Pfrieger's Digest for Niemann-Pick Type C
Summaries of research advances based on selected peer-reviewed publications in scientific journals.

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Dear Readers.
Welcome to a new edition of Pfrieger's Digest, which covers the period from 1st of September 2021 to 30th of April 2022. The link for the PubMed query is:

((niemann-pick type C disease OR niemann-pick disease type C OR niemann-pick type C1 OR niemann-pick type c2 OR npc1 OR npc2) AND (("2021/09/01"[Date - Publication] : "2022/04/30"[Date - Publication]))) NOT (("2020/01/01"[Date - Publication] : "2021/8/31"[Date - Publication]))

During this period, **102** articles were published in scientific journals including **9** reviews. Notably, the query covers articles where relevant terms appear in their title, abstract/summary or keyword list. Therefore, a fraction of articles is not directly related to Niemann-Pick type C disease (NPC). If – just for fun – the query is limited to titles and abstract/summary by adding "[tiab]" to each search term, it yields **89** articles including **8** reviews.

The following applies as stated previously: 1) My selection is subjective, 2) I do not comment on review articles or case studies, 3) I only describe articles that I can read entirely, 4) I try to ensure correctness of statements, but I cannot guarantee it, 5) my judgements and interpretations expressed in this document are subjective and reflect my personal opinion, they do not claim any validity, 6) I apologize for errors (grammar, orthography etc.) and any wrong, quirky or otherwise weird expressions. As for previous issues, the original German version of this one was translated by a non-native speaker – i.e. me. Please feel free to distribute and forward this issue. Feedback welcome to: fw-pfrieger@gmx.de or frank.pfrieger@unistra.fr.

Patients

https://pubmed.ncbi.nlm.nih.gov/34682919
https://pubmed.ncbi.nlm.nih.gov/35242574

Based on 26 NPC1 or NPC2 patients Dardis and colleagues corroborate previous evidence that the neurofilament L protein (for light chain) can be used as biomarker. Neurofilament is an important structural component of nerve cells (neurons), where it lives mainly in their extensions, the axons. As often in biology, there are several forms that differ in size – light, middle and heavy chain. Once neurons die, these proteins probably reach the extracellular space, then the cerebrospinal fluid and finally the bloodstream. The group found elevated plasma levels of neurofilament L in young patients compared to age-matched controls. In older patients, the difference was largely lost, probably because of an age-dependent but disease-independent increase in extracellular neurofilament – the older the more dead neurons! Therefore, neurofilament L is not a specific marker for NPC. On the other hand, it seems to indicate the presence of neurologic symptoms. Therefore, this protein may help to monitor disease progression and potential drug effects. The second article by Welford and colleagues shows that blood levels of neurofilament L are also strongly elevated in patients with gangliosidoses.

https://pubmed.ncbi.nlm.nih.gov/34794481

The publication by Evans and colleagues asks an old but important question: how can we assess quantitatively – meaning with numbers – the actual state of NPC patients and the disease progression. Biomarkers are not the whole story. The prevalent answer are severity scales. In the new study, the socalled Delphi process (named after the Greek oracle although I am not sure this term makes sense to me) was used to define a sort of basic consensus about severity scales using successive questioning of 20 physicians from seven countries. Indeed, over the years the NPC field has developed or used six different severity scales covering distinct cateories. The consensus of the new study was that the 17 domain severiy scale is the first choice to estimate disease severity and progress in clinical studies. Moreover, the group agreed that the reduced five domain scale covering ambulation, cognition, fine motor control, speech and swallowing is sufficient for daily clinical practice. It was noted that the number of participants was relatively small and that it was not easy to reach a consensus. It not clear though who selected the experts based on what.

