

## WHAT ALKAPTONURIA PATIENTS NEED TO KNOW ABOUT NITISINONE (ORFADIN®, NTBC)

### Introduction

This article is designed to give people suffering from Alkaptonuria (AKU) a patient's viewpoint on what Nitisinone is, why it is a possible treatment for AKU and what is known about the effects from using it over a long period.

I've referenced some websites in this article. Please note I have no control over the contents of these sites, neither do I have any connection, either commercial or otherwise, with them and therefore neither endorse nor recommend them (apart from the website for the AKU Society, of which I am a member).

### What is Alkaptonuria?

Alkaptonuria (AKU) is a rare autosomal recessive metabolic disorder with fewer than 1,000 patients currently identified worldwide. It is caused by a faulty enzyme, homogentisic acid 1,2-dioxygenase (HGO), in the metabolic pathway of tyrosine, an essential amino acid. As a consequence, there is an excess of homogentisic acid (HGA) in the body, the majority of which gets excreted in the urine, causing it to turn black if left for a time. However, the remaining HGA embeds itself in various soft tissues within the body, especially cartilage. This leads in particular to problems in the spine and major joints with consequent pain and loss of mobility as the cartilage is degraded and ultimately destroyed. Its effects are similar to arthritis, although AKU has further complications: it can affect two of the heart valves (the aortic and the mitral); it can increase the likelihood of painful stones (kidney and prostate); and muscles and ligaments can be affected. There is no known cure for AKU and current treatment relies heavily on pain management and invasive surgery.

There are many web sites that will give you an in-depth perspective on AKU. They may be found by clicking on the following links:

<http://www.alkaptonuria.info>

<http://www.ncbi.nlm.nih.gov/pubmedhealth/PMH0002180>

<http://en.wikipedia.org/wiki/Alkaptonuria>

### What is Nitisinone?

Nitisinone blocks an enzyme called 4-hydroxyphenylpyruvate dioxygenase (HPPD), which is found both in animals and plants. In plants it affects chlorophyll production in the leaves and as a consequence, Nitisinone and other related compounds are used as herbicides. In 1992, in a big leap of faith, a very sick child in Sweden was administered Nitisinone to treat Hereditary Tyrosinaemia Type 1 (HT-1), and hence a treatment for this condition was born. Nitisinone is now licensed to be prescribed for the treatment of HT-1 and, once again, there are many websites that will tell you all you need to know

about Nitisinone. As a sample I've added three links here for reference. Please remember the disclaimer outlined above.

<http://www.rxlist.com/orfadin-drug.htm#drugs>

<http://www.flexyx.com/O/Orfadin.html>

<http://en.wikipedia.org/wiki/Nitisinone>

### **Why is Nitisinone a potential treatment for Alkaptonuria?**

Imagine a factory where cars are built by robots on a conveyer belt, and the robot responsible for putting the engine into the car breaks down. The whole process has to stop and there is a build up of cars without engines. The next robot down the line cannot put the wheels on until there is an engine present. This simplistic analogy is how many of the processes in the body work.

For Alkaptonuria and other related conditions, the process affected is a metabolic pathway called the Tyrosine catabolism. Tyrosine is an amino acid required for life: it is consumed as dietary protein, and excess tyrosine is removed from the human body by this pathway. Tyrosine catabolism is automatic, and consists of a series of enzymes that systematically degrade tyrosine to non-toxic metabolites that can be easily excreted. Like the conveyer belt, the pathway is very specific and interdependent. Any faults in the enzymes in this pathway (mutations) can lead to a variety of medical conditions.

The medical condition is dependent on which enzyme is at fault and is usually described by the excess of metabolite produced. In the car analogy, if the robot putting in the engines stopped, then there would be a lot of cars without engines; if it was the robot that puts on the brakes, then there would be a lot of cars without brakes. Either way, you don't get a working car at the end of it. In HT-1 the condition is caused by the mutation of the fumarylacetoacetate hydrolase (FAH) enzyme, leading to the build up of toxic metabolites causing acute liver disease in children that, if untreated, leads to death.

