erickson, R.P.; Howison, C.M.; Hunter, R.J.; Heidenreich, R.H.; Hicks, C.; Trouard, T.P.; Gilles, R.J. Tamoxifen and vitamin E treatments delay symptoms in the mouse model of Niemann-Pick C. *J. Appl. Genet.* 2004, 45, 461–467.

172. Marín, T.; Contreras, P.; Castro, J.F.; Chamorro, D.; Balboa, E.; Bosch-Morató, M.; Muñoz, F.J.; Alvarez, A.R.; Zanlungo, S. Vitamin E dietary supplementation improves neurological symptoms and decreases c-Abl/p73 activation in Niemann-Pick C mice. *Nutrients* 2014, 6, 3000–3017. [CrossRef]

173. Repa, J.J.; Li, H.; Frank-Cannon, T.C.; Valasek, M.A.; Turley, S.D.; Tansey, M.G.; Dietschy, J.M. Liver X receptor activation enhances cholesterol loss from the brain, decreases neuroinflammation, and increases survival of the NPC1 mouse. *J. Neurosci.* 2007, 27, 14470–14480. [CrossRef]

174. Langmade, S.J.; Gale, S.E.; Frolov, A.; Mohri, I.; Suzuki, K.; Mellon, S.H.; Walkley, S.U.; Covey, D.F.; Schaffer, J.E.; Ory, D.S. Pregnane X receptor (PXR) activation: A mechanism for neuroprotection in a mouse model of Niemann-Pick C disease. *Proc. Natl. Acad. Sci. USA* 2006, 103, 13807–13812. [CrossRef]

175. Chen, G.; Li, H.M.; Chen, Y.R.; Gu, X.S.; Duan, S. Decreased estradiol release from astrocytes contributes to the neurodegeneration in a mouse model of Niemann-Pick disease type C. *GLIA* 2007, 55, 1509–1518. [CrossRef] [PubMed]

176. Alvarez, A.R.; Klein, A.; Castro, J.; Cancino, G.I.; Amigo, J.; Mosquera, M.; Vargas, L.M.; Yévenes, L.F.; Bronfman, F.C.; Zanlungo, S. Imatinib therapy blocks cerebellar apoptosis and improves neurological symptoms in a mouse model of Niemann-Pick type C disease. *FASEB J.* 2008, 22, 3617–3627. [CrossRef]

177. Liu, B.; Li, H.; Repa, J.J.; Turley, S.D.; Dietschy, J.M. Genetic variations and treatments that affect the survival of the NPC1 mouse. *J. Lipid Res.* 2008, 49, 663–669. [CrossRef] [PubMed]

178. Liu, B.; Turley, S.D.; Burns, D.K.; Miller, A.M.; Repa, J.J.; Dietschy, J.M. Reversal of defective lysosomal transport in NPC disease ameliorates liver dysfunction and neurodegeneration in the npc1/- mouse. *Proc. Natl. Acad. Sci. USA* 2009, 106, 2377–2382. [CrossRef] [PubMed]
179. Davidson, C.D.; Ali, N.F.; Micsenyi, M.C.; Stephney, G.; Renault, S.; Dobrenis, K.; Ory, D.S.; Vanier, M.T.; Walkley, S.U. Chronic cyclodextrin treatment of murine Niemann-Pick C disease ameliorates neuronal cholesterol and glycosphingolipid storage and disease progression. *PloS ONE* **2009**, *4*, e6951. [CrossRef]

180. Aquil, A.; Liu, B.; Ramirez, C.M.; Pieper, A.A.; Estill, S.J.; Burns, D.K.; Liu, B.; Repa, J.J.; Turley, S.D.; Dietchy, J.M. Unesterified cholesterol accumulation in late endosomes/lysosomes causes neurodegeneration and is prevented by driving cholesterol export from this compartment. *J. Neurosci.* **2011**, *31*, 9404–9413. [CrossRef]

181. Vite, C.H.; Bagel, J.H.; Swain, G.P.; Prociuk, M.; Sikora, T.U.; Stein, V.M.; O’Donnell, P.; Ruane, T.; Ward, S.; Crooks, A.; et al. Intracisternal cyclodextrin prevents cerebellar dysfunction and Purkinje cell death in feline Niemann-Pick type C1 disease. *Sci. Transl. Med.* **2015**, *7*, 276ra226. [CrossRef]

182. Hao, Y.; Pan, D.; Zhang, M.; Xu, J.; Li, L.; Wei, J.; Wang, X. The neuroprotective effects of cyclin-dependent kinase-5 inhibition in mice with Niemann-Pick disease type C. *J. Huazhong Univ. Sci. Technolog. Med. Sci.* **2009**, *29*, 324–329. [CrossRef]

183. Borbon, I.A.; Hillman, Z.; Duran, E., Jr.; Kiela, P.R.; Frautschy, S.A.; Erickson, R.P. Lack of efficacy of curcumin on neurodegeneration in the mouse model of Niemann-Pick C1. *Pharmacol. Biochem. Behav.* **2012**, *101*, 125–131. [CrossRef]

184. Stein, V.M.; Crooks, A.; Ding, W.; Prociuk, M.; O’Donnell, P.; Bryan, C.; Sikora, T.; Dingemanse, J.; Vanier, M.T.; Walkley, S.U.; et al. Miglustat improves purkinje cell survival and alters microglial phenotype in feline niemann-pick disease type C. *J. Neuropathol. Exp. Neurol.* **2012**, *71*, 434–448. [CrossRef]

185. Nietupski, J.B.; Pacheco, J.J.; Chuang, W.L.; Maratea, K.; Li, L.; Foley, J.; Ashe, K.M.; Cooper, C.G.; Aerts, J.M.; Copeland, D.P.; et al. Inosugar-based inhibitors of glucosylceramide synthase prolong survival but paradoxically increase brain glucosylceramide levels in Niemann-Pick C mice. *Mol. Genet. Metab.* **2012**, *105*, 621–628. [CrossRef] [PubMed]

186. Fu, R.; Wassif, C.A.; Yanjani, N.M.; Watkins-Chow, D.E.; Baxter, L.L.; Incao, A.; Liscum, L.; Siddhu, R.; Firmkes, S.; Graham, M.; et al. Efficacy of N-acetylcysteine in phenotypic suppression of mouse models of Niemann-Pick disease, type C1. *Hum. Mol. Genet.* **2013**, *22*, 3508–3523. [CrossRef] [PubMed]

187. Argüello, G.; Martinez, P.; Peña, J.; Chen, O.; Platt, F.; Zanlungo, S.; González, M. Hepatic metabolic response to restricted copper intake in a Niemann-Pick C murine model. *Metallomics* **2014**, *6*, 1527–1539. [CrossRef] [PubMed]

188. Seo, Y.; Shin, Y.; Kim, H.S.; Kang, I.; Hong, I.S.; Choi, S.W.; Yu, K.R.; Kang, K.S. Donepezil enhances Purkinje cell survival and alleviates motor dysfunction by inhibiting cholesterol synthesis in a murine model of Niemann-Pick disease, type C1. *Mol. Genet. Metab.* **2012**, *105*, e2147. [CrossRef]

189. Williams, I.M.; Wallom, K.L.; Smith, D.A.; Al Eisa, N.; Smith, C.; Platt, F.M. Improved neuroprotection using miglustat, curcumin and ibuprofen as a triple combination therapy in Niemann-Pick disease type C1 mice. *Brain* **2013**, *136*, 9–17. [CrossRef]

190. Marques, A.R.; Aten, J.; Ottenhoff, R.; van Roomen, C.P.; Herrera Moro, D.; Claessen, N.; Vinueza Veloz, M.F.; Zhou, K.; Lin, Z.; Mirzaian, M.; et al. Reducing GBA2 Activity Ameliorates Neuropathology in Niemann-Pick Type C Mice. *Sci. Transl. Med.* **2016**, *8*, 355ra118. [CrossRef]

191. Cougnoux, A.; Clifford, S.; Salmon, A.; Ng, S.L.; Bertin, J.; Porter, F.D. Necroptosis inhibition as a therapy for Niemann-Pick disease, type C1: Inhibition of RIP kinases and combination therapy with 2-hydroxypropyl-β-cyclodextrin. *Mol. Genet. Metab.* **2018**, *125*, 345–350. [CrossRef]

192. Cougnoux, A.; Cluzeau, C.; Mitra, S.; Li, R.; Williams, I.; Burkert, K.; Xu, X.; Wassif, C.A.; Zheng, W.; Porter, F.D. Necroptosis in Niemann-Pick disease, type C1: A potential therapeutic target. *Cell Death Dis.* **2016**, *7*, e2147. [CrossRef]

193. Kirkegaard, T.; Gray, J.; Priestman, D.A.; Wallom, K.L.; Atkins, J.; Olsen, O.D.; Klein, A.; Drndarski, S.; Petersen, N.H.; Ingemann, L.; et al. Heat shock protein-based therapy as a potential candidate for treating the sphingolipidosis. *Sci. Transl. Med.* **2016**, *8*, 355ra118. [CrossRef]

194. Munkacsi, A.B.; Hammond, N.; Schneider, R.T.; Senanayake, D.S.; Haghi, K.; Lagutin, K.; Bloor, S.J.; Ory, D.S.; Maue, R.A.; Chen, F.W.; et al. Normalization of Hepatic Homeostasis in the Npc1(nmf164) Mouse Model of Niemann-Pick Type C Disease Treated with the Histone Deacetylase Inhibitor Vorinostat. *J. Biol Chem.* **2017**, *292*, 4395–4410. [CrossRef]

195. Xie, C.; Gong, X.M.; Luo, J.; Li, B.L.; Song, B.L. AAV9-NPC1 significantly ameliorates Purkinje cell death and behavioral abnormalities in mouse NPC disease. *J. Lipid Res.* **2017**, *58*, 512–518. [CrossRef] [PubMed]
196. Chandler, R.J.; Williams, I.M.; Gibson, A.L.; Davidson, C.D.; Incao, A.A.; Hubbard, B.T.; Porter, F.D.; Pavan, W.J.; Venditti, C.P. Systemic AAV9 gene therapy improves the lifespan of mice with Niemann-Pick disease, type C1. Hum. Mol. Genet. 2017, 26, 52–64. [CrossRef] [PubMed]