https://pubmed.ncbi.nlm.nih.gov/34819124

A new study by Mengel et al. asks the important and seemingly easy question how does NPC impact the quality of life of patients and of their caregivers, which are

mostly family members. To this end, 43 family members with patients of different ages and six adult NPC patients from the US and UK were polled. The goal was to obtain quantitative, number-based data. The results aren't surprising, only few shall be mentioned. Caregivers rated as the most severe categories affected by disease ambulation, swallowing, language, memory loss and cognition. Accordingly, these are part of the severity scales. How do these symptoms impact the quality of life of patients. Most participants mentioned frustration, inability to pursue favourite activities, fear and reduction of social contacts. For caregivers, changes in daily life, additional time required for care, pressure to be vigilant certainly during meals, and transport of patients. Caregivers further suffer from sadness, fear, distress and social isolation. I assume that this is nothing new for many readers. Still, it's probably helpful to document this black on white and with numbers, last not least to inform regulatory bodies.

https://pubmed.ncbi.nlm.nih.gov/34810067
A study from the vast children's hospital *Bambino Gesù* in Rome, Italy, shows that a sort of fat fingerprint in blood, reporting the concentrations of many distinct lipids differs strongly between NPC patients and healthy controls. The group of Federica Deodato found distinct subgroups of patients with distinct lipid patterns in their blood. It remains to be seen whether such fingerprints can serve as biomarkers.

https://pubmed.ncbi.nlm.nih.gov/35389779
https://pubmed.ncbi.nlm.nih.gov/35389781/
https://pubmed.ncbi.nlm.nih.gov/35513515
Attention, please, I will try something new: I will write about three new studies that are unrelated to NPC – they are about autoimmune diseases (Yazar et al., 2022; Perez et al., 2022 both published in SCIENCE) and Parkinson disease (Kamath et al., 2022 Nature Neuroscience). In my opinion, they provide new insight that is relevant for NPC – and many other diseases. What is it all about? Two important questions: first, why do only specific neurons die in NPC? Second, why do the symptoms, disease progress and reactions to therapeutic drugs differ from patient to patient. High on the list of possible culprits figure genetic factors that vary between human individuals. How does this work? There are probably genes/proteins that must be expressed in cell X to ensure its function and that at the same time increase the vulnerability of cell X to a defect in NPC1. In addition, there are small variants of the genetic code spread all over the DNA that may affect expression of NPC1 – regardless whether it's mutated. One example are the socalled *single nucleotide polymorphisms*. Assume that the genetic code reads at whatever location of the DNA – A for adenin in human number 1 and T for Tymin in human number 2. So, does

this small variant impact how much NPC1 is produced by cells in human 1 and 2 or how cells react to damaged NPC1. The answer is "maybe", we need to find out. The identification of these disease modifiers is a major goal of biomedical research and therefore, many studies are searching for the modifiers – however few for NPC and rare diseases in general. The two articles in SCIENCE show now that the impact of the genetic variants varies even from cell to cell. In our example, variant A could increase expression of NPC1 in cell type X or decrease it in cell type Y of human 1, whereas the variant T could... you get the picture! If this is true, the whole effort to identify these variants and their impact will increase drastically.

The third study is about Parkinson's disease. This similarly Herculean work investigated gene expression in hundreds of thousands single cells from post-mortem brains of patients and age-matched healty controls. This study identifies at least ten types of dopaminergic nerve cells in humans, which differ in the gene expression pattern and notably their vulnerability to Parkinson. Importantly, the study shows that some of these nerve cell types neither exist in mouse nor monkey. So? The take-home message is that we may have to look into single cells, in differently affected brain regions from human post-mortem material. The effort to understand NPC will increase, but with mouse or cat we may not reach our goals. Needless to mention that NPC is not a skin or fibroblast disease. Indeed, hard to swallow, but welcome to biology, the result of hundreds of million years try-error tinkering by evolution. One should mention the enormous resources that went into these and other studies cited here. Some probably cost several million USDs plus many woman/man years.