One way of stopping the build up of these toxins is to stop one of the enzymes further up the chain. In the car analogy, it might be safer and easier to stop the production line at an earlier stage so that you don't get a build up of cars without engines. This is how Nitisinone works in the Tyrosine catabolism. It blocks an enzyme further up the chain called 4-hydroxyphenylpyruvate dioxygenase (HPPD), which stops the degradation process before the body can create the toxins seen in HT-1.

Then why do we think Nitisinone is a potential treatment for AKU? Crucially, the enzyme HPPD appears in the tyrosine degradation pathway before the enzyme mutation that causes AKU and as such, should reduce the levels of HGA produced, resulting in either a reduction or elimination of the onset of AKU related complications.

### **Why isn't Nitisinone currently available for AKU?**

Like all regulated medical products, there is a well-defined process to prove the efficacy and safety of any new drug. In the case of Nitisinone, the safety profile has been established, allowing it to be sold as a medicine, but its efficacy in treating AKU has still to be statistically proven.

A formal three year trial, run by a team in the USA and funded by the National Institute of Health (NIH), was completed in 2009. This trial was carried out on 40 AKU patients, 20 taking Nitisinone, 20 taking a placebo, and the clinicians tested a series of movements on the patient's joints before, during and at the end of the trial. Although there was some improvement, it was not statistically significant. This does not mean that Nitisinone doesn't work, just that there is more work to prove that it does work. The USA trial highlighted that the creation of a definitive 'test' that was a true measure of the medical condition under review is key. Moreover, this is not a simple task for complex conditions such as AKU. This is compounded by the fact that the standard route to increasing statistical significance of a clinical trial is to use a larger patient group, something not available to the AKU community.

### **Potential Long-Term Side Effects**

Most of the toxicology and safety data available relates to human trials for HT-1. In these patients the average dose is 1 milligram of drug per kilogram of bodyweight per day (1mg/kg/day), and can be raised to 2mg/kg/day if the patient does not respond to the lower dose. The known side effects are reported through the links in the "What is Nitisinone?" section above. In particular, there is a significant incidence of eye problems, which is controlled by maintaining a low tyrosine diet. The treatment regime is designed to maintain the levels of tyrosine seen in the blood plasma under 500  $\mu$ M.

More recently, there have been a couple of reports of children with HT-1 who have been on Nitisinone since infancy. A few of these children have shown developmental and behavioural problems when they reached school age. The authors hypothesise that it may be related to the elevated levels of tyrosine. However, the authors do admit that the children in question may not have followed the strict medication and diet regime required for HT-1 and as a consequence the levels of tyrosine are exceptionally high. Alternatively, these behavioural issues may be due to HT-1 itself, but have never been seen before because most patients did not survive long enough without Nitisinone treatment. The investigators do point out that there is not enough evidence to definitely prove the cause of these changes, and that further studies need to be undertaken to clarify the situation.

### **Are there any differences seen between HT-1 and AKU patients taking Nitisinone?**

In theory, as Nitisinone blocks HPPD, then there should be many similarities with the treatment of these two conditions. However, the average dose for HT-1 patients is 1mg/kg/day while for AKU it is 2mg per patient (0.05-0.01mg/kg/day). There has been a report that some patients with HT-1 are successfully treated (the presence of the toxic metabolite was not detected in blood samples) with doses of 0.5-0.65mg/kg/day, whilst an AKU patient taking 0.01mg/kg/day showed undetectable amounts of HGA in their urine. Why the metabolites from the two conditions are controlled with such extremes of

doses is unclear. However, with the lower doses used with AKU patients the amount of tyrosine in the blood plasma is reduced to such an extent that in the NIH trial no dietary modification was deemed necessary. Further dose ranging studies may be appropriate to balance the levels of HGA and tyrosine in blood plasma for AKU patients.

