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# Optic Neuropathy: Characteristic Pathology for Identifying Drugs with Potential to Cause Mitochondrial Toxicity

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**Abstract:** Deleterious effects on mitochondria are known to be frequently caused by prescription drugs, however, there is no standard and specific testing by pharmaceutical companies on these effects on mitochondria during drug development. The objective of this study is to determine if cases of optic neuropathy are related to drug-induced mitochondrial toxicity in a way that could be used to predict patient harm. To determine this, all cases of optic neuropathy within a fifty-eight-month timeframe were reviewed in the food and drug administration adverse effect reporting system. The drugs listed as the suspected causes of optic neuropathy in each case were analyzed for frequency of occurrence and frequently occurring drugs were evaluated for documented mechanisms of mitochondrial toxicity. The odds of occurrence of drugs with and without mitochondrial toxicity implemented in these cases were compared with all other adverse drug events excluding optic neuropathy. Our findings indicate that mitochondrial toxicity plays a major role in drug-induced optic neuropathy. We found that the drugs with known mitochondrial toxicity had a 1.47 (95% CI 1.30-1.67, $p<0.0001$) times higher risk of causing optic neuropathy compared to drugs associated with non-mitochondrial mechanisms of toxicity. Therefore, optic neuropathy is a significantly correlated adverse event of drugs with mitochondrially toxic properties. Although most of these properties are known, there were still 691 cases of optic neuropathy reported in less than a five-year period, which could have potentially been avoided using effective postmarket research and warnings. Implementing standard mitochondrial toxicity analyses during drug development will help identify drugs with the potential to cause optic neuropathy or other potential harm. Furthermore, several drugs were identified without known mitochondrially toxic mechanisms but possessed significant rates of optic neuropathy suggesting that a mechanism may exist and some of them have been proven mitochondrially toxic in other species.

**Keywords:** Optic Neuropathy, Mitochondrial Toxicity, Adverse Event Reporting

## Introduction

Mitochondria play a role in many physiological processes in our cells, including the production of 95% of the cellular ATP requirement, regulation of apoptosis, beta-oxidation of fatty acids and biosynthesis of home, cholesterol and phospholipids (Vuda and Kamath, 2016; Hynes *et al*., 2006; Hart *et al*., 2013). Mitochondrial dysfunction has been linked to aging, cardiovascular disease, diabetes, neuromuscular disorders and neurological disorders as both an independent event and a pathogenic factor accelerating the progression of chronic disease (Newsholme *et al*., 2012).

As such, mitochondrial dysfunction and toxicity have potential associations with many homeostatic and disease states, some of which are easily characterized due to the high ATP demand of the organ or tissues such as the heart, liver, brain and other nervous system structures such as those related to the eyes. We believe these characterizations are exploitable to help discover unknown mechanisms of mitochondrial toxicity. This type of postmarket analysis provides sufficient evidence that symptoms characteristic of mitochondrial dysfunction such as hepatotoxicity, cardiotoxicity, or

optic neuropathy could be significantly correlated with drugs that have mitochondrially toxic mechanisms and a similar method could be used to find drugs that the FDA or pharmaceutical industry have not identified as mitochondrially toxic.

It has been noted that drugs that do not show immediate evidence of affecting mitochondria can prove surprisingly toxic after the drug administration stops, even years later (Will and Dykens, 2014). The database called MitoTox lists over 870 mitochondrial targets affected by over 4100 different drugs indicating mitochondrial toxicity. Unfortunately, many of these targets have been discovered after the drugs were reported safe and without documented effects on mitochondria (Lin *et al*., 2021). This highlights the importance of such analysis of adverse events. Drugs used to treat conditions that are not primarily induced by mitochondrial pathogenic mechanisms including HIV, diabetes, hypercholesterolemia, cancer, bacterial infections and mental health conditions, have been shown to have off-target toxic effects on mitochondria. Though there is a longstanding concern for the off-target effects of drugs, testing for mitochondrial toxicity is debated as an ethical dilemma and even an impediment to scientific progress. For example, some extremely beneficial drugs such as Metformin, have been shown to be toxic using current assays for mitochondrial toxicity (Stoker *et al*., 2019). If Metformin and similar drugs were deemed toxic too early in the research phase and discontinued, then we would not know their therapeutic potential or be able to assess their benefits. This puts pharmaceutical companies, scientists and physicians in a position to weigh the pros and cons of research, testing and usage of all types of medications that could be toxic to mitochondria (Stoker *et al*., 2019). When and how should assays be performed and if proven toxic to mitochondria, how would it change the course of development of the drug and management of the disease it is intended for.