https://pubmed.ncbi.nlm.nih.gov/35140266

The topics stays with us, there is an article from the NIH (not the Muppett) laboratories (Baxter et al., 2022). This work follows up on an earlier study dealing with two questions: first, whether we can deduce by whatever method from patient skin fibroblasts the age of disease onset and neurologic symptoms. A daring enterprise! The well-known filipin staining cannot really accomplish this. The new work shows that a tool named Lysotracker may allow to do this. This chemical substance likes the acid environment of lysosomes and - if labeled with a dye - allows to stain this part of cells. The accumulation of cholesterol in the endosomal-lysosomal system of NPC cells results in a strong increase of lysosomal volume, which can be detected and measured with the LysoTracker at least in fibroblasts. This does not require a microscope, it can be done with a socalled cytometer (which costs as much as a microscope, though). The machine measures the fluorescence intensity in each cell – and if you spend some more bucks – it will sort the cells ("The Good into the Pot, the Bad into the Crop"). The second part of the study deals with

the aforementioned topic, genetic factors. Baxter and colleagues examined the gene expression patterns of sorted fibroblasts to determine whether there are genes/proteins whose expression level correlates with the LysoTracker signal and thus with the clinical picture of the patient. This was indeed the case, at least roughly. The authors present a list of genes including some component that may serves as therapeutic targets. Now, based on the previously mentioned articles, one could note: the authors studied fibroblasts, but their genetic modifiers may not work in brain cells. And the other way round: genetic factors active in brain cells may not be detectable in fibroblasts. Which in turn raises the question whether there are subtypes of fibroblasts where genetic factors have distinct effects. That's the crux (and charm) of biology: each new answer to an old question provokes new questions.

https://pubmed.ncbi.nlm.nih.gov/35164809
A study of Bolton and colleagues analyses data from the international NPC patient registry illustrating its usefulness. To mention a few points: of the 203 registered patients 10 died from the fatal neonatal form and 168 showed neurologic symptoms. This represents 87% of patients, where these symptoms could be noticed. Of the 97 patients with known genetics, a third shows the I1062T variant. And 63% of patients take Miglustat/Zavesca.

Animal models
https://pubmed.ncbi.nlm.nih.gov/34410604/
A Chinese group reports something quite new (Han et al., 2021). The authors studied changes in the socalled *long non-coding RNA* (or lncRNA) in NPC1-deficient mice compared to wildtype animals. A bit of biology: The DNA contains instructions to make cells and ultimately an entire human being. Classically, the cellular machinery transcribes the parts of the DNA containing the gene information to messenger RNA, which is then used to make proteins in a process called translation. For a long time, it was thought "That's it!". Decoding the DNA revealed many stretches that contained non-sense, at least scientists thought so. But that's not true. Many of these stretches are transcribed to RNA except that these snippets do not code for proteins, they are non-coding. Moreover, this *lncRNA* comes in two versions, long and short. Cells probably make thousands of these threads. The question is what for. Since a few years, there is a true hype around this stuff, since it was found that lncRNAs regulate all sorts of processes in specialized cells including for example cell division: of big interest for the "tumor field"! There are also links to neurodegeneration. So far, it is unclear how many and which of these lncRNAs are truly non coding and have

regulatory roles. Evidently, these snippets may serve as biomarkers or as therapeutic targets, the hunt is open: *hot topic!*
The Chinese group reports that NPC1 deficiency in mice changes the amounts of specific lncRNAs, which may influence cells and their interactions. One of them bears the name H19. Important questions are raised: what is their function? Do they contribute to the disease, and if so, how? This topic will probably stay with us.

https://pubmed.ncbi.nlm.nih.gov/34580197
The Dickson group (Tiscione et al., 2021) shows how the defect NPC1 may kill nerve cells. This requires a bit of background. It's all about calcium. One may think of bones the other one of water pipes. In cells – notably in nerve cells – calcium serves as an important messenger: the local calcium concentration in cellular compartments controls many processes, notably signal transmission at synapses. Anything wrong with calcium and cells are doomed. Because of this, cells have sophisticated mechanisms to control the calcium concentration in their interior. It's known since some time that NPC deficiency perturbs calcium regulation, the Dickson group contributed to this notion previously. The new work shows a relatively complicated mechanism leading from NPC1 deficiency to death. It looks as if nerve cells with defective NPC1 react to specific stimuli with a strong release of calcium from intracellular stores, which in turn damages mitochondria and leads to cell death. The work brings a new player to the field, the socalled IP3 receceptor type 1, which may serve as new therapeutic target.