### **What further clinical trials could be run to show that Nitisinone is effective in treating AKU?**

Like most things in life, taking time to analyse what you are trying to achieve before you start can save you a lot of time, money and frustration in the future. This is particularly important when designing a clinical trial, which is why drug companies spend a lot of money ensuring they get it right before they start. For a clinical trial for any treatment of AKU there are several issues we need to think about.

#### **Option A: Traditional Clinical Trial**

What do I mean by a traditional clinical trial? Standard practice when running a clinical trial (to give its full title, Phase III Clinical Trial) looking at the efficacy of a new drug is to have an observable outcome. For example, a reduction in blood pressure for those testing a new anti-hypertensive. For more complex medical conditions where a direct cause and effect **cannot** be measured, an internationally agreed scoring system that looks at all the symptoms of the condition is used. For example, clinical trials for rheumatoid arthritis (**RA**) have for many years used the ACR (short for American College of Rheumatology) scoring system, which measures the overall changes in a persons' RA rather than just in the hands, feet or other joints. Scoring systems have the added benefit of minimizing the subjective nature of patients' feedback, especially for conditions involving chronic pain, as everyone's pain threshold is different.

Over the past three years clinicians involved in AKU Research have been developing an AKU Severity Score Index (AKUSSI), which has now been agreed and validated. It is this scoring system that could be used for any further clinical trials.

#### **How long will the clinical trial need to run?**

In many clinical trials, there is often an observable effect after a short period of time and these outcomes can be readily measured. Even in the original treatment of HT-1 with Nitisinone, there was a marked improvement in the condition of the patients after only a few days or weeks. Unfortunately, there are many conditions, such as AKU, where there is not a quick response, and it takes many years to conclusively show that the treatment is working, even using scoring systems such as the AKUSSI. As the effects of AKU take such a long time to be recognized in a patient, it has been suggested that a 5-10 year timeframe to run a clinical trial, with further regular monitoring for the rest of the patients life, would be necessary to prove definitively that Nitisinone works. Not only would it be expensive to run such a trial, but the regulatory authorities would need to be convinced of a high chance of a positive outcome before allowing it. There is also an ethical argument that those patients that are not on the clinical trial may be missing out on the only available treatment. By the time Nitisinone is approved for AKU the non-participants may have developed the joint problems associated with AKU.

## **Option B: Designing a Clinical Trial with a Surrogate Endpoint**

The first question that should be asked is “What is a Surrogate Endpoint?” A surrogate endpoint is defined by the NIH as “a biomarker intended to substitute for a clinical endpoint”. What does this mean in practice? Normally, as described above, clinical trials are looking for a benefit in alleviating the symptoms of a medical condition. Regulatory authorities will allow surrogate endpoints when the primary endpoint has to be avoided at all costs, e.g. death, or when the patient population is so small that it will be impossible to get a statistically significant outcome. For example, high levels of cholesterol can lead to increased risk of heart failure and that taking statins reduced the risk. It is not the statin that is reducing the incidence of heart failure, but it is medically accepted that reducing the levels of cholesterol is the beneficial effect, i.e. the level of cholesterol is the surrogate endpoint.

The regulatory authorities in the USA and EU have identified that rare diseases, by their very nature, are difficult to design clinical trials for. Hence they are willing to consider surrogate endpoints, as long as the link between the endpoint and beneficial effects on the medical condition are proved. For AKU, it has long been known that the increased levels of HGA are caused by the faulty enzyme and that the deposition of HGA leads to the degradation of soft tissue. More recently there have been several reports that show that Nitisinone reduces the levels of HGA in both urine and blood plasma by more than 95% compared to before treatment started. The difficulty lies in getting the regulatory authorities to accept that the reduction in HGA, the surrogate endpoint, reduces the debilitating effects of AKU. If the authorities do accept that this argument is valid, then the design of the clinical trial would be a lot simpler, would be quicker to complete and have a better chance of success. Research is already underway to validate this link.