197. Hughes, M.P.; Smith, D.A.; Morris, L.; Fletcher, C.; Colaco, A.; Huebecker, M.; Tordo, J.; Palomar, N.; Massaro, G.; Henckaerts, E.; et al. AAV9 intracerebroventricular gene therapy improves lifespan, locomotor function and pathology in a mouse model of Niemann-Pick type C1 disease. Hum. Mol. Genet. 2018, 27, 3079–3098. [CrossRef] [PubMed]

198. Markmann, S.; J, J.C.-R.; Rosenberg, J.B.; De, B.P.; Kaminsky, S.M.; Crystal, R.G.; Sondhi, D. Attenuation of the Niemann-Pick type C2 disease phenotype by intracisternal administration of an AAVrh.10 vector expressing Npc2. Exp. Neurol. 2018, 306, 22–33. [CrossRef] [PubMed]

199. Torres, S.; Matias, N.; Baulies, L.; Nuñez, S.; Alarcon-Vila, C.; Martinez, L.; Nuñez, N.; Fernandez, A.; Caballera, J.; Levade, T.; et al. Mitochondrial GSH replenishment as a potential therapeutic approach for Niemann Pick type C disease. Redox Biol. 2017, 11, 60–72. [CrossRef] [PubMed]

200. Ferrante, A.; Pezzola, A.; Matteucci, A.; Di Biase, A.; Attorri, L.; Armida, M.; Martire, A.; Chern, Y.; Popoli, P. The adenosine A(2A) receptor agonist T1-11 ameliorates neurovisceral symptoms and extends the lifespan of a mouse model of Niemann-Pick type C disease. Neurobiol. Dis. 2018, 110, 1–11. [CrossRef]

201. Kulkarni, A.; Caporali, P.; Dolas, A.; Johny, S.; Goyal, S.; Dragotto, J.; Macone, A.; Jayaraman, R.; Fiorenza, M.T. Linear Cyclodextrin Polymer Prodrugs as Novel Therapeutics for Niemann-Pick Type C1 Disorder. Sci. Rep. 2018, 8, 9547. [CrossRef]

202. Houben, T.; Magro Dos Reis, I.; Oligschlaeger, Y.; Steinbusch, H.; Gijbels, M.J.J.; Hendrikx, T.; Binder, C.J.; Cassiman, D.; Westerterp, M.; Prickaerts, J.; et al. Pneumococcal Immunization Reduces Neurological and Hepatic Symptoms in a Mouse Model for Niemann-Pick Type C1 Disease. Front. Immunol. 2018, 9, 3089. [CrossRef]

203. Moreau, D.; Vacc, F.; Vossio, S.; Scott, C.; Colaco, A.; Paz Montoya, J.; Ferguson, C.; Damme, M.; Moniatte, M.; Parton, R.G.; et al. Drug-induced increase in lysobisphosphatidic acid reduces the cholesterol overload in Niemann-Pick type C cells and mice. EMBO Rep. 2019, 20, e47055. [CrossRef]

204. Yasmin, N.; Ishitsuka, Y.; Fukaura, M.; Yamada, Y.; Nakahara, S.; Ishii, A.; Kondo, Y.; Takeo, T.; Nakagata, N.; Motoyama, K.; et al. In Vitro and In Vivo Evaluation of 6-O-α-Maltosyl-β-Cyclodextrin as a Potential Therapeutic Agent Against Niemann-Pick Disease Type C. Int. J. Mol. Sci. 2019, 20, 1152. [CrossRef]

205. Park, M.H.; Choi, B.J.; Jeong, M.S.; Lee, J.Y.; Jung, I.K.; Park, K.H.; Lee, H.W.; Yamaguchi, T.; Marti, H.H.; Lee, B.H.; et al. Characterization of the Subventricular-Thalamo-Cortical Circuit in the NP-C Mouse Brain, and New Insights Regarding Treatment. Mol. Ther. 2019, 27, 1507–1526. [CrossRef] [PubMed]

206. Mitroi, D.N.; Pereyra-Gómez, G.; Soto-Huelin, B.; Senovilla, F.; Kobayashi, T.; Esteban, J.A.; Ledesma, M.D. NPC1 enables cholesterol mobilization during long-term potentiation that can be restored in Niemann-Pick disease type C by CYP46A1 activation. EMBO Rep. 2019, 20, e48143. [CrossRef] [PubMed]

207. Schultz, M.L.; Fawaz, M.V.; Azaria, R.D.; Hollon, T.C.; Liu, E.A.; Kunkel, T.J.; Halseth, T.A.; Krus, K.L.; Ming, R.; Morin, E.E.; et al. Synthetic high-density lipoprotein nanoparticles for the treatment of Niemann-Pick diseases. BMC Med. 2019, 17, 200. [CrossRef]

208. Levy, J.M.; Yeh, W.H.; Pendse, N.; Davis, J.R.; Hennessy, E.; Butcher, R.; Koblan, L.W.; Comander, J.; Liu, Q.; Liu, D.R. Cytosine and adenine base editing of the brain, liver, retina, heart and skeletal muscle of mice via adeno-associated viruses. Nat. Biomed. Eng. 2020, 4, 97–110. [CrossRef]

209. Hung, Y.H.; Lotan, A.; Yeshurun, S.; Schroeder, A.; Bush, A.I. Iron chelation by deferiprone does not rescue the Niemann-Pick Disease Type C1 mouse model. Biomaterials 2020, 33, 87–95. [CrossRef]

210. Jiang, D.; Lee, H.; Partridge, W.M. Plasmid DNA gene therapy of the Niemann-Pick C1 mouse with transferrin receptor-targeted Trojan horse liposomes. Sci. Rep. 2020, 10, 13334. [CrossRef] [PubMed]

211. Millat, G.; Marçais, C.; Rafi, M.A.; Yamamoto, T.; Morris, J.A.; Petchev, P.G.; Ohno, K.; Wengier, D.A.; Vanier, M.T. Niemann-Pick C1 disease: The I1061T substitution is a frequent mutant allele in patients of Western European descent and correlates with a classic juvenile phenotype. Am. J. Hum. Genet. 1999, 65, 1321–1329. [CrossRef] [PubMed]

212. Park, W.D.; O’Brien, J.F.; Lundquist, P.A.; Kraft, D.L.; Vockley, C.W.; Karnes, P.S.; Patterson, M.C.; Snow, K. Identification of 58 novel mutations in Niemann-Pick disease type C: Correlation with biochemical phenotype and importance of PTC1-like domains in NPC1. Hum. Mutat. 2003, 22, 313–325. [CrossRef]
213. Millat, G.; Marçais, C.; Tomasetto, C.; Chikh, K.; Fensom, A.H.; Harzer, K.; Wenger, D.A.; Ohno, K.; Vanier, M.T. Niemann-Pick C1 disease: Correlations between NPC1 mutations, levels of NPC1 protein, and phenotypes emphasize the functional significance of the putative sterol-sensing domain and of the cysteine-rich luminal loop. *Am. J. Hum. Genet.* 2001, 68, 1373–1385. [CrossRef]

214. Benussi, A.; Alberici, A.; Premi, E.; Bertasi, V.; Cotelli, M.S.; Turla, M.; Dardis, A.; Zampieri, S.; Marchina, E.; Paghera, B.; et al. Phenotypic heterogeneity of Niemann-Pick disease type C in monozygotic twins. *J. Neurol. 2015*, 262, 642–647. [CrossRef]

215. Miyawaki, S.; Yoshida, H.; Mitsuoka, S.; Enomoto, H.; Ikehara, S. A mouse model for Niemann-Pick disease. Influence of genetic background on disease expression in splen/spm mice. *J. Hered 1986*, 77, 379–384. [CrossRef]

216. Zhang, J.; Erickson, R.P. A modifier of Niemann Pick C1 maps to mouse chromosome 19. *Mamm. Genome 2000*, 11, 69–71. [CrossRef] [PubMed]

217. Parra, J.; Klein, A.D.; Castro, J.; Morales, M.G.; Mosqueira, M.; Valencia, I.; Cortés, V.; Rigotti, A.; Zanlungo, S. Npc1 deficiency in the C57BL/6J genetic background enhances Niemann-Pick disease type C spleen pathology. *Biochem. Biophys. Res. Commun. 2011*, 413, 400–406. [CrossRef] [PubMed]

218. Marshall, C.A.; Watkins-Chow, D.E.; Palladino, G.; Deutsch, G.; Chandran, K.; Pavan, W.J.; Erickson, R.P. In Niemann-Pick C1 mouse models, glial-only expression of the normal gene extends survival much further than do changes in genetic background or treatment with hydroxypropyl-beta-cyclodextrin. *Gene 2018*, 643, 117–123. [CrossRef] [PubMed]

219. Xie, C.; Turley, S.D.; Pentchev, P.G.; Dietschy, J.M. Cholesterol balance and metabolism in mice with loss of function of Niemann-Pick C protein. *Am. J. Physiol 1999*, 276, E336–E344. [CrossRef] [PubMed]

220. Xie, C.; Lund, E.G.; Turley, S.D.; Russell, D.W.; Dietschy, J.M. Quantitation of two pathways for cholesterol excretion from the brain in normal mice and mice with neurodegeneration. *J. Lipid Res. 2003*, 44, 1780–1789. [CrossRef]

221. Li, H.; Turley, S.D.; Liu, B.; Repa, J.J.; Dietschy, J.M. GM2/GD2 and GM3 gangliosides have no effect on cellular cholesterol pools or turnover in normal or NPC1 mice. *J. Lipid Res. 2008*, 49, 1816–1828. [CrossRef]

222. Kaptzan, T.; West, S.A.; Holicky, E.L.; Wheatley, C.L.; Marks, D.L.; Wang, T.; Peake, K.B.; Vance, J.; Walkley, S.U.; Pagano, R.E. Development of a Rab9 transgenic mouse and its ability to increase the lifespan of a murine model of Niemann-Pick type C disease. *Am. J. Pathol. 2009*, 174, 14–20. [CrossRef]