To further complicate the issue, the vulnerabilities of mitochondria in the population are different when compared between individuals and can change with each person's lifestyle, age and chronic disease conditions. This makes the ability to accurately assess the potential for mitochondrial toxicity before phase III trials or before FDA approval even more difficult. Some researchers concluded that the best solution to this dilemma is to aggressively assess the latest adverse events reported to the FDA that may indicate that a drug is toxic and then determine whether the mechanism of toxicity is acting on mitochondria (Kamitaki *et al*., 2021; Rana *et al*., 2021). It has also been demonstrated that the selective screening of Adverse Drug Events (ADE) databases can yield insight into potentially dangerous drugs however this has not been done thoroughly for mitochondrially toxic drugs or for all symptoms characteristic of mitochondrial toxicity (Rana *et al*., 2021). Our study attempts to bridge this gap by assessing more recently approved drugs that could be toxic to mitochondria based on the outcome of optic neuropathy. This data has implications for treating certain patient populations considered to be at high risk for mitochondrial toxicity, such as patients who are elderly or have a family history of mitochondrial disease or suggestive personal medical history of mitochondrial vulnerability. Data related to optic neuropathy from the FDA Adverse Event Reporting System (FAERS) was collected in order to identify drug classes with the potential to cause mitochondrial toxicity.

With this study, we aim to compare drugs with and without known properties of mitochondrial toxicity to predict the potential of mitochondrial injury. Additionally, this will aid in the care of the community of patients suffering from primary mitochondrial disorders or patients with chronic conditions (aging, metabolic syndrome, cardiovascular diseases) for which acquired mitochondrial defects accelerate their progression (Kamitaki *et al*., 2021). Our study aligns with other efforts that support the concept that selective mitochondrial screening of adverse drug event databases can yield insight into potentially dangerous drugs with the ability to cause mitochondrial toxicity (Rana *et al*., 2021).

## Materials and Methods

### *Design*

The FAERS database was utilized to identify cases where patients presented with optic neuropathy as an adverse drug event. Any drug that was associated with 5 or more cases of optic neuropathy was labeled as high frequency and investigated to determine if the drug had a known mechanism of mitochondrial toxicity. Drugs were then separated into the categories of established mechanisms for mitochondrial toxicity and unestablished mechanisms. FAERS was then utilized again to search for all other adverse events other than mitochondrial toxicity related to each individual drug. High-frequency drugs with mitochondrially toxic mechanisms were compared against other drugs of high frequency of occurrence that did not have identified mitochondrial toxic mechanisms and the likelihood of a drug with and drug without a mitochondrially toxic mechanism to cause optic neuropathy was computed as an odds ratio.

### *Data Collection*

The FAERS public dashboard was developed by the U.S. Food and drug administration to provide a user-friendly way to improve transparency in drug reporting and to permit public access to information on pharmaceutical adverse events. Adverse events are reported by the pharmaceutical industry, healthcare providers and the consumers of pharmaceuticals. The information most frequently reported in cases of optic

neuropathy were the symptoms, the age of the patients, the drugs that were suspected of causing the optic neuropathy and other drugs that the patient may have been taking. Other categories of data recorded with the cases were not always included such as weight, reaction intensity and the actual date of the event. These factors may have been useful to the study but were not reliably available. This data provides an early and up-to-date viewpoint on drug toxicity. The search terms used to find all potential cases of optic neuropathy included "toxic optic neuropathy" and "optic neuropathy". Any adverse drug event where optic neuropathy was reported as a symptom was captured in our search. For the date range, we included cases reported to the FDA between January 1st, 2018 and October 8, 2022. After the drugs of interest were determined based on the frequency of association with cases of optic neuropathy, another search was conducted in FAERS. This second search captured all adverse drug events reported to the FDA in the same period of time. Cases of optic neuropathy or toxic optic neuropathy were excluded and then the same inclusion and exclusion criteria were applied similarly as in the initial search. This created a list of adverse drug event cases with reactions other than optic neuropathy to use for comparison.

