

Introduction

Spider silk has been around for 100s of millions of years and it has caught the attention of researchers around the world for its incredible mechanical properties. There is no man-made or natural material quite like it and it has captivated medical research due to its biocompatible and adhesive properties. The recombinant spider silk used in this research is completely biocompatible.

Applications for spider silk coatings are broad, however the interest in this subset of research is to successfully coat medical implants and other medical materials, such as heart stents and catheters, which will drastically increase their biocompatibility, decreasing the chance of infection as well as recovery time due to decreased tissue growth in response to the foreign object.

Spider silk coatings can be produced using a solvent of 1,1,1,3,3,3-Hexafluoro-2-propanol (HFIP) or water, however the toxicity of HFIP causes it to not be a suitable choice for in vivo applications. Aqueous coatings do not have the limitations of HFIP based coatings. This makes it possible to incorporate cytokines, growth factors, antibodies, stem cells, and antibiotics into aqueous coatings.

This study will focus on the preparation and application of spider silk solutions to stainless steel (a popular choice for biomedical implants) and the use other additives to increase the adhesive properties of the silk solution. Coatings provide strength, but are also elastic as they transparently coat many different materials.

Methods

Standard water based coatings are made using dopes which contain a small percentage (w/v) recombinant spider silk protein dissolved in water along with different additives such as acids, bases and detergents. The recombinant spider silk powder (insoluble in water by nature) was dissolved by a proprietary process that results in 100% solvation and 100% recovery of protein. After solubilization the dope is transferred into a microcentrifuge tube and spun to remove particulates and impurities. The dope is then poured onto stainless steel or put into an atomizer to be sprayed onto the stainless steel and then allowed to dry. The solution on the steel is then put through a process that enhances the presence of B-pleated sheets. Evidence of this enhancement is shown in the pictures on the right with the use of Congo red dye.

Before the dope is made it is necessary to treat the stainless steel so that its surface is optimal for coating. This method includes polishing and liquid treatment using different kinds of acids and bases. It is evident that this treatment process drastically increases the adhesive properties of the surface.

Protein powder was attained from the purification of transgenic goats milk expressing the spider silk protein (MW approx. 65 Kda).