223. Maulik, M.; Ghoshal, B.; Kim, J.; Wang, Y.; Yang, J.; Westaway, D.; Kar, S. Mutant human APP exacerbates early inflammation in Niemann-Pick disease Type C. *J. Neuroinflamm. 2019*, 16, 269. [CrossRef] [PubMed]

224. Kaptzan, T.; West, S.A.; Holicky, E.L.; Wheatley, C.L.; Marks, D.L.; Wang, T.; Peake, K.B.; Vance, J.; Walkley, S.U.; Pagano, R.E. Development of a Rab9 transgenic mouse and its ability to increase the lifespan of a murine model of Niemann-Pick type C disease. *Am. J. Pathol. 2009*, 174, 14–20. [CrossRef]

225. Maulik, M.; Thinakaran, G.; Kar, S. Alterations in gene expression in mutant amyloid precursor protein exacerbates early inflammation in Niemann-Pick disease Type C. *Am. J. Physiol. Liver Physiol. 2018*, 315, G454–G463. [CrossRef] [PubMed]

226. Lee, H.; Lee, J.K.; Bae, Y.C.; Yang, S.H.; Okino, N.; Schuchman, E.H.; Yamashita, T.; Bae, J.S.; Jin, H.K. Inhibition of GM3 synthase attenuates neuropathology of Niemann-Pick disease Type C by affecting sphingolipid metabolism. *Mol. Cells 2014*, 37, 161–171. [CrossRef]

227. Chung, C.; Elrick, M.J.; Dell’Orco, J.M.; Qin, Z.S.; Kalyana-Sundaram, S.; Chinnaiyen, A.M.; Shakkottai, V.G.; Lieberman, A.P. Heat Shock Protein Beta-1 Modifies Anterior to Posterior Purkinje Cell Vulnerability in a Mouse Model of Niemann-Pick Type C Disease. *PLoS Genet. 2016*, 12, e1006042. [CrossRef] [PubMed]

228. Lopez, A.M.; Jones, R.D.; Repa, J.J.; Turley, S.D. Niemann-Pick C1-deficient mice lacking sterol O-acyltransferase 2 have less hepatic cholesterol entrapment and improved liver function. *Am. J. Physiol. Liver Physiol. 2018*, 315, G454–G463. [CrossRef] [PubMed]

229. Shin, S.D.; Shin, A.; Mayagotia, K.; Siebold, L.; Rubini, M.; Wilson, C.G.; Bellinger, D.L.; Soriano, S. Loss of amyloid precursor protein exacerbates early inflammation in Niemann-Pick disease type C. *J. Neuroinflamm. 2019*, 16, 269. [CrossRef] [PubMed]

230. Cawley, N.X.; Lyons, A.T.; Abebe, D.; Wassif, C.A.; Porter, F.D. Evaluation of the Potential Role of Proprotein Convertase Subtilisin/Kexin Type 9 (PCSK9) in Niemann–Pick Disease, Type C1. *Int. J. Mol. Sci. 2020*, 21, 2430. [CrossRef]
231. Cougnoux, A.; Yerger, J.C.; Fellmeth, M.; Serra-Vinardell, J.; Martin, K.; Navid, F.; Iben, J.R.; Wassif, C.A.; Cawley, N.X.; Porter, F.D. Single Cell Transcriptome Analysis of Niemann–Pick Disease, Type C1 Cerebella. *Int. J. Mol. Sci.* 2020, 21, 5368. [CrossRef]

232. Klein, A.D.; De La Vega, J.G.; Zanlungo, S. Complement Component C3 Participates in Early Stages of Niemann–Pick C Mouse Liver Damage. *Int. J. Mol. Sci.* 2020, 21, 2127. [CrossRef]

233. Liu, E.A.; Schultz, M.L.; Mochida, C.; Chung, C.; Paulson, H.L.; Lieberman, A.P. Fbxo2 mediates clearance of damaged lysosomes and modifies neurodegeneration in the Niemann-Pick C brain. *JCI Insight* 2020, 5. [CrossRef]

234. Ramirez, C.M.; Taylor, A.M.; Lopez, A.M.; Repa, J.J.; Turley, S.D. Delineation of metabolic responses of Npc1(-/-nih) mice lacking the cholesterol-esterifying enzyme SOAT2 to acute treatment with 2-hydroxypropyl-β-cyclodextrin. *Steroids* 2020, 164, 108725. [CrossRef]

235. Smith, A.F.; Vanderah, T.W.; Erickson, R.P. Haploinsufficiency of tau decreases survival of the mouse model of Niemann–Pick disease type C1 but does not alter tau phosphorylation. *J. Appl. Genet.* 2020, 61, 567–570. [CrossRef] [PubMed]

236. Vöikar, V.; Rauvala, H.; Ikonen, E. Cognitive deficit and development of motor impairment in a mouse model of Niemann-Pick type C disease. *Behav. Brain Res.* 2002, 132, 1–10. [CrossRef] [PubMed]

237. Cougnoux, A.; Fellmeth, M.; Gu, T.; Davidson, C.D.; Gibson, A.L.; Pavan, W.J.; Porter, F.D. Maternal immune activation modifies the course of Niemann-pick disease in mice in a gender specific manner. *Mol. Genet. Metab.* 2020, 129, 165–170. [CrossRef] [PubMed]

238. Houben, T.; Bitorina, A.V.; Oligschläger, Y.; Jeurissen, M.L.; Rensen, S.; Köhler, S.E.; Westerterp, M.; Lütjohann, D.; Theys, J.; Romano, A.; et al. Sex-opposed inflammatory effects of 27-hydroxycholesterol are mediated via differences in estrogen signaling. *J. Pathol.* 2020, 251, 429–439. [CrossRef] [PubMed]

239. Tchernof, A.; Després, J.P. Pathophysiology of human visceral obesity: An update. *Physiol. Rev.* 2013, 93, 359–404. [CrossRef] [PubMed]

240. Regitz-Zagrosek, V.; Kararigas, G. Mechanistic Pathways of Sex Differences in Cardiovascular Disease. *Physiol. Rev.* 2017, 97, 1–37. [CrossRef] [PubMed]

241. Lonardo, A.; Nascimbeni, F.; Ballestri, S.; Fairweather, D.; Win, S.; Than, T.A.; Abdelmalek, M.F.; Suzuki, A. Sex Differences in Nonalcoholic Fatty Liver Disease: State of the Art and Identification of Research Gaps. *Hepatology* 2019, 70, 1457–1469. [CrossRef]

242. Mauvais-Jarvis, F.; Bairey Merz, N.; Barnes, P.J.; Brinton, R.D.; Carrero, J.J.; DeMeo, D.L.; De Vries, G.J.; Epperson, C.N.; Govindan, R.; Klein, S.L.; et al. Sex and gender: Modifiers of health, disease, and medicine. *Lancet* 2020, 396, 565–582. [CrossRef]

243. Segatto, M.; Di Giovanni, A.; Marino, M.; Pallottini, V. Analysis of the protein network of cholesterol homeostasis in different brain regions: An age and sex dependent perspective. *J. Cell Physiol.* 2013, 228, 1561–1567. [CrossRef] [PubMed]

244. Tonini, C.; Segatto, M.; Pallottini, V. Impact of Sex and Age on the Mevalonate Pathway in the Brain: A Focus on Effects Induced by Maternal Exposure to Exogenous Compounds. *Metab.* 2020, 10, 304. [CrossRef]

245. Yu, T.; Shakkottai, V.G.; Chung, C.; Lieberman, A.P. Temporal and cell-specific deletion establishes that neuronal Npc1 deficiency is sufficient to mediate neurodegeneration. *Hum. Mol. Genet.* 2011, 20, 4440–4451. [CrossRef] [PubMed]

246. Yu, T.; Lieberman, A.P. Npc1 acting in neurons and glia is essential for the formation and maintenance of CNS myelin. *PLoS Genet.* 2013, 9, e1003462. [CrossRef] [PubMed]

247. Borbon, I.; Totenhagen, J.; Fiorenza, M.T.; Canterini, S.; Ke, W.; Trouard, T.; Erickson, R.P. Niemann-Pick C1 mice, a model of “juvenile Alzheimer’s disease”, with normal gene expression in neurons and fibrillary astrocytes show long term survival and delayed neurodegeneration. *J. Alzheimers Dis* 2012, 30, 875–887. [CrossRef] [PubMed]

248. Klein, A.D.; Oyarzúñ, J.E.; Cortez, C.; Zanlungo, S. Gadolinium Chloride Rescues Niemann–Pick Type C Liver Damage. *Int. J. Mol. Sci.* 2018, 19, 3599. [CrossRef]

249. Slezak, M.; Goritz, C.; Niemiec, A.; Frisen, J.; Chambon, P.; Metzger, D.; Pfrieger, F.W. Transgenic mice for conditional gene manipulation in astroglial cells. *GLIA* 2007, 55, 1565–1576. [CrossRef]

250. Pfrieger, F.W.; Slezak, M. Genetic approaches to study glial cells in the rodent brain. *GLIA* 2012, 60, 681–701. [CrossRef]
251. German, D.C.; Liang, C.L.; Song, T.; Yazdani, U.; Xie, C.; Dietschy, J.M. Neurodegeneration in the Niemann-Pick C mouse: Glial involvement. *Neuroscience* **2002**, *109*, 437–450. [CrossRef]

252. Baudry, M.; Yao, Y.; Simonsen, D.; Liu, J.; Bi, X. Postnatal development of inflammation in a murine model of Niemann-Pick type C disease: Immunohistochemical observations of microglia and astrogliosis. *Exp. Neurol.* **2003**, *184*, 887–903. [CrossRef]

253. Kavetsky, L.; Green, K.K.; Boyle, B.R.; Youssufzai, F.A.K.; Padron, Z.M.; Melli, S.E.; Kuhnle, V.L.; Jackson, H.M.; Blanco, R.E.; Howell, G.R.; et al. Increased interactions and engulfment of dendrites by microglia precede Purkinje cell degeneration in a mouse model of Niemann Pick Type C. *Sci. Rep.* **2019**, *9*, 14722. [CrossRef]