### *Inclusion and Exclusion Criteria*

Cases can be reported to FAERS by a variety of entities including healthcare professionals, manufacturers, consumers and unspecified/unknown reporters. Within each of these reporting entities, there could be several individuals who report the ADE including doctors, nurses, pharmacists and health administrators, but also could include drug company employees, lawyers, family members and the patients themselves. Due to varying consistency and potentially conflicting interests of the latter categories, we limited our database to only include cases reported by healthcare professionals. Many cases had excessive amounts of drugs identified as the suspected cause of optic neuropathy. Due to complex drug-drug interactions and possible reporting inconsistencies, cases with more than three drugs identified as the primary cause of optic neuropathy (suspected product name) were excluded. Many cases did not have primary drugs listed but instead only had active ingredients (suspected active ingredient) listed which were the primary interest of our study. Due to reporting inconsistencies, we couldn't assume that the active ingredients were listed in order of significance or suspicion. For this reason, we did not exclude cases with multiple suspected active ingredients. Other exclusion criteria included cases in which no suspected drug or active ingredients were identified or in cases with pre-existing optic neuropathy that was listed as the indication for drug administration. After accounting for all the inclusion and exclusion criteria detailed above, a total of 128 cases were excluded from the original 819 cases.

### *Classification of Drugs*

Information regarding mitochondrial toxicity was sourced from a variety of online databases and journals including pubmed.gov, mitotox.org and google scholar. The drugs of interest were first searched for in the mitotox.org database, then subsequent relevant databases and finally google scholar to ensure any evidence of mitochondrial toxicity was discovered and properly evaluated. Drugs were determined to have mitochondrial toxicity if they possessed the ability to alter major mitochondrial pathways and functions: Increase cellular production of reactive oxygen species and cause oxidative stress, inhibit respiratory complexes of the electron transport chain, induce mitochondrial membrane permeability transition pore, inhibit mitochondrial fatty acid oxidation, cause mitochondrial DNA damage and impair oxidative phosphorylation by causing a decrease or uncoupling (Rana *et al*., 2021). Drugs that were suspected to have mitochondrially toxic mechanisms but did not have a specific mechanism outlined in the database were classified as not having a mechanism of mitochondrial toxicity.

### *Statistical Analysis*

Drugs were coded as numbers and then SPSS software was used to analyze the number of times a drug was implicated as a potential cause of optic neuropathy in each case. Initially, 253 drugs were identified to be associated with these cases. Any drug appearing to be implicated in 5 or more cases was chosen as a drug of interest, with 5 being the cut-off determined to establish more confidence in a drug's potential connection to optic neuropathy and to account for drugs reported in FAERS that may have been implicated in error. This procedure identified 60 drugs. After we identified if these drugs were linked or not with mitochondrial toxicity mechanisms, subsequent searches were performed for adverse events other than optic neuropathy for all 60 drugs. The drug names were individually searched in the FAERS database allowing the capture of all cases of adverse drug events related to each of the 60 drugs. There were two instances in which the same drug was listed twice (e.g., "amiodarone" and "amiodarone hydrochloride") and these were combined into one line of data as not every case reported to FAERS specified the exact formulary of each drug. The same exclusion criteria were applied to the individual drug searches as to the original data set for optic neuropathy/toxic optic neuropathy. Then all cases that mentioned optic neuropathy or toxic optic neuropathy were excluded. This resulted in all adverse drug events other than optic neuropathy for each drug. This method yielded several cases of adverse drug events for each drug that had outcomes of optic neuropathy and outcomes other than optic neuropathy. For statistical analysis of the results, a reporting odds ratio was calculated as displayed in Table 1 to compare the likelihood of the occurrence of optic neuropathy and other adverse events between each drug class.