The current thinking is to use some sort of combination approach that combines the HGA surrogate endpoint and AKUSSI to monitor patient outcome.

Who should be included on the trial?

For many medical conditions there is a large patient population for the clinicians to choose from. This is not true for rare diseases like AKU even with the excellent work of the AKU Society over the past several years in contacting AKU patients worldwide. For a successful clinical trial, there is a balance between having as few patients as possible on a trial, to make it manageable and reduce costs, and having sufficient numbers to allow some to drop out of the trial and still have enough data to statistically prove that the treatment works. The higher the numbers, the more accurate the statistics.

The other major consideration with any treatment for AKU is the nature of the condition itself and the requirements of individual patients. For those that already have advanced symptoms, the treatment is unlikely to repair existing tissue damage and any existing damage is likely to increase the likelihood of further complications. For example, if the patient already has a damaged hip, then the ongoing wear and tear caused by this

damage is likely to enhance the need for early joint replacement. Trying to measure any increase in damage over the existing tissue damage is therefore very difficult to prove.

Conversely, other than vaccinations, it is unusual to treat 'well' people with drugs to prevent onset of a condition before it shows any symptoms, with the cost of drugs and potential side-effects from long term use being quoted as reasons for not starting early. Perversely, if a patient is treated early and does not become symptomatic, there are those who will argue that the patient would not have developed the debilitating effects of the condition anyway.

Running a Clinical Trial on younger adult patients would appear to be the obvious solution, but this in itself creates its own ethical dilemma. As mentioned above, the length of any clinical trial would have to be run over a very long period. If the trial is very successful, then those on placebo would have started to show the effects of AKU, which would leave them in a similar situation to older patients. Maybe the solution is to allow any young adults with AKU who wants to take Nitisinone to do so, with regular monitoring over an extended period. Comparison with those that decided not to take the medication and the older generation should provide the required data. Getting regulatory approval and health care establishments to finance the cost of the medication and regular monitoring would be a great hurdle to overcome.

As a footnote to this section, there are a significant number of patients that are not diagnosed as having AKU until later in life, often after seeking medical advice because of excessive joint pain. The diagnoses for these patients are very hit and miss. One recent example involved a clinician, suspecting a patient had AKU, leaving a sample of the patients urine on the window sill and waiting to see if it turned black, rather than bothering the pathology laboratory – very simple but effective. At the other extreme, there have been scientific papers reporting on a patient undergoing keyhole surgery on their knees and ochronosis being found. Good to know, but not really a simple tool for diagnosis.

If it is established that taking Nitisinone before symptoms start appearing is beneficial in treating AKU, then those patients diagnosed later in life would not be able to get the full benefits from Nitisinone. There is clearly an unmet need to establish a simple diagnostic test for use in infancy to determine whether someone has AKU or not. Similarly, a diagnostic test should be available to diagnose AKU in older patients and crucially, whether those patients are starting to develop damage or not. The earlier the diagnosis, the better chance of treating the condition.

### **Is anyone looking at using Nitisinone for treating any other conditions?**

There has been a report that lab based tests indicate that Nitisinone may have some clinical value in treating Hawkinsonuria, another disease of the Tyrosine catabolic pathway. Interestingly, there is a clinical trial that is currently in progress that is looking at using Nitisinone as a potential treatment for Parkinson's disease. The logic behind this is that tyrosine is a precursor to dopamine, the chemical stimulus in the brain that is missing in Parkinson's patients. Laboratory tests have shown that increases in tyrosine

levels increase the levels of dopamine and hence alleviate the symptoms of Parkinson's. It is not clear what dosing regime these investigators are using, but it is something to keep an eye open for.