254. Walkley, S.U. Pyramidal neurons with ectopic dendrites in storage diseases exhibit increased GM2 ganglioside immunoreactivity. *Neuroscience* **1995**, *68*, 1027–1035. [CrossRef]

255. Mauler, D.A.; Gandolfi, B.; Reinero, C.R.; O’Brien, D.P.; Spooner, J.L.; Lyons, L.A. Precision Medicine in Neurodegeneration in the feline Niemann-Pick disease type C. *Acta Neuropathol.* **1997**, *94*, 164–172. [CrossRef] [PubMed]

256. Vite, C.H.; Ding, W.; Bryan, C.; O’Donnell, P.; Cullen, K.; Aleman, D.; Haskins, M.E.; Winkle, T.V. Clinical, Electrophysiological, and Serum Biochemical Measures of Progressive Neurological and Hepatic Dysfunction in Feline Niemann-Pick Type C Disease. *Pediatric Res.* **2008**, *64*, 544–549. [CrossRef] [PubMed]

257. Bagel, J.H.; Sikora, T.U.; Prociuk, M.; Pesayco, J.P.; Miszisin, A.P.; Shelton, G.D.; Vite, C.H. Electrodiagnostic testing and histopathologic changes confirm peripheral nervous system myelin abnormalities in the feline model of niemann-pick disease type C. *J. Neuropathol. Exp. Neurol.* **2013**, *72*, 256–262. [CrossRef]

258. Roszell, B.R.; Tao, J.-Q.; Yu, K.J.; Gao, L.; Huang, S.; Ning, Y.; Feinstein, S.I.; Vite, C.H.; Bates, S.R. Pulmonary Abnormalities in Animal Models Due to Niemann-Pick Type C1 (NPC1) or C2 (NPC2) Disease. *PLoS ONE* **2013**, *8*, e67084. [CrossRef] [PubMed]

259. Maurer, D.A.; Gandolfi, B.; Reinero, C.R.; O’Brien, D.P.; Spooner, J.L.; Lyons, L.A. Precision Medicine in Cats: Novel Niemann-Pick Type C1 Diagnosed by Whole-Genome Sequencing. *J. Vet. Intern. Med.* **2017**, *31*, 539–544. [CrossRef]

260. Zampieri, S.; Bianchi, E.; Cantile, C.; Saleri, R.; Bembi, B.; Dardis, A. Characterization of a Spontaneous Novel Mutation in the NPC2 Gene in a Cat Affected by Niemann Pick Type C Disease. *PLoS ONE* **2014**, *9*, e112503. [CrossRef]

261. Sloan, H.R.; Uhlendorf, B.W.; Kanfer, J.N.; Brady, R.O.; Fredrickson, D.S. Deficiency of sphingomyelin-cleaving enzyme activity in tissue cultures derived from patients with Niemann-Pick disease. *Biochem. Biophys. Res. Commun.* **1969**, *34*, 582–588. [CrossRef]

262. Pentechev, P.G.; Booth, A.D.; Kruth, H.S.; Weintroub, H.; Stivers, J.; Brady, R.O. A genetic storage disorder in BALB/C mice with a metabolic block in esterification of exogenous cholesterol. *J. Biol. Chem.* **1984**, *259*, 5784–5791. [PubMed]

263. Liscum, L.; Faust, J.R. Low density lipoprotein (LDL)-mediated suppression of cholesterol synthesis and LDL uptake is defective in Niemann-Pick type C fibroblasts. *J. Biol. Chem.* **1987**, *262*, 17002–17008.

264. Maziere, C.; Maziere, J.C.; Mora, L.; Lageron, A.; Polonovski, C.; Polonovski, J. Alterations in cholesterol metabolism in cultured fibroblasts from patients with Niemann-Pick disease type C. *J. Inherit. Metab. Dis.* **1997**, *20*, 339–346. [CrossRef]

265. Carstea, E.D.; Morris, J.A.; Coleman, K.G.; Loftus, S.K.; Zhang, D.; Cummings, C.; Wu, J.; Rosenfeld, M.A.; Pavan, W.J.; Krizman, D.B.; et al. Niemann-Pick C1 Disease Gene: Homology to Mediators of Cholesterol Homeostasis. *Science* **1997**, *277*, 228–231. [CrossRef] [PubMed]

266. Gelsthorpe, M.E.; Baumann, N.; Millard, E.; Gale, S.E.; Langmade, S.J.; Schaffer, J.E.; Ory, D.S. Niemann-Pick type C1 I1061T mutant encodes a functional protein that is selected for endoplasmic reticulum-associated degradation due to protein misfolding. *J. Biol. Chem.* **2008**, *283*, 8229–8236. [CrossRef] [PubMed]

267. Liscum, L.; Faust, J.R. The intracellular transport of low density lipoprotein-derived cholesterol is inhibited in Chinese hamster ovary cells cultured with 3-beta-[2-(diethylamino)ethoxy]androst-5-en-17-one. *J. Biol. Chem.* **1987**, *262*, 17002–17008.

268. Rodriguez-Lafrasse, C.; Rousson, R.; Bonnet, J.; Pentechev, P.G.; Louisot, P.; Vanier, M.T. Abnormal cholesterol metabolism in imipramine-treated fibroblast cultures. Similarities with Niemann-Pick type C disease. *Biochim. Biophys. Acta* **1990**, *1043*, 123–128. [CrossRef]
269. Roff, C.F.; Goldin, E.; Comly, M.E.; Cooney, A.; Brown, A.; Vanier, M.T.; Miller, S.P.; Brady, R.O.; Pentchev, P.G. Type C Niemann-Pick disease: Use of hydrophobic amines to study defective cholesterol transport. *Dev. Neurosci.* 1991, 13, 315–319. [CrossRef] [PubMed]

270. Koh, C.H.; Cheung, N.S. Cellular mechanism of U18666A-mediated apoptosis in cultured murine cortical neurons: Bridging Niemann-Pick disease type C and Alzheimer’s disease. *Cell Signal.* 2006, 18, 1844–1853. [CrossRef]

271. Cenedella, R.J. Cholesterol synthesis inhibitor U18666A and the role of sterol metabolism and trafficking in numerous pathophysiological processes. *Lipids* 2009, 44, 477–487. [CrossRef]

272. Lu, F.; Liang, Q.; Abi-Mosleh, L.; Das, A.; De Brabander, J.K.; Goldstein, J.L.; Brown, M.S. Identification of NPC1 as the target of U18666A, an inhibitor of lysosomal cholesterol export and Ebola infection. *eLife* 2015, 4, e12177. [CrossRef]

273. Higaki, K.; Ninomiya, H.; Sugimoto, Y.; Suzuki, T.; Taniguchi, M.; Niwa, H.; Pentchev, P.G.; Vanier, M.T.; Schnaar, R.L. Sphingomyelinase and nonspecific phosphodiesterase activities in Epstein-Barr virus-transformed lymphoid cell lines from Niemann-Pick disease A, B and C. *Biochim. Biophys. Acta* 1994, 1226, 173–180. [CrossRef]

274. Dahl, N.K.; Reed, K.L.; Daunais, M.A.; Faust, J.R.; Liscum, L. Isolation and characterization of Chinese hamster ovary cell mutants defective in the intracellular metabolism of low density lipoprotein-derived cholesterol. *J. Biol. Chem.* 1992, 267, 4899–4906. [CrossRef]

275. Brown, A.; Patel, S.; Ward, C.; Lorenz, A.; Ortiz, M.; DuRos, A.; Wieghardt, F.; Esch, A.; Otten, E.G.; Heiser, L.M.; et al. PEG-lipid micelles enable cholesterol efflux in Niemann-Pick Type C1 disease-based lysosomal storage disorder. *Sci. Rep.* 2016, 6, 31750. [CrossRef] [PubMed]

276. Huang, L.; Pike, D.; Sleat, D.E.; Nanda, V.; Lobel, P. Potential pitfalls and solutions for use of fluorescent fusion proteins to study the lysosome. *PLoS ONE* 2014, 9, e88893. [CrossRef] [PubMed]

277. Coleman, E.M.; Walker, T.N.; Hildreth, J.E. Loss of Niemann Pick type C proteins 1 and 2 greatly enhances HIV infectivity and is associated with accumulation of HIV Gag and cholesterol in late endosomes/lysosomes. *Virol. J.* 2012, 9, 31. [CrossRef] [PubMed]

278. Cadigan, K.M.; Spillane, D.M.; Chang, T.Y. Isolation and characterization of Chinese hamster ovary cell mutants defective in intracellular low density lipoprotein-cholesterol trafficking. *J. Cell Biol.* 1990, 110, 295–308. [CrossRef]

279. Dahl, N.K.; Reed, K.L.; Daunais, M.A.; Faust, J.R.; Liscum, L. Isolation and characterization of Chinese hamster ovary cells defective in the intracellular metabolism of low density lipoprotein-derived cholesterol. *J. Biol. Chem.* 1992, 267, 4899–4906. [CrossRef]

280. Koh, C.H.; Cheung, N.S. Cellular mechanism of U18666A-mediated apoptosis in cultured murine cortical neurons: Bridging Niemann-Pick disease type C and Alzheimer’s disease. *Cell Signal.* 2006, 18, 1844–1853. [CrossRef]

281. Lu, F.; Liang, Q.; Abi-Mosleh, L.; Das, A.; De Brabander, J.K.; Goldstein, J.L.; Brown, M.S. Identification of NPC1 as the target of U18666A, an inhibitor of lysosomal cholesterol export and Ebola infection. *eLife* 2015, 4, e12177. [CrossRef]

282. Ebrahimi, V.; Hashemi, A. Challenges of in vitro genome editing with CRISPR/Cas9 and possible solutions: A review. *Gene* 2020, 753, 144813. [CrossRef]
289. Bartz, F.; Kern, L.; Erz, D.; Zhu, M.; Gilbert, D.; Meinhof, T.; Wirkner, U.; Erfle, H.; Muckenthaler, M.; Pepperkok, R.; et al. Identification of cholesterol-regulating genes by targeted RNAi screening. Cell Metab. 2009, 10, 63–75. [CrossRef]

290. Du, X.; Lukmantara, I.; Yang, H. CRISPR/Cas9-Mediated Generation of Niemann-Pick C1 Knockout Cell Line. Methods Mol. Biol. 2017, 1583, 73–83.