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## Results

The dataset included 373 drugs. Most suspected active ingredients occurred at a low frequency (n ≤4) in the database. However, there were 60 drugs that occurred at a higher frequency (n >5). This list of drugs was analyzed for mitochondrial or non-mitochondrial mechanisms of action based on existing literature and they were then categorized as drugs "with" and "without" a known mitochondrial toxicity mechanism. Once separated into the Reporting Odds Ratio (ROR) value was calculated and is shown in Table 1. The ROR was calculated as 1.4762 (95% CI 1.3022-1.6735, p<0.0001). Table 1 shows a high statistical significance with a large odds ratio for drugs with known mitochondrial toxicity implicated in optic neuropathy. This indicates that drugs possessing a known mechanism of mitochondrial toxicity were 47.6% more likely to be implicated in cases of optic neuropathy than drugs without known mitochondrial toxicity. Additionally, the drugs that do have documented mechanisms of mitochondrial toxicity comprise significantly higher percentages of the cases of adverse drug events for each drug than drugs without mechanisms of mitochondrial toxicity when comparing the number of optic neuropathy cases to the total number of ADEs for each drug. Drugs that do not have documented mechanisms of mitochondrial toxicity were significantly less often implicated in cases of optic neuropathy than drugs with such mechanisms (Tables 2-3).

Figure 1 shows the drugs from Table 2 organized by their type. Figure 2 shows the drugs from Table 3 organized by their type. In both analyses, antimicrobials appeared much more frequently than other drug types, followed by antineoplastics and immunosuppressants appearing in both lists. There were also drug types that only appeared in one list, such as corticosteroids, phosphodiesterase inhibitors and monoclonal antibodies only appeared in a list of drugs without known mitochondrial toxicity (Fig. 2).

**Table 1:** Reporting odds ratio of drugs with and without known mitochondrial toxicity compared to the number of reported adverse drug events with and without optic neuropathy

| | Drugs implicated in cases of optic neuropathy | Drugs implicated in ADEs other than optic neuropathy |
|---|---|---|
| Drugs with known mitochondrial toxicity mechanism | 560 | 442949 |
| Drugs without known mitochondrial toxicity mechanism | 434 | 506772 |
| ROR Equation | $\frac{\left(\frac{560}{442949}\right)}{\left(\frac{434}{506772}\right)} = 1.4762$ | ROR = 1.4762<br>CI = 1.3-1.7<br>Z = 60.1<br>p<0.0001 |

**Table 2:** Drugs with known mitochondrial toxicity and the frequency of optic neuropathy cases

| Drug name | Total ADEs reported | Number of ADEs with optic neuropathy |
|---|---|---|
| Linezolid | 8743 | 172 |
| Tacrolimus | 42012 | 95 |
| Levofloxacin | 13056 | 27 |
| Rifampin | 6355 | 23 |
| Isoniazid | 3434 | 19 |
| Amiodarone | 13321 | 18 |
| Clofazimine | 2614 | 17 |
| Moxifloxacin | 2864 | 15 |
| Methotrexate | 65641 | 14 |
| Cyclophosphamide | 54040 | 14 |
| Mycophenolate mofetil | 36903 | 14 |
| Bedaquiline | 2201 | 13 |
| Lamivudine | 2832 | 12 |
| Carboplatin | 33618 | 10 |
| Levetiracetam | 23917 | 10 |
| Oxaliplatin | 24913 | 9 |
| Docetaxel | 16776 | 9 |
| Azithromycin | 2898 | 9 |
| Clarithromycin | 7148 | 8 |
| Pyrimethamine | 283 | 8 |
| Paclitaxel | 31427 | 7 |
| Omeprazole | 15222 | 7 |
| Levothyroxine | 4399 | 7 |
| Sirolimus | 4309 | 7 |
| Itraconazole | 1852 | 6 |
| Etoposide | 22617 | 5 |
| Pentamidine isethionate | 114 | 5 |