https://pubmed.ncbi.nlm.nih.gov/34802899
Another new target is brought in by the Porter group (Cougnoux et al., 2021). They started from earlier observations that NPC1 deficiency decreases the density of glutamate transporters in specific types of glial cells. What's this all about? Well, glutamate is the most frequent neurotransmitter in the brain, as most synapses work with this amino acid. To ensure proper function, glutamate has to be rapidly removed from synapses after its release otherwise excitotoxicty, a sort of death from excitation, looms large. This may occur in other situations in life. However, in the brain glutamate is the culprit. Too much of it overactivates receptors on nerve cells and kills them in the mid-term. So who takes care of the glutamate cleanup? This is done by a specific subtype of glial cells named astrocytes. Their tiny little processes, in the nanometer range, ensheath synapses and carry glutamate transporters. They suck away released glutamate and prevent neuronal "death by excitation". If in NPC too few glutamate transporters are present on glial cells, the loss of neurons could be caused by much glutamate at synapses. Well, *excitotoxicity* is on the menu since a long time as cause of neurodegenerative conditions, and there are approved drugs.

The Porter group tested now one of them named Riluzol, which is used to treat specific forms of multiple sclerosis. Treatment of NPC mice with the drug slowed down progression of neurologic symptoms and prolonged the life span - a bit. It will be interesting to see whether the drug can be tested in patients.

https://pubmed.ncbi.nlm.nih.gov/34944681
A Belgian-German team suggests a new therapeutic approach for NPC (van Hoecke et al., 2021). It is based on extrazellular vesicles, which have become a *hot topic* in biomedical research with the number of publications growing exponentially. It's about a sort of "news exchange" between cells. Extracellular vesicles, also called exosomes, are released by many cells to the extracellular space as their name indicates. These vesicles, sized about 50 to 150 nanometers, contain a cell type-specific mix of signal molecules. They are taken up by target cells, where they induce specific changes depending on their cargo. Nowadays, these vesicles are tested as biomarkers and for the treatment of diseases, notably malignant tumors. One source of these cells are socalled huamnmesenchymal stem cells from bone marrow or umbilical cord. The group shows in NPC1-deficient mice that a multiple injections of extracellular vesicles in the blood stream reduces inflammation and pathologic changes in different organs including the brain. It will be interesting to see how this field evolves.

https://pubmed.ncbi.nlm.nih.gov/34948052
And once again a potential new target uncovered by the Hecimovic group from Zagreb (Dominko et al., 2022). This is about the socalled retromer complex, a central and (evolutionarily) old component of cells – from yeast to humans. The complex regulates the transport of proteins from endosomes to different places within cells. The whole thing is complicated – "quelle surprise" – and incompletely understood. Mutations in the retromer complex cause or contribute to different diseases including Parkinson. The group shows in NPC1-deficient cells lines and in the brain of NPC mice that the intracellular distribution of the retromer complex is perturbed. These results provoke many questions, most importantly namely whether retromer complex is "just" a bystander or whether it can really serve as therapeutic target.

https://pubmed.ncbi.nlm.nih.gov/35081253
A study from Australia (Castiblanko et al., 2022) deals with a specific cell type of the immune system, the cytotoxic T lymphocytes. These cells are a strategic part of the first line immune defense: they kill cells that appear "strange" to them, namely infected cells or tumor cells. To this end, they grab these cells and release topically a mix of toxic stuff to kill their target cells. The toxic components include a protein

named perforin, which drills lethal holes in cellular membranes. Experiments with cells from mice and patients show that in NPC, perforin performs less well, probably because it's littered with the accumulating fat garbage. When treated with cyclodextrin, the whole thing works again. These results suggest that NPC patients have an impaired immune system, the younger the worse. Moreover, activity measurements of cytotoxic T lymphocytes may be useful as test bed to develop therapeutic drugs.