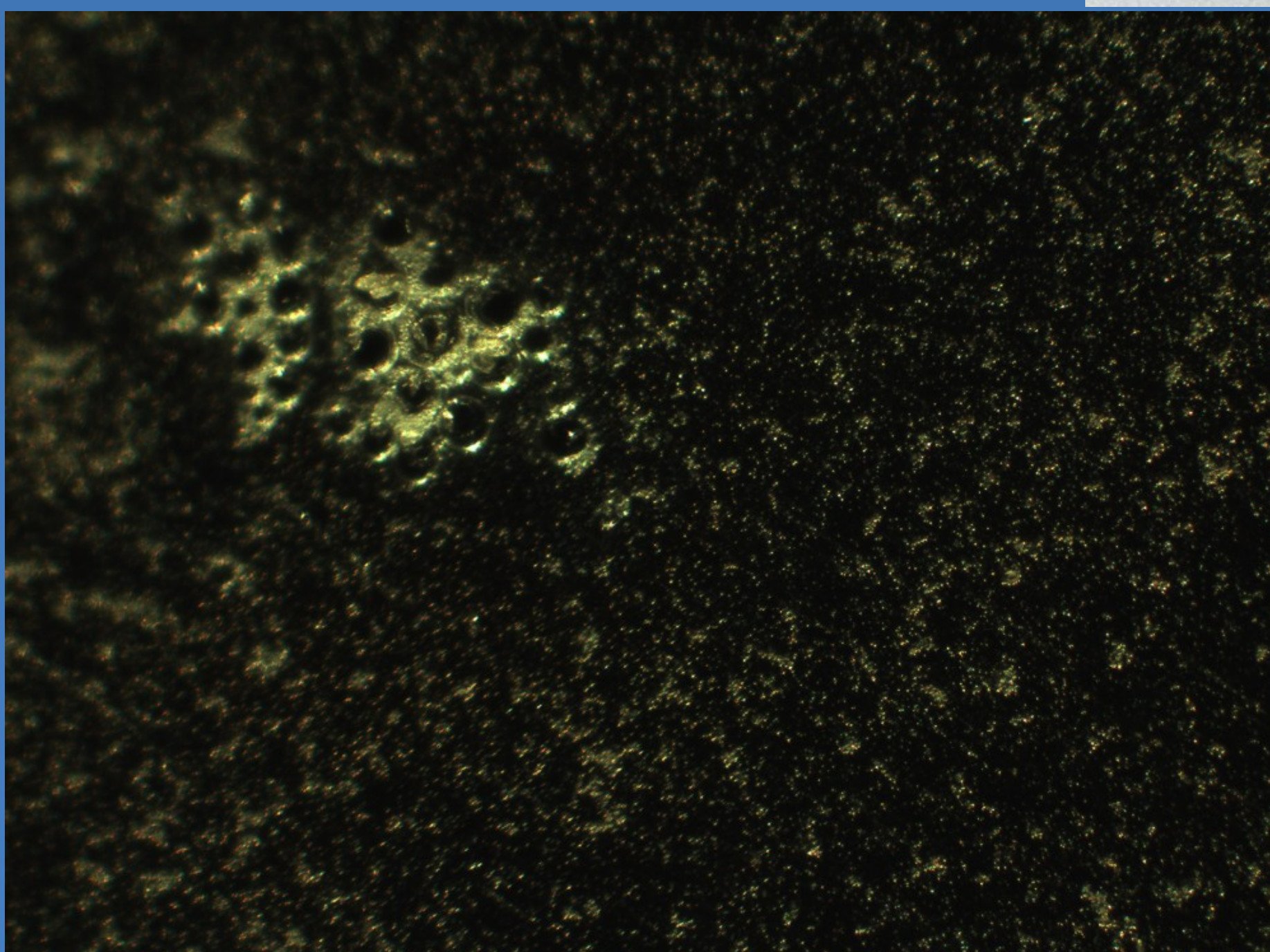


FIGURE 1-Untreated Surface

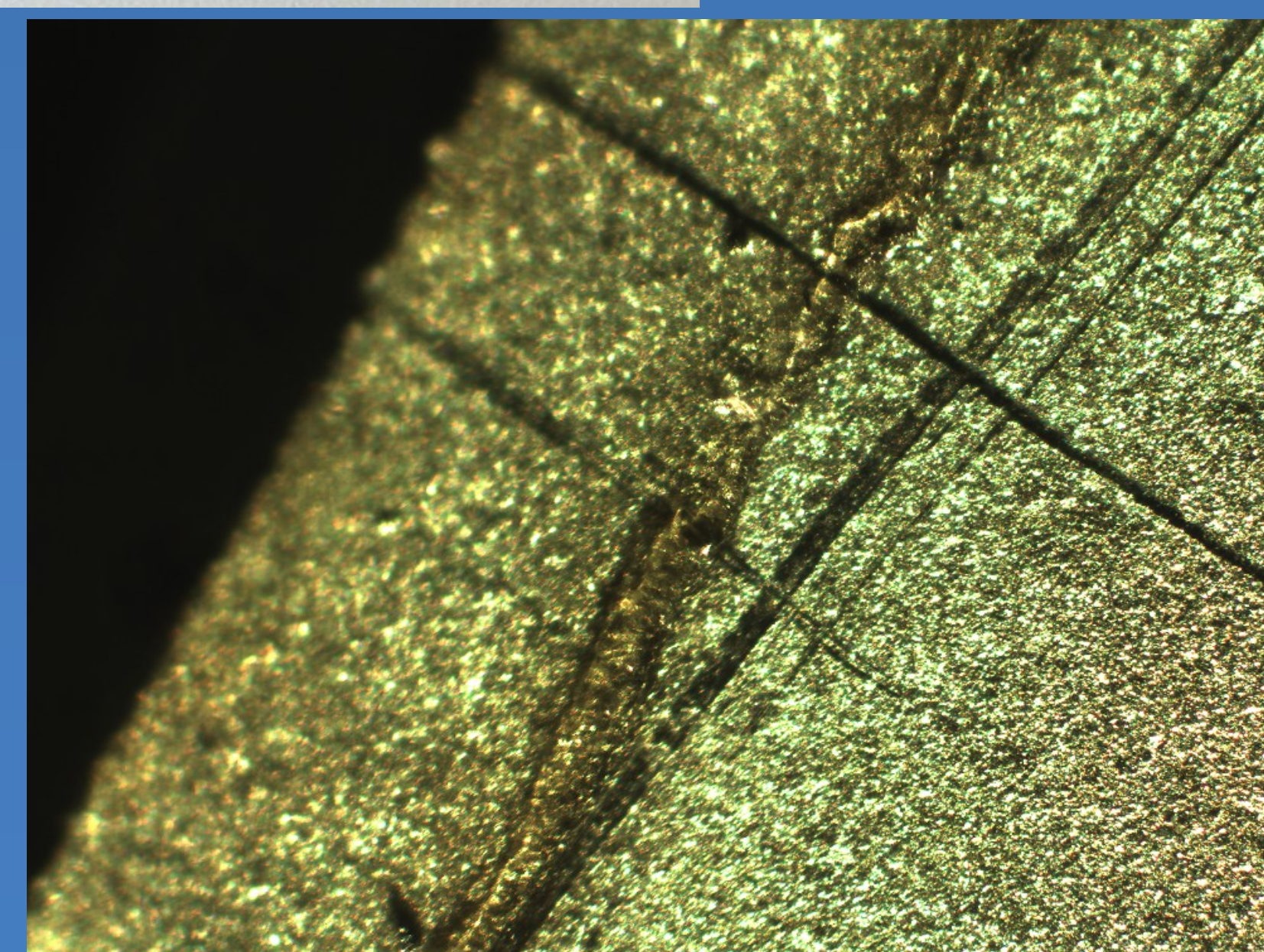


FIGURE 2-Treated Surface

Bubbles on the non-treated surface in Figure 1 show that the coating is not as uniform as on the treated surface in Figure 2. The edge of the spider silk film is shown in Figure 2 on the treated stainless steel surface. This proves both the presence of the film and shows greater consistency in the coating.

Future Work

Stainless steel is not the only surface that has potential to be coated by spider silk. Our lab has already done preliminary work with silicone that shows promise. A silicone coating that can be used for devices such as catheters provide a uniform coating, but are also elastic enough to stay on the silicone material as it bends. Several additives are being explored to enhance the strength, adhesiveness, and elasticity of the coatings on all kinds of materials, including silicone and stainless steel.

Aside from the manipulation of dopes, preparation of the different surfaces has proven that the adhesive properties of the surface can be drastically enhanced. As more surfaces are coated, new and improved techniques for processing these surfaces will be found. In addition to preparing the surface, there are a number of different ways that the coating procedure can be altered, enhancing the quality of the coating that is delivered. Testing the surface of the coating is a procedure that is just starting to be developed and new technologies could show us things about the coating's surface that will allow us to manipulate the coating to our favor.

The preliminary studies of these spider silk coatings are very promising. The future of this research will lead to better coatings that will provide both strength and elasticity. We have established that spider silk solutions can be combined with drugs, protein therapeutics and growth factors that can be released in the body to aid and improve recovery time. The aqueous coatings being developed are perfect solutions to add these factors making the coating multi-functioning inside the body.