*Adverse drug events included occurred between 1/1/2018-10/8/2022

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**Table 3:** Drugs without known mitochondrial toxicity and the frequency of optic neuropathy cases

| Drug name | Total ADEs reported | Number of ADEs without optic neuropathy |
|---|---|---|
| Ethambutol hydrochloride | 2649 | 86 |
| Voriconazole | 6571 | 53 |
| Pyrazinamide | 2716 | 27 |
| Prednisolone | 34134 | 25 |
| Adalimumab | 53666 | 17 |
| Methylprednisolone | 27172 | 16 |
| Metronidazole | 8167 | 16 |
| Sulfamethoxazole-trimethoprim | 13322 | 14 |
| Prednisone | 52964 | 13 |
| Pembrolizumab | 27349 | 10 |
| Alprazolam | 16972 | 9 |
| Fingolimod hydrochloride | 11882 | 9 |
| Leucovorin calcium | 4625 | 9 |
| Dolutegravir | 2798 | 9 |
| Delamanid | 1260 | 9 |
| Amikacin | 2754 | 8 |
| Atovaquone | 762 | 8 |
| Onabotulinumtoxina | 17337 | 6 |
| Hydroxychloroquine | 14127 | 6 |
| Aminosalicylic acid | 745 | 6 |
| Rituximab | 61846 | 5 |
| Etanercept | 50312 | 5 |
| Trastuzumab | 16635 | 5 |
| Ipilimumab | 15857 | 5 |
| Ramipril | 14365 | 5 |
| Melphalan | 6933 | 5 |
| Ribavirin | 5831 | 5 |
| Tadalafil | 5171 | 5 |
| Mesalamine | 4976 | 5 |
| Sildenafil | 4281 | 5 |
| Pemetrexed disodium | 3644 | 5 |
| Clofazimine | 2602 | 5 |
| Mesna | 1967 | 5 |

*Adverse drug events included occurred between 1/1/2018-10/8/2022

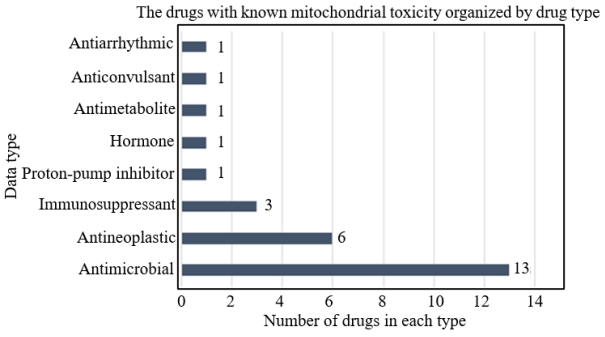


**Fig. 1:** The drugs with known mitochondrial toxicity listed in Table 2 were classified by their type and the frequency of each type is noted in this figure. Drugs that can be classified as a type of antimicrobial were by far the most common type with 13 out of 27. Antineoplastic and immunosuppressant drugs were the second and third most commonly appearing drug type

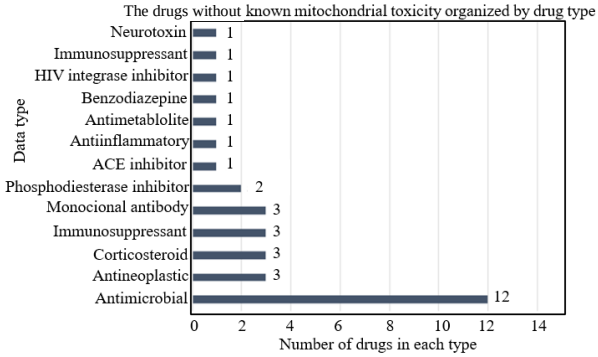


**Fig. 2:** The drugs without known mitochondrial toxicity that are listed in Table 3 were classified by their type and the frequency of each type is noted in this figure. Drugs that can be classified as a type of antimicrobial were by far the most common type with 12 out of 33. Antineoplastic, corticosteroid, immunosuppressant and monoclonal antibody drugs were the next most common drug types appearing on the list