https://pubmed.ncbi.nlm.nih.gov/35452076

Back to the question how to measure disease severity and progress. The problem also applies to mice and other animal models – and evidently to other neurodegenerative diseases. And it becomes pressing with respect to drug development. Not surprisingly, a large number of behavioral tests has been developed within the last decades, notably for mice and rats. A problem here is that often these tests are complicated, require special machines and trained staff. Moreover, it is possible that results from one test to the next cannot be compared. Yerger and colleagues (2022) refined a previously established test that estimates motor behavior in mice in a rather simple and low-cost manner. It comprises five categories and can be performed by "normal" volunteers after a bit of training, but without specific equipment. Do this: look carefully at the mouse with respect to specific features, grab its tail and lift her up and let her walk around. There are points for each category and a total score. It will be interesting to see whether this test will be adopted.

Cell-based models

https://pubmed.ncbi.nlm.nih.gov/34440927/

A study from Chile (Balboa et al., 2021) deals with the recurring topic biomarkers and drug targets. The Zanlungo group looked at liver cells using a meanwhile classic experimental approach. It takes a sort of snap-shot revealing all the proteins in a cell and compares the amount of each protein in healthy and sick cells. Previous studies examined such changes in livers from NPC1-deficient mice and wildtype (normal) littermates. Here, liver cells were isolated from mouse tissue and studied in cell culture. The group could identify roughly 4000 proteins in liver cells. Evidently, these are not all of the proteins that are there, rare proteins or proteins that somehow hide in a inaccessible corner of a cell are often missed. The study reports that NPC1 deficiency changes the levels of 155 proteins, most of which are increased. This includes components regulating lipid metabolism and inflammation. It remains to be seen, which components are of relevance for NPC either as biomarker or drug target.

https://pubmed.ncbi.nlm.nih.gov/34592985
Important news with respect to experimenal models for NPC. Prabhu and colleagues developed a new approach to study the impact of NPC1 variants on nerve cells. This relies on the highly efficient and flexible stem cell technology allowing to convert human stem cells to nerve cells by "genetic reprogramming". The new approach accomplishes the metamorphosis within 10 days. This is record time! Further advantages: the approach uses human cells, but does not require skin fibroblasts, it yields relatively large and homogeneous swarms of nerve cells and it delivers a "clean" isogenic control, meaning the cells are genetically identical ("twins in vitro") except for the NPC variant. This feature eliminates the impact of uncharted genetic background. The work shows that cholesterol accumulates in cells bearing NPC1 mutations. And it shows that broken NPC1 perturbs some nerve cell-specific happenings, for example transport of lysosomes along their prolongations (axons). And nerve cells run out of steam because of damaged mitochondria. Evidently, the new model, named i3Neuron, can be used to hunt for new drugs that save specifically nerve cells. Unclear is of course, how similar these artificial neurons are to the real ones in the brain. Evidently, they come closer than fibroblasts.

https://pubmed.ncbi.nlm.nih.gov/34830064
https://pubmed.ncbi.nlm.nih.gov/35159129
https://pubmed.ncbi.nlm.nih.gov/35159316
In the same context, three studies from the Frech group should be mentioned (Völkner et al., 2021; Völkner et al., 2022; Liedke et al., 2022), they are among the pioneers on the NPC stem cell frontier. In the first work, skin fibroblasts from a female patient with the adult form of NPC were converted first to stem cells and then to nerve and liver cells. The authors determined the distribution of cholesterol and NPC1 within these cells. The results support the idea that a late disease onset is due to residual activity of the corresponding NPC1 variant. The second work addresses new therapeutic approaches. The results deliver first hints that a substance named *abiraterone acetate* reverts the pathologic accumulation of cholesterol in nerve and liver cells in culture. Known targets of this drug are androgen receptors (binding, for example testosterone) and enzymes that produce androgens. Abiraterone is prescribed for the treatment of specific forms of prostate cancer. Further studies notably in animals must show whether the drug is of use for NPC.