291. Tharkeshwar, A.K.; Trekker, J.; Vermeire, W.; Pauwels, J.; Sannerud, R.; Priestman, D.A.; Te Vruchte, D.; Vints, K.; Baatsen, P.; Decuyper, J.P.; et al. A novel approach to analyze lysosomal dysfunctions through subcellular proteomics and lipidomics: The case of NPC1 deficiency. Sci. Rep. 2017, 7, 41408. [CrossRef]

292. Zhao, K.; Ridgway, N.D. Oxysterol-Binding Protein-Related Protein 1L Regulates Cholesterol Egress from the Endo-Lysosomal System. Cell Rep. 2017, 19, 1807–1818. [CrossRef]

293. Castellano, B.M.; Thelen, A.M.; Moldavski, O.; Feltes, M.; van der Welle, R.E.; Mydock-McGrane, L.; Jiang, X.; van Eijkeren, R.J.; Davis, O.B.; Louie, S.M.; et al. Lysosomal cholesterol activates mTORC1 via an SLC38A9-Niemann-Pick C1 signaling complex. Science 2017, 355, 1306–1311. [CrossRef]

294. Rodriguez-Pascau, L.; Coll, M.J.; Casas, J.; Vilageliu, L.; Grinberg, D. Generation of a human neuronal stable cell model for niemann-pick C disease by RNA interference. JIMD Rep. 2012, 4, 29–37.

295. Ulatowski, L.; Parker, R.; Davidson, C.; Yanjanin, N.; Kelley, T.J.; Corey, D.; Atkinson, J.; Porter, F.; Arai, H.; Walkley, S.U.; et al. Altered vitamin E status in Niemann-Pick type C disease. J. Lipid Res. 2011, 52, 1400–1410. [CrossRef] [PubMed]

296. Twu, Y.C.; Lee, T.S.; Lin, Y.L.; Hsu, S.M.; Wang, Y.H.; Liao, C.Y.; Wang, C.K.; Liang, Y.C.; Liao, Y.J. Niemann-Pick Type C2 Protein Mediates Hepatic Stellate Cells Activation by Regulating Free Cholesterol Accumulation. Int. J. Mol. Sci. 2016, 17. [CrossRef] [PubMed]

297. Davies, J.P.; Chen, F.W.; Ioannou, Y.A. Transmembrane molecular pump activity of Niemann-Pick C1 protein. Science 2000, 290, 2295–2298. [CrossRef] [PubMed]

298. Maulik, M.; Peake, K.; Chung, J.; Wang, Y.; Vance, J.E.; Kar, S. APP overexpression in the absence of NPC1 exacerbates metabolism of amyloidogenic proteins of Alzheimer’s disease. Hum. Mol. Genet. 2015, 24, 7132–7150. [CrossRef]

299. Watabe, K.; Ida, H.; Uehara, K.; Oyanagi, K.; Sakamoto, T.; Tanaka, J.; Garver, W.S.; Miyawaki, S.; Ohno, K.; Eto, Y. Establishment and characterization of immortalized Schwann cells from murine model of Niemann-Pick disease type C (sph/sph). J. Peripher. Nerv. Syst. 2001, 6, 85–94. [CrossRef]

300. Erwood, S.; Brewer, R.A.; Bily, T.M.I.; Maino, E.; Zhou, L.; Cohn, R.D.; Ivakine, E.A. Modeling Niemann-Pick disease type C in a human haploid cell line allows for patient variant characterization and clinical interpretation. Genome Res. 2019, 29, 2010–2019. [CrossRef]

301. Wang, M.L.; Motamed, M.; Infante, R.E.; Abi-Mosleh, L.; Kwon, H.J.; Brown, M.S.; Goldstein, J.L. Identification of surface residues on Niemann-Pick C2 essential for hydrophobic handoff of cholesterol to NPC1 in lysosomes. Cell Metab. 2010, 12, 166–173. [CrossRef]

302. Vanharanta, L.; Peränen, J.; Pfisterer, S.G.; Enkavi, G.; Vattulainen, I.; Ikonen, E. High-content imaging and structure-based predictions reveal functional differences between Niemann-Pick C1 variants. Traffic 2020, 21, 386–397. [CrossRef]

303. Carette, J.E.; Raaben, M.; Wong, A.C.; Herbert, A.S.; Obermoster, G.; Mulherkar, N.; Kuehne, A.I.; Kranzusch, P.J.; Griffin, A.M.; Ruthel, G.; et al. Ebola virus entry requires the cholesterol transporter Niemann-Pick C1. Nature 2011, 477, 340–343. [CrossRef]

304. Côté, M.; Misasi, J.; Ren, T.; Bruchez, A.; Lee, K.; Filone, C.M.; Hensley, L.; Li, Q.; Ory, D.; Chandran, K.; et al. Small molecule inhibitors reveal Niemann-Pick C1 is essential for Ebola virus infection. Nature 2011, 477, 344–348. [CrossRef]

305. Stoeck, I.K.; Lee, J.Y.; Tabata, K.; Romero-Brey, I.; Paul, D.; Schult, P.; Lohmann, V.; Kaderali, L.; Bartenschlager, R. Hepatitis C Virus Replication Depends on Endosomal Cholesterol Homeostasis. J. Virol. 2018, 92, e01196-17. [CrossRef] [PubMed]

306. Cahoy, J.D.; Emery, B.; Kaushal, A.; Foo, L.C.; Zamanian, J.L.; Christopherson, K.S.; Xing, Y.; Lubischer, J.L.; Krieg, P.A.; Krupenko, S.A.; et al. A transcriptome database for astrocytes, neurons, and oligodendrocytes: A new resource for understanding brain development and function. J. Neurosci. 2008, 28, 264–278. [CrossRef] [PubMed]
307. Foo, L.C.; Allen, N.J.; Bushong, E.A.; Ventura, P.B.; Chung, W.S.; Zhou, L.; Cahoy, J.D.; Daneman, R.; Zong, H.; Ellisman, M.H.; et al. Development of a method for the purification and culture of rodent astrocytes. *Neuron* 2011, 71, 799–811. [CrossRef] [PubMed]

308. Wohl, S.G.; Geh, T.A. The microRNA expression profile of mouse Müller glia in vivo and in vitro. *Sci. Rep.* 2016, 6, 35423. [CrossRef] [PubMed]

309. Bohlen, C.J.; Bennett, F.C.; Tucker, A.F.; Collins, H.Y.; Mulinyawe, S.B.; Barres, B.A. Diverse Requirements for Microglial Survival, Specification, and Function Revealed by Defined-Medium Cultures. *Neuron* 2017, 94, 759–773.e758. [CrossRef] [PubMed]

310. Sloan, S.A.; Darmanis, S.; Huber, N.; Khan, T.A.; Birey, F.; Caneda, C.; Reimer, R.; Quake, S.R.; Barres, B.A.; Pašca, S.P. Human Astrocyte Maturation Captured in 3D Cerebral Cortical Spheroids Derived from Pluripotent Stem Cells. *Neuron* 2017, 95, 779–790.e776. [CrossRef]

311. Falk, T.; Garver, W.S.; Erickson, R.P.; Wilson, J.M.; Yool, A.J. Expression of Niemann-Pick type C transcript in rodent cerebellum in vivo and in vitro. *Brain Res.* 1999, 839, 49–57. [CrossRef]

312. Henderson, L.P.; Lin, L.; Prasad, A.; Paul, C.A.; Chang, T.Y.; Maue, R.A. Embryonic striatal neurons from niemann-pick type C mice exhibit defects in cholesterol metabolism and neurotrophin responsiveness. *J. Biol. Chem.* 2000, 275, 20179–20187. [CrossRef]

313. Karten, B.; Vance, D.E.; Campenot, R.B.; Vance, J.E. Cholesterol accumulates in cell bodies, but is decreased in distal axons, of Niemann-Pick C1-deficient neurons. *J. Neurochem.* 2002, 83, 1154–1163. [CrossRef]

314. Jin, L.W.; Shtie, F.S.; Maezawa, I.; Vincent, I.; Bird, T. Intracellular accumulation of amyloidogenic fragments of amyloid-beta precursor protein in neurons with Niemann-Pick type C defects is associated with endosomal abnormalities. *Am. J. Pathol.* 2004, 164, 975–985. [CrossRef]

315. Tashiro, Y.; Yamazaki, T.; Shimada, Y.; Ohno-Iwashita, Y.; Okamoto, K. Axon-dominant localization of cell-surface cholesterol in cultured hippocampal neurons and its disappearance in Niemann-Pick type C model cells. *Eur. J. Neurosci.* 2004, 20, 2015–2021. [CrossRef] [PubMed]

316. Wasser, C.R.; Ertunc, M.; Liu, X.; Kavalali, E.T. Cholesterol-dependent balance between evoked and spontaneous synaptic vesicle recycling. *J. Physiol.* 2007, 579, 413–429. [CrossRef] [PubMed]

317. Demais, V.; Barthélémé, A.; Perraut, M.; Ungerer, N.; Keime, C.; Reibel, S.; Pfrieger, F.W. Reversal of Pathologic Lipid Accumulation in NPC1-Deficient Neurons by Drug-Promoted Release of LAMP1-Coated Lamellar Inclusions. *J. Neurosci.* 2016, 36, 8012–8025. [CrossRef] [PubMed]

318. Buard, I.; Pfrieger, F.W. Relevance of neuronal and glial NPC1 for synaptic input to cerebellar Purkinje cells. *Mol. Cell Neurosci.* 2014, 61, 65–71. [CrossRef] [PubMed]

319. Hawes, C.M.; Wiemer, H.; Krueger, S.R.; Karten, B. Pre-synaptic defects of NPC1-deficient hippocampal neurons are not directly related to plasma membrane cholesterol. *J. Neurochem.* 2010, 114, 311–322. [CrossRef]