## Discussion

The findings of this study indicate that drugs possessing a mechanism of mitochondrial toxicity are 47.6% more likely than drugs without mitochondrial toxicity to be implicated in cases of optic neuropathy (Table 1). This is important because this conclusion is based on the occurrence of patients having adverse reactions to these drugs, even though most of them already have a warning specifying optic neuropathy or neuritis in the FDA data sheet of the drug. This indicates that either the information is missing, doesn't make it to the prescriber and consumer, or is ignored. Furthermore, the significance of this relationship is important because there were drugs identified that have not been proven to be mitochondrially toxic that were also identified in the database as significantly correlated with cases of optic neuropathy highlighting the utility of this analysis. There were several drug categories that appeared to be more highly correlated with optic neuropathy and there were several categories that overlapped between the lists of drugs with and without known mitochondrial toxicity. This raises suspicion of the potential for a mitochondrial toxicity mechanism to exist and is currently unknown. It also indicates that new drugs in certain drug categories or types may need more testing or surveillance than others to determine patient risk for mitochondrial toxicity. This also emphasizes the importance of this type of analysis.

It was interesting to note how the same drug types appeared frequently in both lists (drugs with and without known mitochondrial toxicity), which could indicate an unknown link between the two, such as metabolic or genetic similarities between mitochondrial bacterial ancestors and humans or point mutations and genetic anomalies in humans that could lead to unknown off-target effects of some of these drugs. However, these would be extremely challenging to identify in any pre-market toxicity analysis. Unfortunately, in most of these cases, enough data is not gathered to draw conclusions as to what is causing optic neuropathy in patients using non-mitochondrially toxic drugs. It is possible that there are unknown human mitochondrial variants, or variants that have unknown susceptibility to these drugs. This information would be crucial to put into practice when prescribing to individuals with confirmed mitochondrial disorders, populations with decreased mitochondrial function, or those with familial mitochondrial disorders that may carry sub-threshold levels of heteroplasmic mitochondrial DNA mutations. It would be prudent to require more detailed reporting in FAERS, as well as to perform more *in vivo* analysis of these drugs to find out how they are causing symptoms so specific for mitochondrial toxicity such as optic neuropathy or hepatotoxicity.

For example, Atovaquone is a medication that was implicated frequently as a cause of optic neuropathy. This drug doesn't have a documented mechanism of mitochondrial toxicity in humans, but Table 3 shows a very high number of adverse outcomes listed as optic neuropathy. Atovaquone is an antimalarial and anti-parasitic drug that acts to kill through selective toxicity to parasite mitochondria but not their hosts (Srivastava *et al*., 1999). However, there have been single amino acid mutations in malarial mitochondrial DNA altering binding sites of atovaquone to cytochrome b conferring resistance to atovaquone treatment (Srivastava *et al*., 1999). Alteration of this binding site could prevent toxicity and lead to resistance, or it could allow off-target binding and lead to destruction of a mitochondrion or cell in a host in which it was not meant to. It is therefore very possible that there could be mutations in human mitochondrial DNA allowing it to be mitochondrially toxic in a similar manner.

Ethambutol, a medication used to treat tuberculosis, is known to cause optic neuropathy in a small subset of patients. Ethambutol is thought to disrupt mycobacterial cell wall formation, though its mechanism of action is not well known (Pawar *et al*., 2019). Further analyses led to the discovery of mitochondrial mutations in genes involved in oxidative phosphorylation pathways of mitochondria and mitochondrial wall integrity as potential causes of susceptibility to optic neuropathy in patients that receive ethambutol therapy (Kaur *et al*., 2022; Guillet *et al*., 2010). With mitochondria making the most substantial contribution to ATP production, it's easy to understand how alteration of this process in mitochondria leads to acute symptoms in structures highly dependent on energy such as optic nerve or the heart. Guillet *et al*. (2010) used fibroblasts collected from a patient who developed optic neuropathy following ethambutol administration and found that, compared to control cells, the patient carried a mutation in the OPA1 gene. This gene codes for a protein that resides in the mitochondrial cell membrane and plays a role in cristae formation, which are infoldings of the mitochondrial inner membrane to enhance the production of ATP to feed the entire cell. Additionally, this study found that ethambutol administration to the patient-derived cell line caused a decline in complex IV activity, decreased mitochondrial membrane potential and increased mitochondrial network fragmentation (Guillet *et al*., 2010), all of which could indicate metabolically stressed mitochondria. Dolutegravir, an integrase inhibitor used in antiretroviral treatment in HIV patients has been shown to decrease expression of the uncoupling protein 1, a decrease in mitochondrial complex IV and a reduction in mitochondrial respiratory capacity once again indicating mitochondrial toxicity and leading to metabolically stressed cells (Jung *et al*., 2022). Although the authors suggest that mitochondrial dysfunction leads to decreased energy expenditure and weight gain as major side effects of integrase inhibitors, these results indicate that more research is needed to understand the full implications this drug can have on mitochondria (Jung *et al*., 2022). These

