



## Life After

# KHV

*Walk a mile in his shoes*

STORY AND PHOTOGRAPHS BY  
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*Remember, a Koi that causes an outbreak is essentially a Koi that was the victim of a previous outbreak.*



Once upon a time, I lived in a world where I thought that my business was invincible against KHV. I figured that I had so much fear and respect for this dreadful virus that it would never affect me, or those around me. In July 2006, I was proven wrong! The months of June and July 2006 marked both an all-time high—having won Grand, Mature and Jumbo Champions at the BKKS—and then an all-time low. Following is the story of what happened, and what we learned from the experience.

### *From bad to worse*

The BKKS Show has always been a little different for us than for other dealers. Most go there to sell their wares. For us, however, this particular show is one where we showcase all of our best Koi. So, several weeks before the show, we rounded up all of our best Koi and prepared them to take to the show, in order to impress fellow Koi keepers.

On return from the BKKS National Koi Show, and after allowing the Koi to settle back in, I set about the usual process of warming the Koi back up to our normal summer growing temperatures of 24°C/75.2°F. When the ponds reached 23.5°C/74.3°F, I noticed a customer's Sanke breathing heavily. It had resided with us since May 2006, and we had taken it to the show. Naturally, I was a little concerned, as this was a particularly good Koi that had been left in Japan since Tosai, and now measured 74cm/29in.

Upon closer inspection, it was apparent the Koi had a little gill filament damage on one side. I took a scrape from the Koi's gills and found parasite damage. I concluded that this parasite damage had most likely triggered Bacterial Gill Disease. I decided to raise the temperature quickly to just under 28°C/82.4°F, as such a temperature would fix this problem. I was cautious not to raise it any farther, just in case I would be inadvertently fixing KHV. This rise in temperature accelerated the Koi's decline in condition, and the Koi died on the eve of the same day.

On the following day, two more Koi died, and when a fourth one died on July 22, I started to panic, fearing the worst. It was at this stage that I decided I should take measures to stop the problem in its tracks, and to take a Koi to CEFAS (Centre for Environment, Fisheries and Aquaculture Science) on Monday, July 24, for PCR testing. My logic was that if it wasn't KHV, then at least I would have kept the losses to a minimum, rather than just watch Koi die every day until the test results came back, only to find that I had been losing Koi unnecessarily. My countermeasures worked, as my last mortality occurred on that Monday morning, bringing the death toll to seven Koi, out of 80. However, on August 4, our worst nightmare was confirmed by CEFAS: **It was KHV.**

**IMAGE 1:** A dehydrated head is also one of the key symptoms of KHV. If you imagine where the skull bone areas are, and where the fleshy areas are, you will understand how the fleshy areas 'dry out', causing a sunken, wrinkly appearance. If you

stare long enough, you will be able to see the resemblance to Michael Cox.

**IMAGE 2:** In order to obtain a blood sample, you have to draw it from the caudal artery, which runs directly underneath the spine. Some choose

an entry point beneath the tail tube. I favour the route shown for several reasons: 1) You can see afterwards if an infection has occurred; 2) You don't puncture any scales; 3) On a big Koi, you will never obtain a needle long enough to hit the artery.

**IMAGE 3:** We use serum separator tubes, which we then centrifuge. The force of the centrifuge forces the red blood down through the gel barrier, leaving the serum separated out on top, which can easily be drawn off for testing. If not

separated, the red cells will break down and ruin the serum sample. **IMAGE 4:** Gill necrosis is one of the symptoms of KHV. **IMAGE 5:** One of the seven Koi that died before we decided to stop the outbreak in its tracks. This Sanke

was one we had grown from small Tosai, to become 64cm/25in Nisai. **IMAGE 6:** Another Koi bought as Tosai, and measured 71cm/28in as Sansai. **IMAGE 7:** Two of our dead Koi.