320. Suresh, S.; Yan, Z.; Patel, R.C.; Patel, Y.C.; Patel, S.C. Cellular cholesterol storage in the Niemann-Pick disease type C mouse is associated with increased expression and defective processing of apolipoprotein D. *J. Neurochem.* 1998, 70, 242–251. [CrossRef]

321. Strauss, K.; Goebel, C.; Runz, H.; Mobius, W.; Weiss, S.; Feussner, I.; Simons, M.; Schneider, A. Exosome secretion ameliorates lysosomal storage of cholesterol in Niemann-Pick type C disease. *J. Biol. Chem.* 2010, 285, 26279–26288. [CrossRef]

322. Yang, F.; Feng, X.; Rolfs, A.; Luo, J. Lovastatin promotes myelin formation in NPC1 mutant oligodendrocytes. *J. Neurosci.* 2018, 38, 56–63. [CrossRef]

323. De Nuccio, C.; Bernardo, A.; Ferrante, A.; Pepponi, R.; Martire, A.; Falchi, M.; Visentin, S.; Popoli, P.; Minghetti, L. Adenosine A(2A) receptor stimulation restores cell functions and differentiation in Niemann-Pick type C-like oligodendrocytes. *Sci. Rep.* 2019, 9, 9782. [CrossRef]

324. Peake, K.B.; Campenot, R.B.; Vance, D.E.; Vance, J.E. Niemann-Pick Type C1 deficiency in microglia does not cause neuron death in vitro. *Biochim. Biophys. Acta* 2011, 1812, 1121–1129. [CrossRef]

325. Seo, Y.; Kim, H.S.; Kang, I.; Choi, S.W.; Shin, T.H.; Shin, J.H.; Lee, B.C.; Lee, J.Y.; Kim, J.J.; Kook, M.G.; et al. Cathepsin S contributes to microglia-mediated olfactory dysfunction through the regulation of Cx3cl1-Cx3cr1 axis in a Niemann-Pick disease type C1 model. *Glia* 2016, 64, 2291–2305. [CrossRef] [PubMed]

326. Walterfang, M.; Di Biase, M.A.; Cropley, V.L.; Scott, A.M.; O’Keefe, G.; Velakoulis, D.; Pathmaraj, K.; Ackermann, U.; Pantelis, C. Imaging of neuroinflammation in adult Niemann-Pick type C disease: A cross-sectional study. *Neurology* 2020, 94, e1716–e1725. [CrossRef] [PubMed]
327. Marschalek, N.; Albert, F.; Meske, V.; Ohm, T.G. The natural history of cerebellar degeneration of Niemann-Pick C mice monitored in vitro. *Neuropathol. Appl. Neurobiol.* 2014, 40, 933–945. [CrossRef] [PubMed]

328. Reid, P.C.; Sugii, S.; Chang, T.Y. Trafficking defects in endogenously synthesized cholesterol in fibroblasts, macrophages, hepatocytes, and glial cells from Niemann-Pick type C1 mice. *J. Lipid Res.* 2003, 44, 1010–1019. [CrossRef]

329. Vainio, S.; Bykow, I.; Hermansson, M.; Jokitalo, E.; Somerharju, P.; Ikonen, E. Defective insulin receptor activation and altered lipid rafts in Niemann-Pick type C disease hepatocytes. *Biochim. J.* 2005, 391, 465–472. [CrossRef]

330. Teratani, T.; Tomita, K.; Suzuki, T.; Oshikawa, T.; Yokoyama, H.; Shimamura, K.; Tominaga, S.; Hiroi, S.; Irie, R.; Okada, Y.; et al. A high-cholesterol diet exacerbates liver fibrosis in mice via accumulation of free cholesterol in hepatic stellate cells. *Gastroenterology* 2012, 142, 152–164.e110. [CrossRef]

331. Te Vruchte, D.; Jeans, A.; Platt, F.M.; Sillence, D.J. Glycosphingolipid storage leads to the enhanced degradation of the B cell receptor in Sandhoff disease mice. *J. Inherit. Metab. Dis.* 2010, 33, 261–270. [CrossRef]

332. Zhang, J.R.; Coleman, T.; Langmade, S.J.; Scherrer, D.E.; Lane, L.; Lanier, M.H.; Feng, C.; Sands, M.S.; Schaffer, J.E.; Semenkovich, C.F.; et al. Niemann-Pick C1 protects against atherosclerosis in mice via regulation of macrophage intracellular cholesterol trafficking. *J. Clin. Investig.* 2008, 118, 2281–2290. [CrossRef]

333. Roszell, B.R.; Tao, J.Q.; Yu, K.J.; Huang, S.; Bates, S.R. Characterization of the Niemann-Pick C pathway in alveolar type II cells and lamellar bodies of the lung. *Am. J. Physiol. Lung Cell Mol. Physiol.* 2012, 302, L919–L932. [CrossRef] [PubMed]

334. Cougnoux, A.; Movassaghi, M.; Picache, J.A.; Iben, J.R.; Navid, F.; Salman, A.; Martin, K.; Farhat, N.Y.; Chuzeau, C.; Tseng, W.C.; et al. Gastrointestinal Tract Pathology in a BALB/c Niemann-Pick Disease Type C1 Null Mouse Model. *Dig. Dis. Sci.* 2018, 63, 870–880. [CrossRef]

335. Speak, A.O.; Platt, N.; Salio, M.; te Vruchte, D.; Smith, D.A.; Shepherd, D.; Veerapen, N.; Besra, G.S.; Yanjinan, N.M.; Simmons, L.; et al. Invariant natural killer T cells are not affected by lysosomal storage in patients with Niemann-Pick disease type C. *Eur. J. Immunol.* 2012, 42, 1886–1892. [CrossRef]

336. Pereira, C.S.; Pérez-Cabezas, B.; Ribeiro, H.; Maia, M.L.; Cardoso, M.T.; Dias, A.F.; Azevedo, O.; Ferreira, M.F.; García, P.; Rodrigues, E.; et al. Lipid Antigen Presentation by CD1b and CD1d in Lysosomal Storage Disease Patients. *Front. Immunol.* 2019, 10, 1264. [CrossRef] [PubMed]

337. Sagiv, Y.; Hudspeth, K.; Mattner, J.; Schrantz, N.; Stern, R.K.; Zhou, D.; Savage, P.B.; Teyton, L.; Bendelac, A. Cutting edge: Impaired glycosphingolipid trafficking and NKT cell development in mice lacking Niemann-Pick type C1 protein. *J. Immunol.* 2006, 177, 26–30. [CrossRef] [PubMed]

338. Platt, N.; Speak, A.O.; Colaco, A.; Gray, J.; Smith, D.A.; Williams, I.M.; Walлом, K.L.; Platt, F.M. Immune dysfunction in Niemann-Pick disease type C. *J. Neurochem.* 2016, 136, 74–80. [CrossRef] [PubMed]

339. Csepeegi, C.; Jiang, M.; Frolov, A. Somatic cell plasticity and Niemann-pick type C2 protein: Adipocyte differentiation and function. *J. Biol. Chem.* 2010, 285, 30347–30354. [CrossRef] [PubMed]

340. Busso, D.; Oñate-Alvarado, M.J.; Balboa, E.; Castro, J.; Lizama, C.; Morales, G.; Vargas, S.; Härtel, S.; Moreno, R.D.; Zanlungo, S. Spermatozoa from mice deficient in Niemann-Pick disease type C2 (NPC2) protein have defective cholesterol content and reduced in vitro fertilising ability. *Reprod. Fertil. Dev.* 2014, 26, 609–621. [CrossRef]

341. Yang, S.R.; Kim, S.J.; Byun, K.H.; Hutchinson, B.; Lee, B.H.; Michikawa, M.; Lee, Y.S.; Kang, K.S. NPC1 gene deficiency leads to lack of neural stem cell self-renewal and abnormal differentiation through activation of p38 mitogen-activated protein kinase signaling. *Stem. Cells* 2006, 24, 292–298. [CrossRef]

342. Ordonez, M.P.; Roberts, E.A.; Kidwell, C.U.; Yuan, S.H.; Plaisted, W.C.; Goldstein, L.S.B. Disruption and therapeutic rescue of autophagy in a human neuronal model of Niemann Pick type C1. *Hum. Mol. Genet.* 2012, 21, 2651–2662. [CrossRef]

343. Bergamin, N.; Dardis, A.; Beltrami, A.; Cesselli, D.; Rigo, S.; Zampieri, S.; Domenis, R.; Bembi, B.; Beltrami, C.A. A human neuronal model of Niemann Pick C disease developed from stem cells isolated from patient’s skin. *Orphanet J. Rare Dis.* 2013, 8, 34. [CrossRef]

344. Trlck, M.; Hübner, R.; Seibler, P.; Klein, C.; Rolfs, A.; Frech, M.J. Niemann-Pick type C1 patient-specific induced pluripotent stem cells display disease specific hallmarks. *Orphanet J. Rare Dis.* 2013, 8, 144. [CrossRef]
345. Lee, H.; Lee, J.K.; Park, M.H.; Hong, Y.R.; Marti, H.H.; Kim, H.; Okada, Y.; Otsu, M.; Seo, E.J.; Park, J.H.; et al. Pathological roles of the VEGF/SphK pathway in Niemann-Pick type C neurons. *Nat. Commun.* 2014, 5, 5514. [CrossRef]

346. Maetzel, D.; Sarkar, S.; Wang, H.; Abi-Mosleh, L.; Xu, P.; Cheng, A.W.; Gao, Q.; Mitalipova, M.; Jaenisch, R. Genetic and Chemical Correction of Cholesterol Accumulation and Impaired Autophagy in Hepatic and Neural Cells Derived from Niemann-Pick Type C Patient-Specific iPS Cells. *Stem. Cell Rep.* 2014, 2, 866–880. [CrossRef] [PubMed]