findings indicate a need to further investigate drugs that are known to cause mitochondrial dysfunction, in order to identify the causal mechanism, but also indicate that postmarket analyses such as ours are crucial to identify these drugs with unknown mechanisms.

Examining mitochondrial off-target effects, mitochondrial mutations and conditions that can cause mitochondrial dysfunction is part of the solution to preventing adverse drug reactions. Determining other drugs or supplements that can be used in conjunction with high-risk drugs to mitigate negative effects is just as important. One such study found that mitoquinone, a derivative of ubiquinone, reduced amikacin ototoxicity in guinea pigs (Dirain *et al*., 2018). Amikacin, an aminoglycoside antibiotic, inhibits protein synthesis by binding to the 30 s ribosomal subunit and does not have any known mitochondrial mechanism of action. Although animal studies do not always translate to human physiology, this finding suggests that supplementation with mitochondrial-specific compounds can help prevent adverse reactions in mitochondria.

These examples demonstrate a need to further investigate biological and genetic differences between patients that predispose them to poor drug outcomes. These biomarkers and genetic variants, if present, could inform and aid physicians in prescribing alternative therapies. Likewise, investigating concomitant therapies with supplements or other drugs that may lower the risk of mitochondrial harm in susceptible patients would lower the chances of detrimental drug outcomes and improve patient safety and quality of life. All of this drives the point that more research is needed to understand drug mechanisms, especially pertaining to mitochondrial toxicity and the effects these drugs may have on patients who have vulnerable mitochondrial health. Postmarket analyses such as this that start with adverse drug events with specific symptoms related to pathologic mechanisms are reactive rather than proactive, but due to numerous unknown factors, maybe the only way to identify dangerous drugs and eventually their mechanisms of toxicity. As of the time of this analysis, there is a lack of similar research methods being conducted to aid in the identification of dangerous drugs with unknown mechanisms of toxicity.

## Conclusion

Mitochondria are especially vulnerable to drug toxicity and the resultant side effects of mitochondrial derangement can have serious effects on patients. Mitochondrial toxicity is under-analyzed in drug development despite its potential to have severe, long-term and irreversible consequences for patients (Vuda and Kamath, 2016). Drug classes that are known to cause mitochondrial toxicity should be considered for boxed warnings issued by the FDA, as many drug classes with less severe adverse events already have specific warnings (Delong and Preuss, 2023). While some medications known to cause mitochondrial toxicity can be a patient's only option or last resort in treating their health conditions that does not absolve the need for better understanding and warning of a drug's or drug classes' potential for toxicity. Mitochondrial toxicity induced by drugs and many of the mechanisms behind that toxicity are well-studied at this point (Vuda and Kamath, 2016), but there is a gap in the literature in assessing and stratifying the risk of new drugs that have the potential to cause mitochondrial toxicity. Based on this study's analysis, we provide evidence that mitochondrial toxicity can be assessed by analyzing adverse drug events based on symptoms in the larger population provided by FAERS. We recommend that, at a bare minimum, this kind of analysis should be considered part of post-marketing surveillance so that risk can be better assessed in prescribing these drugs. Patients also need to be better informed of these risks. Ultimately, mitochondrial toxicity risk should be more preemptive than assessing adverse events after patients have already used the drug and this study supports the need to standardize the assessment of mitochondrial toxicity during the drug's initial development and clinical trials.