FIGURE 3-Dyed treated surface

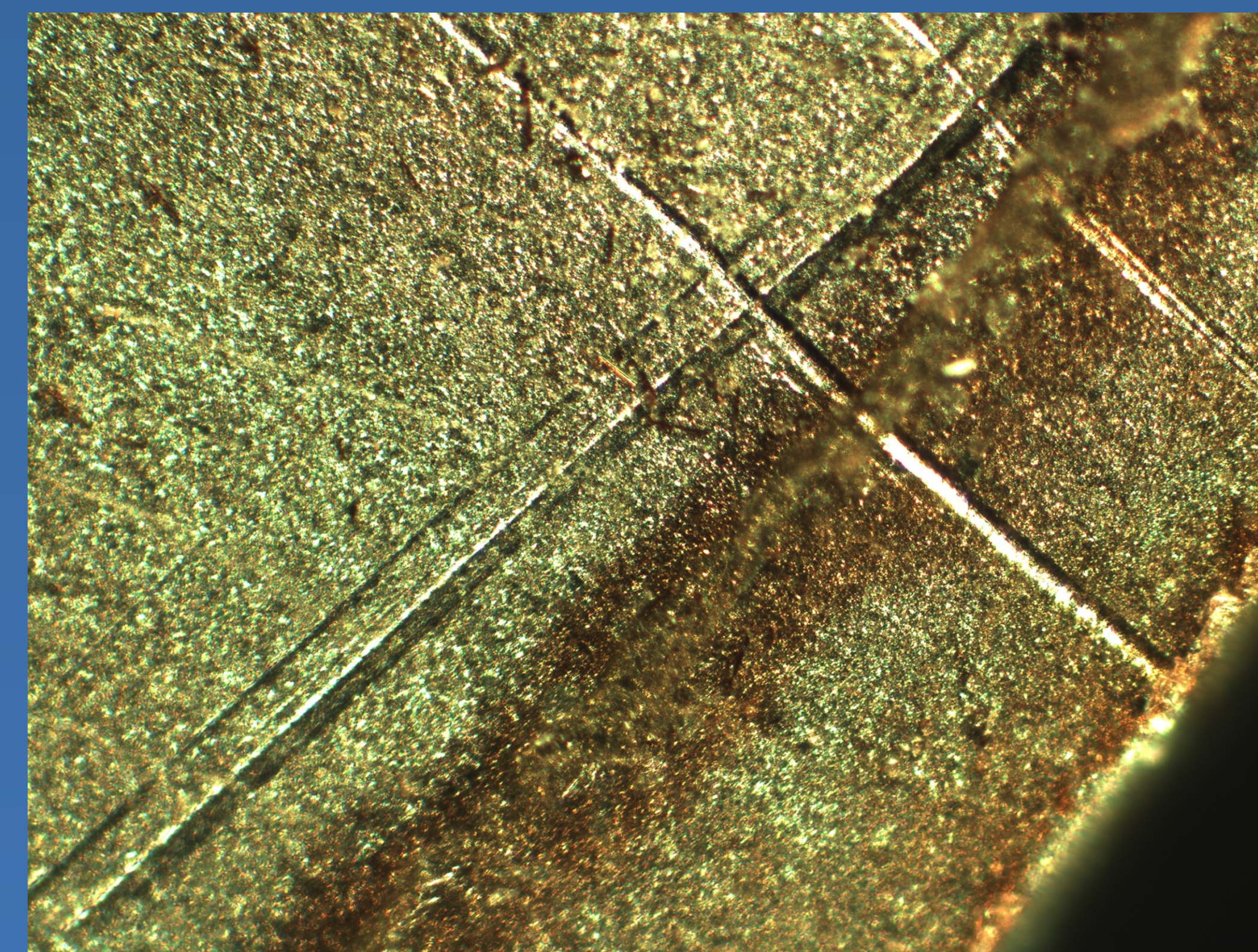