347. Yu, D.; Swaroop, M.; Wang, M.; Baxa, U.; Yang, R.; Yan, Y.; Coksaygan, T.; DeTolla, L.; Marugan, J.J.; Austin, C.P.; et al. Niemann-Pick Disease Type C: Induced Pluripotent Stem Cell-Derived Neuronal Cells for Modeling Neural Disease and Evaluating Drug Efficacy. *J. Biomol. Screen* 2014, 19, 1164–1173. [CrossRef] [PubMed]

348. Efthymiou, A.G.; Steiner, J.; Pavan, W.J.; Winiovitch, S.; Larson, D.M.; Porter, F.D.; Rao, M.S.; Malik, N. Rescue of an in vitro neuron phenotype identified in Niemann-Pick disease, type C1 induced pluripotent stem cell-derived neurons by modulating the WNT pathway and calcium signaling. *Stem Cells Transl. Med.* 2015, 4, 230–238. [CrossRef] [PubMed]

349. Völkner, C.; Peter, F.; Liedtke, M.; Krohn, S.; Lindner, I.; Murua Escobar, H.; Cimmaruta, C.; Lukas, J.; Hermann, A.; Frech, M.J. Generation of the Niemann–Pick type C2 patient-derived iPSC line AKOSi001-A. *Stem Cell Res.* 2019, 41, 101606. [CrossRef]

350. Kuo, S.Y.; Castoreno, A.B.; Aldrich, L.N.; Lassen, K.G.; Goel, G.; Dančík, V.; Kuballa, P.; Latorre, I.; Conway, K.L.; Sarkar, S.; et al. Small-molecule enhancers of autophagy modulate cellular disease phenotypes suggested by human genetics. *Proc. Natl. Acad. Sci. USA* 2015, 112, E4281–E4287. [CrossRef]

351. Gläser, A.; Hammerl, F.; Gräler, M.H.; Coldewey, S.M.; Völkner, C.; Frech, M.J.; Yu, D.; Yang, R.; Luo, J.; Tönnies, E.; et al. Identification of Brain-Specific Treatment Effects in NPC1 Disease by Focusing on Cellular and Molecular Changes of Sphingosine-1-Phosphate Metabolism. *Int. J. Mol. Sci.* 2020, 21, 4520. [CrossRef]

352. Liscum, L.; Arnio, E.; Anthony, M.; Howley, A.; Sturley, S.L.; Agler, M. Identification of a pharmaceutical compound that partially corrects the Niemann-Pick C phenotype in cultured cells. *J. Lipid Res.* 2002, 43, 1708–1717. [CrossRef]

353. Swaroop, M.; Thorne, N.; Rao, M.S.; Austin, C.P.; McKew, J.C.; Zheng, W. Evaluation of cholesterol reduction activity of methyl-beta-cyclodextrin using differentiated human neurons and astrocytes. *J. Biomol. Screen.* 2012, 17, 1243–1251. [CrossRef]

354. Sung, E.-A.; Yu, K.-R.; Shin, J.-H.; Lassen, K.G.; Goel, G.; Dančík, V.; Kuballa, P.; Latorre, I.; Conway, K.L.; Sarkar, S.; et al. Small-molecule enhancers of autophagy modulate cellular disease phenotypes suggested by human genetics. *Proc. Natl. Acad. Sci. USA* 2015, 112, E4281–E4287. [CrossRef]

355. Pipalia, N.H.; Huang, A.; Ralph, H.; Rujoi, M.; Maxfield, F.R. Automated microscopy screening for compounds that partially revert cholesterol accumulation in Niemann-Pick C cells. *J. Lipid Res.* 2006, 47, 284–301. [CrossRef] [PubMed]

356. Xu, M.; Liu, K.; Swaroop, M.; Porter, F.D.; Sidhu, R.; Firnkes, S.; Ory, D.S.; Marugan, J.J.; Xiao, J.; Southall, N.; et al. δ-Tocopherol reduces lipid accumulation in Niemann-Pick type C1 and Wolman cholesterol storage disorders. *J. Biol. Chem.* 2012, 287, 39349–39360. [CrossRef] [PubMed]

357. Pugach, E.K.; Feltes, M.; Kaufman, R.J.; Ory, D.S.; Bang, A.G. High-content screen for modifiers of Niemann-Pick type C disease in patient cells. *Hum. Mol. Genet.* 2018, 27, 2101–2112. [CrossRef] [PubMed]

358. Platt, F.M.; Neises, G.R.; Dwek, R.A.; Butters, T.D. N-butyldeoxynojirimycin is a novel inhibitor of glycolipid biosynthesis. *J. Biol. Chem.* 1994, 269, 8362–8365. [CrossRef]

359. Cruz, J.C.; Chang, T.Y. Fate of endogenously synthesized cholesterol in Niemann-Pick type C1 cells. *J. Biol. Chem.* 2000, 275, 41309–41316. [CrossRef]

360. Zervas, M.; Somers, K.L.; Thrall, M.A.; Walkley, S.U. Critical role for glycosphingolipids in Niemann-Pick disease type C. *Curr. Biol.* 2001, 11, 1283–1287. [CrossRef]

361. Crini, G. Review: A history of cyclodextrins. *Chem. Rev.* 2014, 114, 10940–10975. [CrossRef]

362. Camargo, F.; Erickson, R.P.; Garver, W.S.; Hossain, G.S.; Carbone, P.N.; Heidenreich, R.A.; Blanchard, J. Cyclodextrins in the treatment of a mouse model of Niemann-Pick C disease. *Life Sci.* 2001, 70, 131–142. [CrossRef]
363. Davidson, J.; Molitor, E.; Moores, S.; Gale, S.E.; Subramanian, K.; Jiang, X.; Sidhu, R.; Bell, P.; Zhang, J.; Fujiwara, H.; et al. 2-Hydroxypropyl-β-cyclodextrin is the active component in a triple combination formulation for treatment of Niemann-Pick C1 disease. Biochim. Biophys. Acta Mol. Cell Biol. Lipids 2019, 1864, 1545–1561. [CrossRef]

364. Kao, M.L.; Stellar, S.; Solon, E.; Lordi, A.; Kasica, N.; Swain, G.; Bagel, J.H.; Gurda, B.L.; Vite, C.H. Pharmacokinetics and distribution of 2-hydroxypropyl-β-cyclodextrin following a single intrathecal dose to cats. J. Inherit. Metab. Dis. 2020, 43, 618–634. [CrossRef]

365. Pontikis, C.C.; Davidson, C.D.; Walkley, S.U.; Platt, F.M.; Begley, D.J. Cyclodextrin alleviates neuronal storage of cholesterol in Niemann-Pick C1 neurons without evidence of detectable blood-brain barrier permeability. J. Inherit. Metab. Dis. 2013, 36, 491–498. [CrossRef] [PubMed]

366. Davidson, C.D.; Fishman, Y.I.; Puskas, I.; Szeman, J.; Sohajda, T.; McCauli, M.; Moniatte, M.; Gruenberg, J. Susceptibility of outer hair cells to cholesterol chelator 2-hydroxypropyl-β-cyclodextrine is prein-dependent. Sci Rep. 2016, 6, 21973. [PubMed]

367. Cougnoux, A.; Drummond, R.A.; Collar, A.L.; Iben, J.R.; Salman, A.; Westgarth, H.; Wassif, C.A.; Sawley, N.X.; Farhat, N.Y.; Ozato, K.; et al. Microglia activation in Niemann-Pick disease, type C1 is amenable to therapeutic intervention. Hum. Mol. Genet. 2018, 27, 2076–2089. [CrossRef] [PubMed]

368. Ebner, L.; Gläser, A.; Bräuer, A.; Kasica, N.; Swain, G.; Bagel, J.H.; Gurda, B.L.; Vite, C.H. Evaluation of outer hair cells to cholesterol chelator 2-hydroxypropyl-β-cyclodextrine is prein-dependent. J. Inherit. Metab. Dis. 2016, 3, 366–380. [CrossRef] [PubMed]

369. Takahashi, S.; Homma, K.; Zhou, Y.; Nishimura, S.; Duan, C.; Chen, J.; Ahmad, A.; Cheatham, M.A.; Zheng, J. Susceptibility of outer hair cells to cholesterol chelator 2-hydroxypropyl-β-cyclodextrine is prein-dependent. J. Biol. Chem. 2012, 287, 9290–9298. [PubMed]

370. Wehrmann, Z.T.; Hulett, T.W.; Huegel, K.L.; Vaughan, K.T.; Wiest, O.; Helquist, P.; Goodson, H. Quantitative comparison of the efficacy of various compounds in lowering intracellular cholesterol levels in niemann-pick type C fibroblasts. PLoS ONE 2010, 5, e15054. [CrossRef]

371. Chen, F.W.; Li, C.; Ioannou, Y.A. Cyclodextrin induces calcium-dependent lysosomal exocytosis. PLoS ONE 2012, 7, e48561. [CrossRef]

372. Abi-Mosleh, L.; Infante, R.E.; Radhakrishnan, A.; Goldstein, J.L.; Brown, M.S. Cyclodextrin overcomes deficient lysosome-to-endoplasmic reticulum transport of cholesterol in Niemann-Pick type C cells. Proc. Natl. Acad. Sci. USA 2009, 106, 19316–19321. [CrossRef]

373. Rosenbaum, A.I.; Zhang, G.; Warren, J.D.; Maxfield, F.R. Endocytosis of beta-cyclodextrins is responsible for cholesterol reduction in Niemann-Pick type C mutant cells. Proc. Natl. Acad. Sci. USA 2010, 107, 5477–5482. [CrossRef]

374. Vacca, F.; Vossos, S.; Mercier, V.; Mooreau, D.; Johnson, S.; Scott, C.C.; Montoya, J.P.; Monti, M.; Gruenberg, J. Cyclodextrin triggers MCOLN1-dependent endo-lysosome secretion in Niemann-Pick type C cells. J. Lipid Res. 2019, 60, 832–843. [CrossRef]

375. Vacca, F.; Vossos, S.; Mercier, V.; Moreau, D.; Johnson, S.; Scott, C.C.; Montoya, J.P.; Monti, M.; Gruenberg, J. Cyclodextrin triggers MCOLN1-dependent endo-lysosome secretion in Niemann-Pick type C cells. J. Lipid Res. 2019, 60, 832–843. [CrossRef]

