Podcast Proposal: R399E at angry@arthritis

Steve: Intro…

Q: Martin, you are a Professor holding a medical degree, and you have been working for Merck KGaA in Darmstadt, Germany, until your recent retirement. Tell us a little bit about your background and working life.

A: I started my professional career as a scientist at the department of Physiology at the University of Kiel, Germany. As a medical doctor by training, I wanted to better understand why patients are in chronic pain. There are so many different diseases where pain is the prominent aspect for patients, so this was and is a huge medical problem. By the way, one of these conditions with people in chronic pain is Osteoarthritis, I will come back to this in more detail later. During my academic career I was conducting preclinical research focussing on mechanisms that could contribute to chronic pain. After some time I became more and more interested in combining knowledge about mechanisms of pain with opportunities to develop new medicines that can modulate such mechanisms in order to finally help patients. In consequence, in 2000 I took over a first position in pharmaceutical industry as program lead “Pain in degenerative Joint Diseases” at Aventis. While starting my second career in pharmaceutical industry I stayed connected to academia and became Adjunct Professor of Physiology at Universoity of Erlangen/Nuremberg, Germany. The last 10y until my retirement in 2022 I was Head OA Research and Early Clinical Development at Merck KGaA. By the way, Merck KgaA is a globally acting pharmaceutical company headquarted in Darmstadt, Germany, not to be mixed up with american Merck&Co, which is MSD in Europe. During my tenure with Merck KGaA I had a group of around 20 scientists and technicians, and was responsible for starting and advancing a portfolio of programs in Osteoarthritis.

Q: We speak today about a potential new treatment for Osteoarthritis, called R399E. Before we come to R399E, I want to ask you as a physician scientist what do you consider the most important challenge in the treatment of OA?

A: The challenge in the treatment of OA derives from what is actually happening within joints in the course of the disease OA. So let us have a look into a joint, as an example a knee joint, but what I´m going to outline is happening in all other OA-affected articulating joints as well. In a knee joint of an adult person, there are continuously endogenous processes ongoing that affect tissues of the joint, e.g. cartilage. These are build-up processes like formation of new cartilage, and processes of breaking down existing tissue, e.g. enzymatic degradation of cartilage building blocks. The tissues of the knee joint are healthy as long as such build-up and degradation processes are balanced. Now, Osteoarthritis is a disturbance of the balance between build-up and degradation processes in the joint. Enzymes break down the cartilage and inflammatory mediators are released. More and more cartilage is being destroyed while on the other hand less cartilage matrix than normal is newly formed. The result is a progressive pathologic remodeling of all tissues of the joint. This has overall structural consequences: e.g. the amount of cartilage in the knee joint is getting smaller and smaller. The breakdown of cartilage and the release of inflammatory mediators has another important consequence for the patient: the patient feels pain in the affected knee. For patients, the most

devastating aspects of OA are exactly this pain, and the accompanying functional limitations. This has tremendous impact on normal life, it hinders patients for example to rise from bed or to ascend stairs. Both, pain and functional impairement get worse with time while the disturbance of joint homeostasis rises. Today, therapy of OA starts with self-management to improve fitness and joint health, which can be assisted by physiotherapy. The knee pain can be treated with analgesics but the treatment success is often reported by patients as not being satisfactory. When pain intensity further incresases with time, surgical joint replacement becomes necessary when the patient can no longer tolerate the pain. However, surgical interventions are not a perfect solution, e.g. >20% of patients remain in pain after surgery, and the knee replacements have a limited survival.

So, based on what is happening in the knee joint during Osteoarthritis, the real challenge in the treatment of OA patients in essence is twofold: first, the treatment should permanently improve symptoms, i.e. reduce pain. Secondly, the treatment should re-balance build-up and degradation processes because such a re-balance will ultimately postpone or even supersede the need for joint replacement surgery.

Q: Tell us about R399E. What was the initial idea?

A: So, it all started in 2013. As I pointed out I was at that time head of OA research at Merck KGaA. A colleague who had recently joined my group, had in her past scientifically worked with a molecule called growth and differentiation factor 5, GDF5. This is a growth factor which is naturally occurring in all of us. Early in life, GDF5 is very important for the initial formation of cartilage and bone. And therefore, others had already the idea that GDF5 could be beneficial in situations where cartilage is being lost, e.g. due to Osteoarthritis. They tested this hypothesis by injecting GDF5 into knees of rats with OA. Animals who received GDF5 had more cartilage than rats who did not receive GDF5 injections into their knees. This was a promising result. However, rats who got GDF5, also had bone growth within treated joints at locations where there should be no bone. So, this was a warning signal. Obviously, when GDF5 was injected into knees of adult rats, it initiated cartilage growth but at the same time also bone formation which would not be tolerated by OA patients. My colleague had the idea, that if one would modify GDF5 in a way that the modification still initiates cartilage formation but no longer bone formation, that such a modified GDF5 could be a good treatment option for OA patients.