The third study addresses pathologic changes in mitochondria of nerve cells that once again were created from fibroblast-derived stem cells. As we learn, this approach is *en vogue*. The new results confirm that defective NPC1 undermines the function, transport and recycling of mitochondria and show that the severity of the

defects depends on the patient. These observations further support the case against mitochondria as culprits of neuronal dysfunction in NPC.

https://pubmed.ncbi.nlm.nih.gov/34296265
A study from the Pavan lab (Rodriguez-Gil et al., 2021) reports results of another Herculean (not Sisyphean!) work. The authors studied how treatment with cyclodextrin affects gene expression in skin fibroblasts from 42 patients. The work shows on the one hand that cyclo affects relatively few genes. On the other hand, it reveals that the reaction to cyclo varies from one patient to the next depending on the patient's sex. Moreover, the study confirms that a protein bearing the cryptic name GPNMB can serve as biomarker. Its level is increased in NPC patients and mice (as well as other diseases) and lowered by cyclo. The function of the protein is largely unknown. It is located in lysosomal membrane, but probably also released by cells, eventually from microglial cells in the brain.

https://pubmed.ncbi.nlm.nih.gov/34851695
And we continue with this peculiar cell type, the fire police and garbage collectors of the brain. The Wyss-Coray group at Stanford University reports something new, but a complicated. The plot starts with a surprise. A previous study of the group published 2019 in NATURE had shown that in mice a specific protein named CD22 inhibits phagocytosis in microglial cells thus lowering their capacity to take care of brain trash. Much to their surprise, the group found now that in the human brain this protein is not produced by microglial cells, but by another type of glial cells: the oligodendrocytes. Those guys produce the cable (myelin) isolation for axons forming nerves. Hundreds of experiments and probably several million dollars down the road the group provides evidence that CD22 may serve as a new therapeutic target for NPC. Fact is that CD22 somehow interferes with lysosomal function in microglial cells. The group identified the *insulin-like growth factor 2 receptor* as part of the complex, which CD22 binds to on microglial cells. The group developed a new cell culture model of microglial cells for NPC. And they used this to show that inhibition of CD22 using - ok, encore, newly developed - antibodies reduces cholesterol accumulation in microglial cells bearing the I1061T variant of NPC1. Apart from all these new results, an important message is that microglial cells from mice and humans differ and that studies using mice may not reveal the true role of microglial cells in NPC.

https://pubmed.ncbi.nlm.nih.gov/34886684
A new player in cellular cholesterol regulation, even a potentialy new binding partner for NPC1, is introduced by the Boucher group working in Strasbourg (Awan

et al., 2022). This team originally worked on arteriosclerosis and the Wnt5a protein. A bit of background: the Wnt proteins form a large family of signaling components. Humans produce 19 different varieties. They shuttle between cells, bind to receptors on target cells (humans have 10 of those) and induce a myriad of actions such as cell division. Why this strange name? Well, admittedly, biologists are at times lazy when naming proteins. The first member of the Wnt family was uncovered in 1981 and named *int1* for *integration site 1*. No details! Later studies found that *int1* is similar to *wingless*, a gene in fruit flies (*Drosophila*) that controls specific phases of their development (wing formation!). At some point, wingless and int1 morphed into *Wnt*. Interestingly, all Wnt proteins are hydrophobic, they don't like water and require a carrier to get through the watery pond between cells. In the case of Wnt5a, these are possibly lipoproteins, which brings us closer to cholesterol. The new study suggests that Wnt5a binds to NPC1 and regulates cholesterol metabolism, mTORC signaling and the production of lysosomes.