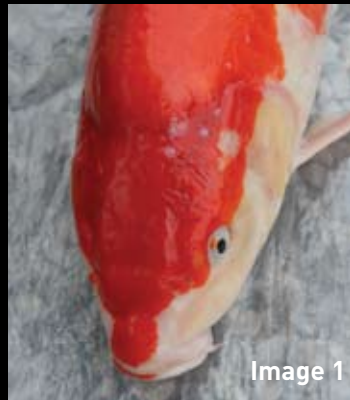


Image 1



Image 2



Image 3



Image 4



Image 5



Image 6



Image 7

### Diagnosis and Discovery

On Tuesday, August 8, we received blood sampling equipment and a centrifuge. We then set about the task of blood sampling each and every Koi in our 'healthy' ponds to establish if any Koi posed a risk of KHV. Once blood testing was completed, we hit the road and traveled the length and breadth of the UK, testing every Koi received and sold from our May 15 shipment, as well as a large number of Koi that dated back more than 12 months prior to this date, in an attempt to try to establish where our KHV came from, and whether we had placed any of our clients' collections at risk. We also tested Tosai that we had sold to three other dealers, as these were siblings to those in our infected ponds. The results of these blood tests proved to be very interesting! In short, the few Koi that we sold at the BKKS Show were fine; they had been up at around 25°C/77°F in their new ponds, due to a heatwave we were having at that time, with no problems. But, four Koi that we had sold as Tosai, and had been grown in Japan through to Nisai, tested positive. These four Koi resided in three clients' ponds. In

addition, one Koi tested positive in one of our healthy ponds. This one had a common link with the other four Koi: They were all exposed in Japan.

The shocker behind the testing, however, was that, of the three dealers' Tosai that we tested, dealer 'A' had experienced no health issues, dealer 'B' had experienced a few losses that were a mystery to him, and dealer 'C' had experienced a bacterial problem, resulting in ulcers on some Koi. The testing, however, revealed that Koi from dealer 'A' returned positives from Kohaku Tosai, but not the Sanke that cohabited with them. Dealer 'B' had positives on many Koi, including pre-existing Koi. Dealer 'C' had the same proportion of positives as dealer 'A'. So, from this, it was possible to conclude that dealer 'A' had potential carriers among his Kohaku Tosai. Dealer 'B' had experienced an outbreak of KHV among his Tosai, but had "fixed" it himself. Dealer 'C' had experienced bacterial problems, but not KHV. Dealer 'A' was the vital link to our KHV outbreak. His Koi had been heavily temperature cycled after their arrival in the UK on May 15, but with

no issues. However, after we got the results back, the dealer cooperated by temperature cycling his Koi once again. The strange thing was that, despite using an anti-viruside in his pond, an outbreak proved remarkably easy to induce!

The conclusion of our outbreak was that the cause of it wasn't the Koi we would have expected, but the Tosai that arrived on the same shipment. It was a pure miracle that I didn't take these Koi to the show, as this would have certainly caused losses if sold. By chance, of the other dealers, only one of them had sold one of the Tosai, and this one was euthanised without causing any problems. I had myself sold three, and these were also euthanised and replaced, as were the other dealers' Koi. As far as the positive Nisai were concerned, the ones in the customers' ponds were destroyed. The one that we found at our place was removed to a temporary pool to which we added some small Tosai from another source (believed naïve). We then temperature-cycled this pool very heavily between 15°C/59°F and 25°C/77°F, three times. We also repeatedly blood tested it up until January 2007, and each

time it returned a result that was 'off the scale.' In the end, we sent this Koi to CEFAS for further research. They, too, could not induce an outbreak, but concluded that it was most likely a carrier, since it was still producing antibodies.

### The Bright Side

Our experiences with KHV in 2006, and all of the testing that we had done as a result, was a huge eye-opener for us, and indeed for many others in the Koi industry. I believe that, although we experienced great losses, and our outbreak most likely frightened many hobbyists away from buying new Koi, there was also a positive side, as it resulted in many other dealers also improving their own protocols. It seems, however, that some still choose to bury their head in the sand a little, figuring they will hear inside information from agents or Japanese dealers if anyone in Japan is seen as a risk. The truth is, however, that Japanese Koi farms, even nowadays, are still generally very unscientific. Many breeders don't even possess a microscope, and they base all of their treatments on a hunch. My opinion is that many KHV cases on Koi

farms in Japan are stemming from the parent Koi. To my mind, KHV is transmitted vertically from the parents, but not to every offspring. The scary part in all of this is that some breeders are getting KHV without even realising it, and others choose to sweep it under the carpet and 'deal with it' rather than tell the government and be shut down whilst testing is carried out.