Q: How did you come from the initial idea to R399E? It has been discovered in a collaborative approach, right?

A: Yes, that´s true. My colleague Kerstin who had the initial idea, knew about a small company called Biopharm in Heidelberg, Germany. Biopharm had a long-standing experience with the generation and modification of growth factors incl. GDF5. So, under a collaboration agreement of Merck KGaA with Biopharm, we told the Biopharm experts about Kerstin´s initial idea. And Biopharm experts started to work. They knew which part of GDF5 binds to specific receptors which is the basis for exerting its biological effects. Exactly there, they modified GDF5 and produced a number of different modifications. These different modifications were then tested in my group and compared with respect to several properties. R399E turned out to be the best among all tested modifications showing good ability to grow cartilage but less potential to grow bone than GDF5. GDF5 is a protein, a string of 120 amino acids, and the exchange of one amino acid (R, arginine) by another amino acid (E, glutamic acid) at position 399 resulted in what got the funny name R399E. I find it fascinating that the exchange of just one out of 120 amino acids can significantly modify the biological properties of

the molecule. So, yeah, R399E is truly the result of a creative collaboration bringing together scientists with different expertises which complement each other.

Q: What can R399E do?

A: Once we at Merck KGaA had selected the molecule R399E, we started to characterize its potential to treat OA in a number of different assay systems. We used cartilage and other tissues which came from OA patients who just had received a joint replacement, so tissues which otherwise would have been discarded. Using these human tissues, we could demonstrate that R399E reduced the production of cartilage-destroying enzymes and thus the degradation of cartilage. In addition, and this was a really surprising result, R399E also reduced the production of inflammatory mediators and pain messengers. When such messengers are released within OA-knee joints of patients, they evoke the sensation of pain, i.e. OA patients suffer from pain since in their knees, pain messengers are released. And R399E did reduce the production of such pain messengers, at least this is what we found in our experiments with patient derived material. So we went on and tested R399E in animal models of knee OA. But how to measure knee pain in rabbits? You cannot ask rabbits about their pain as physicians can do with their patients. Briefly, in our model rabbits develop OA in their right knee joint but not in the left. While OA develops rabbits put less weight on their right OA knee compared to the healthy left knee, like OA patients when they are in pain, also patients relieve the painful joint. We developed a method to objectively measure how much weight a rabbit puts on the right OA knee and how much on the left healthy knee. The bigger the difference, the more pain the rabbits feels, this is our fair assumption. We could show that rabbits who received R399E injections into their right OA knee loaded the diseased limb significantly more than animals who received a placebo injection. Based on our assumption this indicates that R399E reduced knee pain in rabbits! Importantly, we could see the pain reduction as early as 6 hours after the first injection of R399E. The degree of pain reduction kept constant during the complete study duration of 3 months. The effect on pain was greater and longer lasting than when doses of triamcinolone equivalent to those effective in humans were adminstered. Triamcinolone is a steroid which is injected into painful knees of OA patients. Last but not least, injection of R399E into OA joints in rabbits not only resulted in this clear pain reduction but it also reduced structural changes of cartialge and other joint tissues. In a second model of OA in sheep R399E injections resulted very similarly in reduction of pain and structural changes.

Overall, R399E addresses both challenges that we have in the treatment of OA patients as I have formulated them earlier: Based on our tissue and animal data, R399E acts on different cell types in OA joints and is doing several things in parallel. With this, R399E reduces OA-related pain, and also seems to re-balance build-up and degradation processes in cartilage and other joint tissues. What has to come next is to investigate the potential of R399E in patients with knee OA.

Q: You pointed out that the data in tissue culture and in animal models are promising but that there is yet no data existing which demonstrate that R399E can treat human OA patients. How big do you consider the chance that R399E also works in human OA patients?