### **Personal Experience with Taking Nitisinone.**

I have been taking Nitisinone since October 2008. Originally I started on a 1mg per day dose, but due to supply issues the dose was increased to the more readily available 2 mg capsule. Whilst on the 1mg/day dose, the HGA in my urine was below the detectable limits of the analytical technique used (Nuclear Magnetic Resonance Spectroscopy, NMR). No measurements of plasma tyrosine were taken. Being a substantial man, I weigh approximately 100kg, the effective dose I was taking at this time was 0.01mg/kg/day, which equates to 1% of the dose taken by an HT-1 patient.

I have not noticed any adverse effects from taking Nitisinone. The question I often get asked is "Is it working?" and the truthful answer is "I don't know". By the time I started taking Nitisinone I was already suffering many of the effects of AKU, back and joint problems. I had already had three arthroscopies on my left knee and have since had a total left knee replacement. Recent MRI scans on my hips show that I have extensive degradation of the cartilage in both hips. My view is that because of the existing damage to my joints, Nitisinone will not help to combat the existing damage, but it will still help by either preventing or slowing down the rate at which further damage may occur. For example, at present I do not appear to have any of the complications associated with cardiac valves. Taking Nitisinone now will hopefully prevent complications in the future. I will have to live with the existing damage to my joints, and the increased wear and tear the existing damage will cause to these joints.

### **In summary**

AKU was first called an inborn error of metabolism by Garrod nearly eleven decades ago. For the first ten decades there was very little research done into AKU and a lot of misinformation was generated in that time. For example, one biochemistry text book from the 1980s, on describing AKU, states that "...more people have worried themselves sick over AKU than from the condition itself..."

It is only in the last decade that the understanding and treatment of AKU has finally been recognized as an unmet clinical need. In this last decade a lot of progress has been made. The formation of the AKU Society in 2003 along with sister organisations in France, North America and Italy has helped to provide support to patients around the world. More patients have been identified in this past 10 years than ever before. As a result, formal clinical studies have been performed to identify when patients start to be affected by AKU, identifying all the parts of the body that can be affected and how degeneration of soft tissue occurs with age. Formal research into AKU has been carried out and is still ongoing and scientific conferences organised to discuss the latest findings. A potential drug treatment has been identified and an initial clinical trial performed. There is a definite plan to get an effective treatment available for all as soon as possible and the long-term goal is to find a cure. For this momentum to continue there are several key areas that need continuing support:

- 1) There are still a lot of patients that are not diagnosed with AKU until later in life. The consensus is that any treatment will be most effective by administering earlier in life, so early detection is vital.
- 2) Maintaining the level of interest in AKU is essential. It is human nature to become bored with a subject over time. Unfortunately, AKU will not go away just because we are bored with it.
- 3) Finally, as with most things, money has to come into it. Much of the work already carried out has been funded by grants and donations. For the work to continue the levels of funding need to increase. Clinical trials do not come cheap.

### **About the Author**

Duncan Batty gained his PhD in Synthetic Organic Chemistry in 1992 from University College London. He spent two years performing academic research in both Chicago and Paris followed by five years working at a government research institute investigating the isolation and synthesis of novel natural insecticides from plants. More recently he has over 12 years' experience in pharmaceutical research, primarily in the medicinal chemistry department of a world leading biopharmaceutical company looking into synthesizing test material as potential new drug molecules for inflammation and immunology diseases. Since 2002 he was directly responsible for managing all the compounds in the corporate collection, advising which molecules could have potential uses in various disease conditions.

Duncan was diagnosed with AKU as an infant after his parents noticed that his nappies (diapers) turned brown/black on standing. He is the fourth of five children. His three older siblings do not have AKU, but his younger brother does have AKU. His parents are unrelated and there is no family history of AKU. He is married with three teenage children, none of whom have HGA in their urine.

Duncan started feeling the effects of AKU in the late 1990s, and his mobility degenerated to the point where he was unable to continue working full-time and was invalided out of work in June 2010 at the age of 48. He now volunteers for the AKU Society on a part-time basis.

The author had no affiliation to either the manufacturers or the distributors of Nitisinone.

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