On a brighter note, an increasing number of breeders are now sampling Koi that have been recovered from sickness, and new parent Koi, and sending the samples to America for antibody testing. I have sampled and had parent Koi tested for four breeders in Japan. With those breeders that have never seen KHV, the worrying part is that, if a disease fits into the 'none of the above' category, then heat and salt tends to be the answer. So, if Fufusho (sleeping disease) occurs, then the answer is 30°C/86°F and salt. If Bacterial Gill Disease is suspected, then 30°C/86°F and salt is the answer. If Holes Disease occurs, then 30°C/86°F and salt is the answer. BGD and Holes Disease show somewhat similar symptoms to KHV, to a degree. It is my opinion

that KHV is sometimes being inadvertently fixed, as the same treatment regime will also recover fish from KHV.

### Lessons Learned

One of the scariest things about KHV is that a carrier that has been recently infected will be much stronger than one that was infected perhaps 12 months ago. So, assuming that a Koi had been recovered a month ago, and then imported, it is actually very unlikely to shed virus to other Koi, as its antibody count will be so high that the virus will be kept 'in check' within the Koi. However, post-exposure, the Koi's immune system will relax, and the antibody count will fall and an outbreak will become easier to induce. Remember, a Koi that causes an outbreak is essentially a Koi that was the victim of a previous outbreak. KHV doesn't happen unless a carrier is present.

Some people don't believe in temperature ramping. They believe that if the Koi arrive and are then placed into warmer water, then this will be enough. The assumption is that the cargo hold water will be maintained at below 15°C/59°F.

**IMAGE 8:** This Koi looked dreadful during the outbreak, and we thought it would be the first one to die. But, after we stopped the virus, the Koi recovered to look like nothing was ever wrong. We had countless offers from people wishing to

buy this Koi but, my reason for euthanising it, despite most people promising never to sell the Koi, is that someone might inherit and then decide to sell it.

**IMAGE 9:** The author running a blood sample through the centrifuge.

**IMAGE 10:** This Koi tested positive and was moved to another pond before the outbreak. We temperature-cycled it repeatedly with some naïve Koi, but it didn't trigger an outbreak. CEFAS also couldn't induce an outbreak with it.



Image 8



Image 9



Image 10

*To date, we haven't found any Koi that have shown an increase in antibody levels after cycling.*

However, quite often, the Koi are packed in water of 18°C/64.4°F or 19°C/66.2°F in Japan and will often arrive at much the same temperature. The shipment we had was via KLM and should have been kept at below 15°C/59°F in the cargo hold. When the Koi arrived at our end, the water was around 15.5°C/60°F, and placed straight into water of just under 18°C/64.4°F, which we then ramped to 24°C/75.2°F. This wasn't enough to trigger an outbreak. When we went to the BKKS National Show in June 2006, we left Tosai in our two 1700UK/2041US gallon ponds. These ponds were maintained at around 18°C/64.4°F whilst we were at the show. On our return, we reintroduced all of the Koi from the show to these two ponds and heated them back up to growing temperatures of 23.5°C/74.3°F. At this point, the Tosai caused our outbreak. It would appear that this somewhat later temperature cycle triggered the virus.

#### ***New Protocol***

As a result of our outbreak, we now see things very differently. When we import Koi, we now blood test

each and every one of them using the CEFAS ELISA test. This test is highly sensitive but, unfortunately, not incredibly selective. I believe this test is extremely good for finding safe Koi, but not good for finding dangerous ones. What I mean by this is that there does seem to be some reaction with Carp Pox antibodies. So, any positives we find are isolated then retested using Serology test. If these Koi come back positive again, they are destroyed. If they come back negative, the Koi are extensively temperature cycled. Any Koi with an antibody level anywhere close to the negative threshold is retested after temperature cycling to make certain that its antibody level hasn't risen. This confirms that the Koi isn't a risk, as a carrier will increase its antibody level after being temperature cycled.

To date, we haven't found any Koi that have shown an increase in antibody levels after cycling. This is extremely encouraging. While this method of testing is expensive, for us, it's easy, as the Koi are generally pretty valuable, and the number of Koi is low, so the cost is relatively insignificant. Since July 2006, we have had around 2000 Koi tested.

It's not all doom and gloom, however. Most dealers are now being extremely responsible with new shipments, and are double-cycling new shipments in the correct manner. Given that these shipments will have many Koi from one breeder, if there are issues, there will most likely be a number of carriers within one group of Koi, so an outbreak will most likely be induced in a controlled quarantine environment. For me, though, I have very low numbers of Koi from each source I buy from, so my own methods are more suited to my needs. If I buy three Koi from one breeder, there is a high chance that they are all unrelated, so there is no point in losing all three when only one may be a risk. A large number of Koi that are siblings of one another is another matter... ❄



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