376. Fen, W.O.; Li, C.; Ioannou, Y.A. Cyclodextrin induces calcium-dependent lysosomal exocytosis. PLoS ONE 2010, 5, e15054. [CrossRef]

377. Chen, F.W.; Li, C.; Ioannou, Y.A. Cyclodextrin induces calcium-dependent lysosomal exocytosis. PLoS ONE 2010, 5, e15054. [CrossRef]

378. Peake, K.B.; Vance, J.E. Normalization of cholesterol homeostasis by 2-hydroxypropyl-β-cyclodextrin in neurons and glia from Niemann-Pick C1 (NPC1)-deficient mice. J. Biol. Chem. 2012, 287, 9290–9298. [CrossRef] [PubMed]

379. Meske, V.; Erz, J.; Priesnitz, T.; Ohm, T.G. The autophagic defect in Niemann-Pick disease type C neurons differs from somatic cells and reduces neuronal viability. Neurobiol. Dis. 2014, 64, 88–97. [CrossRef] [PubMed]

380. Sarkar, S.; Carroll, B.; Buganim, Y.; Maetz, D.; Ng, A.H.; Cassady, J.P.; Cohen, M.A.; Chakraborty, S.; Wang, H.; Spooner, E.; et al. Impaired autophagy in the lipid-storage disorder Niemann-Pick type C disease. Cell Rep. 2013, 5, 1502–1515. [CrossRef]

381. Ory, D.S.; Ottinger, E.A.; Farhat, N.Y.; King, K.A.; Jiang, X.; Weissfeld, L.; Berry-Kravis, E.; Davidson, C.D.; Bianconi, S.; Keener, L.A.; et al. Intrathecal 2-hydroxypropyl-β-cyclodextrin decreases neurological disease progression in Niemann-Pick disease, type C1: A non-randomised, open-label, phase 1-2 trial. Lancet 2017, 390, 1758–1768. [CrossRef]

382. Berry-Kravis, E.; Chin, J.; Hoffmann, A.; Winston, A.; Stoner, R.; LaGorio, L.; Friedmann, K.; Hernandez, M.; Ory, D.S.; Porter, F.D.; et al. Long-Term Treatment of Niemann-Pick Type C1 Disease With Intrathecal 2-Hydroxypropyl-β-Cyclodextrin. Pediatr. Neurol. 2018, 80, 24–34. [CrossRef] [PubMed]
380. Kim, S.J.; Lee, B.H.; Lee, Y.S.; Kang, K.S. Defective cholesterol traffic and neuronal differentiation in neural stem cells of Niemann-Pick type C disease improved by valproic acid, a histone deacetylase inhibitor. *Biochem. Biophys. Res. Commun.* 2007, 360, 593–599. [CrossRef]

381. Pipalia, N.H.; Cosner, C.C.; Huang, A.; Chatterjee, A.; Bourbon, P.; Farley, N.; Helquist, P.; Wiest, O.; Maxfield, F.R. Histone deacetylase inhibitor treatment dramatically reduces cholesterol accumulation in Niemann-Pick type C1 mutant human fibroblasts. *Proc. Natl. Acad. Sci. USA* 2011, 108, 5620–5625. [CrossRef]

382. Nunes, M.J.; Moutinho, M.; Gama, M.J.; Rodrigues, C.M.; Rodrigues, E. Histone deacetylase inhibition decreases cholesterol levels in neuronal cells by modulating key genes in cholesterol synthesis, uptake and efflux. *PloS ONE* 2013, 8, e53394. [CrossRef]

383. Newton, J.; Hait, N.C.; Maceyka, M.; Colaco, A.; Maczis, M.; Wassif, C.A.; Porte, F.D.; Milstien, S.; Platt, N.; et al. FTY720/fingolimod increases NPC1 and NPC2 expression and reduces cholesterol and sphingolipid accumulation in Niemann-Pick type C1 mutant fibroblasts. *FASEB J.* 2017, 31, 1719–1730. [CrossRef]

384. Subramanian, K.; Hutt, D.M.; Scott, S.M.; Gupta, V.; Mao, S.; Balch, W.E. Correction of Niemann-Pick type C1 trafficking and activity with the histone deacetylase inhibitor valproic acid. *J. Biol. Chem.* 2020, 295, 8017–8035. [CrossRef]

385. Subramanian, K.; Rauniyar, N.; Lavallée-Adam, M.; Yates, J.R., 3rd; Balch, W.E. Quantitative Analysis of the Proteome Response to the Histone Deacetylase Inhibitor (HDACi) Vorinostat in Niemann-Pick Type C1 disease. *Mol. Cell Proteom.* 2017, 16, 1938–1957. [CrossRef] [PubMed]

386. Contreras, P.S.; Gonzalez-Zuñiga, M.; Gonzalez-Hódar, L.; Yáñez, M.J.; Dulcey, A.; Marugan, J.; Seto, E.; Alvarez, A.R.; Zanlungo, S. Neuronal gene repression in Niemann-Pick type C1 mice models is mediated by the c-Abi/HDAC2 signaling pathway. *Biochim. Biophys. Acta* 2016, 1859, 269–279. [CrossRef]

387. Alam, M.S.; Getz, M.; Haldar, K. Chronic administration of an HDAC inhibitor treats both neurological and systemic Niemann-Pick type C disease in a mouse model. *Sci. Transl. Med.* 2016, 8, 326ra323. [CrossRef] [PubMed]

388. Nakasone, N.; Nakamura, Y.S.; Higaki, K.; Oumi, N.; Ohno, K.; Ninomiya, H. Endoplasmic reticulum-associated degradation of Niemann-Pick C1: Evidence for the role of heat shock proteins and identification of lysine residues that accept ubiquitin. *J. Biol. Chem.* 2014, 289, 19714–19725. [CrossRef]

389. Paul, C.A.; Reid, P.C.; Boegle, A.K.; Karten, B.; Zhang, M.; Jiang, Z.G.; Franz, D.; Lin, L.; Chang, T.Y.; Vance, J.E.; et al. Adenovirus expressing an NPC1-GFP fusion gene corrects neuronal and nonneuronal defects associated with Niemann pick type C disease. *J. Neurosci. Res.* 2005, 81, 706–719. [CrossRef]

390. Loftus, S.K.; Erickson, R.P.; Walkley, S.U.; Bryant, M.A.; Incao, A.; Heidenreich, R.A.; Pavan, W.J. Rescue of neurodegeneration in Niemann-Pick C mice by a prion-promoter-driven Npc1 cDNA transgene. *Hum. Mol. Genet.* 2002, 11, 3107–3114. [CrossRef] [PubMed]

391. Luo, J.; Yang, H.; Song, B.L. Mechanisms and regulation of cholesterol homeostasis. *Nat. Rev. Mol. Cell Biol.* 2020, 21, 225–245. [CrossRef] [PubMed]

392. Pfrieger, F.W.; Ungerer, N. Cholesterol metabolism in neurons and astrocytes. *Prog. Lipid Res.* 2011, 50, 357–371. [CrossRef] [PubMed]

393. Martin, M.G.; Pfrieger, F.; Dotti, C.G. Cholesterol in brain disease: Sometimes determinant and frequently implicated. *EMBO Rep.* 2014, 15, 1036–1052. [CrossRef]

394. Arenas, F.; Garcia-Ruiz, C.; Fernandez-Checa, J.C. Intracellular Cholesterol Trafficking and Impact in Neurodegeneration. *Front. Mol. Neurosci.* 2017, 10, 382. [CrossRef]

395. Lamri, A.; Pigeyre, M.; Garver, W.S.; Meyre, D. The Extending Spectrum of NPC1-Related Human Disorders: From Niemann-Pick C1 Disease to Obesity. *Endocr. Rev.* 2018, 39, 192–220. [CrossRef] [PubMed]

396. Trezza, V.; Campolongo, P.; Vanderschuren, L.J. Evaluating the rewarding nature of social interactions in laboratory animals. *Dev. Cogn. Neurosci.* 2011, 1, 444–458. [CrossRef] [PubMed]

397. Halliwell, J.; Barbaric, I.; Andrews, P.W. Acquired genetic changes in human pluripotent stem cells: Origins and consequences. *Nat. Rev. Mol. Cell Biol.* 2020, 21, 715–728. [CrossRef] [PubMed]

398. Horvath, P.; Aulner, N.; Bickle, M.; Davies, A.M.; Nery, E.D.; Ebner, D.; Montoya, M.C.; Östling, P.; Pietiäinen, V.; Price, L.S.; et al. Screening out irrelevant cell-based models of disease. *Nat. Rev. Drug Discov.* 2016, 15, 751–769. [CrossRef] [PubMed]

399. Kim, J.; Koo, B.K.; Knoblich, J.A. Human organoids: Model systems for human biology and medicine. *Nat. Rev. Mol. Cell Biol.* 2020, 21, 571–584. [CrossRef] [PubMed]
400. Krystal, J.H.; State, M.W. Psychiatric disorders: Diagnosis to therapy. *Cell* 2014, *157*, 201–214. [CrossRef] [PubMed]

401. Becker, R.E.; Seeman, M.V.; Greig, N.H.; Lahiri, D.K. What can triumphs and tribulations from drug research in Alzheimer’s disease tell us about the development of psychotropic drugs in general? *Lancet Psychiatry* 2015, *2*, 756–764. [CrossRef]

402. R Core Team. *R: A Language and Environment for Statistical Computing*; R Foundation for Statistical Computing: Vienna, Austria, 2019.

403. Dowle, M. Data.Table: Extension of ‘Data.Frame’. Available online: https://CRAN.R-project.org/package=data.table (accessed on 25 November 2020).

404. Wickham, H. *Ggplot2: Elegant Graphics for Data Analysis*; Springer: New York, NY, USA, 2016.

405. Wickham, H. Readr: Read Rectangular Text Data. Available online: https://cran.r-project.org/web/packages/readr/index.html (accessed on 25 November 2020).

**Publisher’s Note:** MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.

© 2020 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (http://creativecommons.org/licenses/by/4.0/).