### *Limitations and Strengths*

The FDA Adverse Event Reporting System (FAERS) is a public database that contains adverse drug events and medication error reports that can be submitted to the FDA by almost anyone. It is designed as a post-marketing surveillance program for drugs and therapeutics. These events are linked to specific reports that may or may not be associated with a clinical trial. Strengths of FAERS include that it is a standardized reporting system, is user-friendly and can capture events within 15 calendar days of the manufacturer receiving notice of the event. However, some pitfalls are that some, although few, reports are incomplete and many have missing information. FAERS does not include all drug and ADE information, such as how many patients have taken the drug, patients' medical history and all other drugs the patient was taking, so incidence rates and causal relationships between drugs and toxic effects are hard to determine. Further studies will need to be completed to determine the true causality of drug-mediated mitochondrial effects and toxicity.

In order to strengthen our analysis, we excluded adverse events reported exclusively by consumers. Due to a lack of medical training leading to increased inaccuracy and inconsistency in reporting, we focused on ADEs reported only by healthcare professionals. Although the consumer-reported events should have been reviewed by nurses and other medical professionals prior to

submission to FAERS, we still felt that removing these cases was necessary given the large volume of cases and potential inconsistencies. We recommend that FAERS update their reporting standards in an attempt to gain more information including demographics that may be useful to identify at-risk populations. Although we understand that requiring certain information to file a report of an ADE may hinder reporting, if FAERS could provide motivation to make more complete reports, include more data and ensure reports came from reliable sources, then the accuracy of determining exactly why ADEs happen will be improved and perhaps it would be easier to determine exact off-target drug effects.

### *Future Directions*

In the future, it is pertinent to continue to monitor the potential mitochondrial toxicity of newly released drugs. Analysis of post-marketing adverse effects of drugs should be continuous and include analyses of demographic data to determine populations at risk. In this study, we analyzed optic neuropathy, but other symptoms that may be related to mitochondrial toxicity such as cardiotoxicity, myopathy and other neuropathies should be considered. Additionally, to strengthen our findings, the next step would be to analyze the mitochondrial toxicity mechanisms of high-risk drugs. These drugs should be tested in healthy cells to elucidate any potential mitochondrial mechanisms but would also ideally be tested in mitochondrial disease patient-derived cells, or any mitochondrial disease model to identify effects of toxicity. These findings would determine any changes in dosage that would be pertinent to high-risk patients. Additionally, oral supplementation of l-carnitine, alpha-lipoic acid, coenzyme Q10, reduced Nicotinamide Adenine Dinucleotide (NADH), membrane phospholipids, vitamins C, D, E, B1, B2 and others have been used in the treatment of mitochondrial diseases to reduce mitochondrial dysfunction (Nicolson, 2014; Mantle and Hargreaves, 2022). These supplements may be protective if a high-risk drug is required to be taken by a high-risk patient. The testing of these combinations could help identify potential prophylactic protocols for patients at risk of developing mitochondrial dysfunction pathologies in response to taking these high-risk drugs. Overall, works such as this should be carried out periodically and encompass all ADEs with symptoms highly likely to be due to specific pathology in an effort to identify novel mechanisms of toxicity that have yet to be discovered.

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## Author's Contributions

**Connor Dyer:** Conceived the study idea, co-wrote the research proposal, conducted half of the literature search and review, drafted the submitted article, submitted the article to the journal, and made significant edits to the article based on recommended peer-reviewed.

**Katlyn Droke:** Contributed to drafting the proposal, reviewed the literature search findings provided by the other authors, drafted the submitted article, created the tables and figures, and made significant edits to the article based on the recommended peer-reviewed revisions.

**Ugne Zekonyte:** Conceived the study idea, co-wrote the proposal, conducted half of the literature search and review, and co-wrote the draft of the article submitted to the journal. Ugne also provided expertise in mitochondrial genetics.

**Mariana Rosca:** Reviewed the research proposal, provided mentorship during the process of analyzing results, and provided significant edits to the draft of the article. Dr. Rosca also provided subject matter expertise and extensive research experience in mitochondrial diseases.

## Ethics

This study is a database analysis using the FAERS database, which is publicly available and does not contain personally identifiable data. Ethical considerations of data collection include anonymity of participants and avoidance of biased results by variation of search terms as explained in the previous section and exclusion of unverifiable information. Furthermore, case submissions are voluntary and exclusion criteria were carefully defined to avoid bias in data collection as detailed earlier in this article. Therefore, no review board approval was required for this study. The authors have no conflicts of interest.

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