https://pubmed.ncbi.nlm.nih.gov/35088900
A Czech group introduced a new biochemical test to determine the accumulation of cholesterol in patient fibroblasts. This test may also allow to distinguish forms of the disease (Majer et al., 2022). It requires cholesterol that contains the isotope deuterium instead of hydrogen and socalled dextrans. These are long, non toxic sugar molecules that are produced on industrial scale by bacteria and used for all sorts of purposes. Dextrans are taken up by cells and degraded. The test may be more precise than the filipin staining, but it requires skin fibroblasts, the chemicals and the necessary equipment.

https://pubmed.ncbi.nlm.nih.gov/34936700
A study from the Pfeffer lab reports results from a new genetic screen. The screen aimed to identify new cellular components that regulate the metabolism and intracellular distribution of cholesterol and LBPA/BMP, a key component of late endosomes (Lu et al., 2022). These screens are a favourite toy of biomedical research to identify genes/proteins that mediate a cellular process of interest (see Pfrieger's Digest 4). The work revealed a new component named SNX13. This protein is located at contact sites between the endoplasmatic reticulum (the protein factory) and endosomes/lysosomes. Notably, its elimination normalizes the cholesterol distribution in NPC1-deficient cells. The group also compared results of different screens and noticed a limited overlap in components. This suggests that each screen identifies new components - probably not surprisingly, as each screen uses distinct cells or animals, tools and measures.

https://pubmed.ncbi.nlm.nih.gov/35190686
This study may turn out groundbreaking for biomedical research - curiously the authors used NPC as an example whereto apply their technology (Erwood et al., 2022). It's once again about the question which variants in the genetic code impact disease severity and progression. One of the hurdles on this course is that most cells of our body (and under study in NPC) are diploid. This means that each cells has two versions of the chromosome and two editions of a gene - for example NPC1. If one wants to unearth the impact of genetic factors, one needs to ensure that the two chromosomes carry the same variant. This is not easily accomplished and extremely rare in patients, where parents mostly contribute distinct variants. An alternative approach are cells that are haploid. They carry only set of chromosomes. However, these guys are unstable and difficult to maintain in the lab – two is better than one: diploid cells win! The group now came up with the idea to "haploidize" the part of the DNA where your gene variants of interest are located and leave the rest as is. Ok, but howdunnit? The colleagues used a new technology that was developed by another group in 2019 and that is called CRISPR prime edition. This advanced version of the CRISPR-CAS is a sort of text editor for DNA allowing for example search/replace operations that seemed undoable hitherto. No details! The whole thing seems to work: the group showed that the method can be used to generate admittedly artificial cells bearing whatever NPC1 mutation and genetic factor. This will allow to assess the impact of these factors on the production, distribution and function of NPC proteins and on cholesterol accumulation. It will be interesting to see whether this technology will be used to advance NPC research and to support the quest for therapeutic approaches.

https://pubmed.ncbi.nlm.nih.gov/35408815
Let's continue with a study from Italy, where a new therapeutic approach was tested in patient fibroblasts (Pepponi et al., 2022). It is based on the observation that patient fibroblasts show a low concentration of adenosine. This molecule – on its own or tied to other molecules – serves as important signal within cells and outside. The group shows that treatment of cells with a drug named dipyrimadol lowers cholesterol accumulation in fibroblasts. The drug is used to treat thromboses and embolies. Studies in animals will have to reveal its usefulness in NPC.

Miscellaneous
https://pubmed.ncbi.nlm.nih.gov/35261748
To all rice lovers: one of the worst pests in rice cultivation which goes by the name *Nilaparvata lugens* or *brown planthopper* has a tiny, but mighty enemy, the fairyfly *Anagrus nilaparvatae*. Chinese colleagues report that this little guy expresses a sort of

NPC2, males more than females. Why is unclear, possibly to transfer odors. Ok, still a long way to your beloved bowl of rice.