A: This is a very important question that every prospective medicine has to answer. The short answer is that we simply don´t know. But let us take a little closer look. I think that the chance to work in

humans depends on the quality of assay data which have been collected with the drug candidate. As I have pointed out, R399E has been tested on human tissues, cartilage and other tissues from patients who received a new joint, i.e. joints which were already destroyed to such a degree that joint replacement surgery became necessary. If R399E works in tissues of such advanced disease stage this increases the likelihood that it could work when injected into patient knees. Another point is pain measurement in animals. Merck KGaA has used a quantifiable measure, weight bearing behavior, which has the advantage that it reflects the voluntary expression of the rabbits, produced without any interference of the experimenter, so I would consider this an objective measure of pain. This certainly increases reliabilty. Moreover, using this objective pain measure, R399E performed better than a steroid, triamcinolone, which since many years is one of the treatment options for OA patients and for which it is known that it can reduce OA knee pain in patients to a certain degree.

Overall, I would say that there is still a considerable risk that R399E when tested in patients may not work. However, based on the quality of available data, as outlined before, I would say that the chance that R399E may work in OA patients is as good as it can be at this stage.

Q: How far is R399E developed today, and what is the timeline to market?

A: All preclinical investigations which are necessary and mandatory to be conducted before entering into clinical development have been concluded. If the next step of R399E´s development would be started tomorrow, it will take approximately 3 years until first clinical data from human patients with knee OA will become available. Depending on the outcome of the first clinical trials, further clinical development may be accelerated so that in an optimistic scenario, timeline to market could be 6 years from now.

Q: What would you say are advantages of R399E over other approaches to treat OA?

A: Let me first look into direct competitors of R399E, i.e. novel pharmacological treatment options which are given either orally or via injection into the knee, and which are in clinical development with the ambition to address both challenges of OA, namely to reduce pain and to improve structure. There are currently around 10 different clinical development programs ongoing. The important differentiation factor of R399E is, that R399E – at least as seen in rabbits – can start to reduce pain already hours after the first injection into the knee. To my knowledge, no other of the aforementioned competitors has shown pain relief of such quick onset. For patients, this could be a significant advantage since they do not want to wait for months or even longer until their pain gets better. Another advantage for patients could be that treatment with R399E could re-balance build-up and degradation processes which may ultimately postpone or even supersede the need for joint replacement surgery.

There are other approaches like autologous chondrocyte transplantation which are considered to treat OA. This is an invasive approach which requires two operations, one to harvest cartilage cells from the patient´s own knee to grow them in the lab, and a second to implant these new cells. Certainly, this is a more invasive approach than to inject a drug like R399E into knee joints.

Q: You mentioned that R399E is to be injected into knee joints of OA patients. How often has this to be done, and how do you compare this form of administration with systemic administration, i.e. taking a pill?

A: As said, R399E has to be administered directly into affected joints, knees or hips, via injection. Since R399E will be given to reduce existing pain and also to improve health of cartilage it is expected – based on the existing data - that 4-6 injections will be needed during the course of a year, and that the therapy probably has to be continued. Each injection will be done by an experienced physician. This is a disadvantage over taking a pill at home. On the other hand, injecting R399E directly into the OA-knee brings the right amount of drug exactly and exclusively to that place where it is needed, namely the OA knee joint in contrast to a pill where after absorption the blood stream brings the drug to all tissues of the body whether it is affected by OA or not. Overall, when injected into the knee joint R399E has the potential to be effective against OA while not reaching other organs and tissues thereby minimizing the risk of unwanted side effects. In striking contrast to what is known as opioid crisis, it is an important point for patients that injections of R399E into knee joints – based on how R399E is working there and because it is only working where it is injected – do not bear a risk of inducing any overuse, misuse or abuse, and even overdose deaths.

Q: You told us at the beginning that last year you retired from Merck KGaA, thus a good time to leave former professional activities behind. Why are you still engaged with R399E?

A: As I tried to outline, the profile of R399E appears very appealing and my personal view is that the existing data definitely justify that R399E is tested in human patients with knee OA. What I did not mention so far is that Merck KGaA has decided for strategic reasons to be no longer engaged in OA. In consequence Biopharm, the small company which originally designed R399E, received back all rights to R399E. Based on my deep insight in the existing data, in light of the huge medical need in OA and the fact that only a few other approaches are currently in clinical development, I´m highly motivated in supporting Biopharm to find partners and investors, to enable start of clinical development of R399E.

OA is such a huge indication, nevertheless not a single disease modifying therapy is approved yet, so the medical need is tremendous, and on the other hand the engagement of pharmaceutcal industry and venture capital is rather limited. I know that you, Steve, have come to a similar diagnosis of the current situation. I hope that in the future interest in academic research to explore novel mechanisms of the disease OA will raise, and that more money will become available for preclinical as well as clinical innovation, ultimately for the benefit of many millions of OA patients, and I´m here to support this progress.