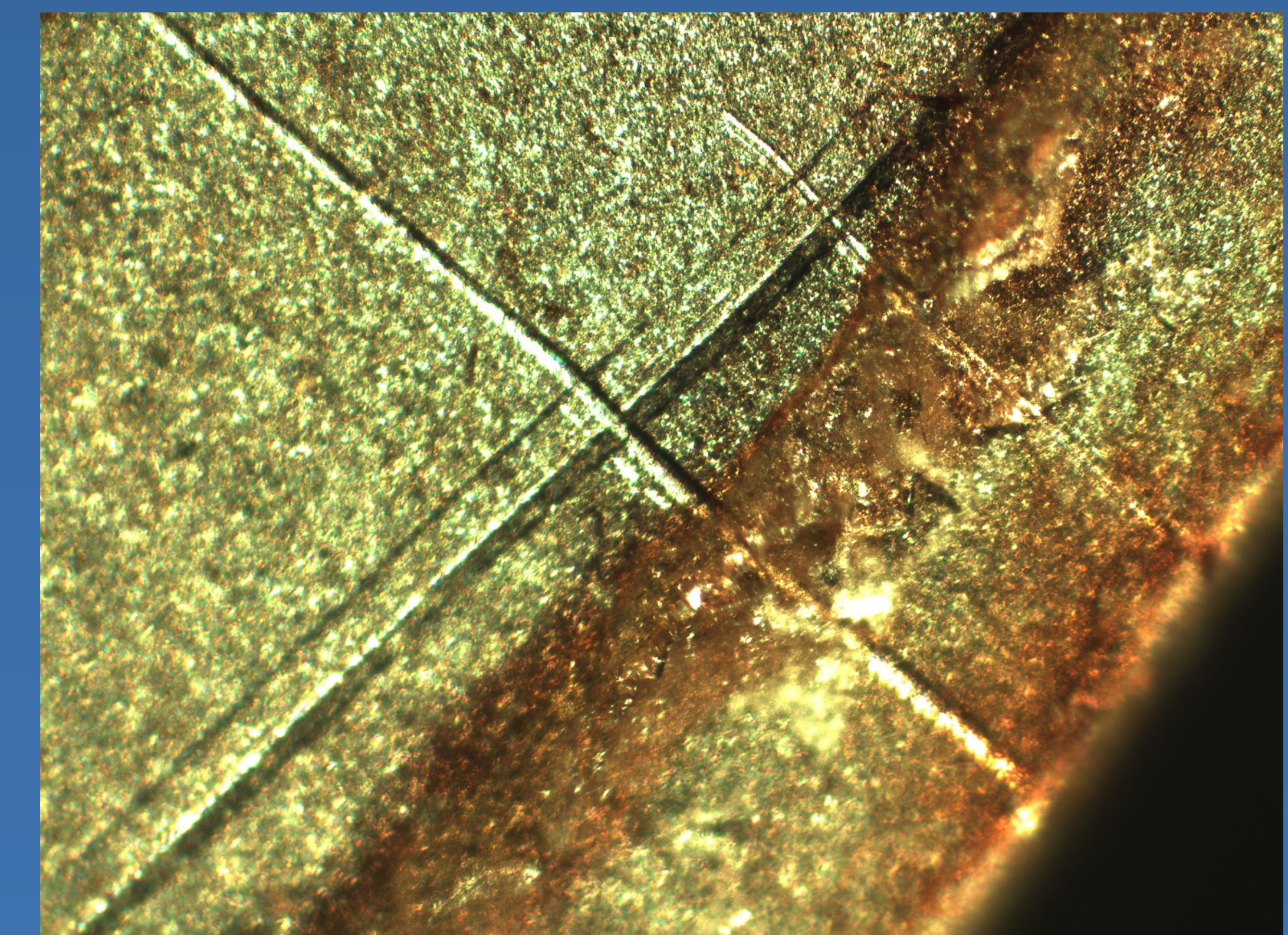
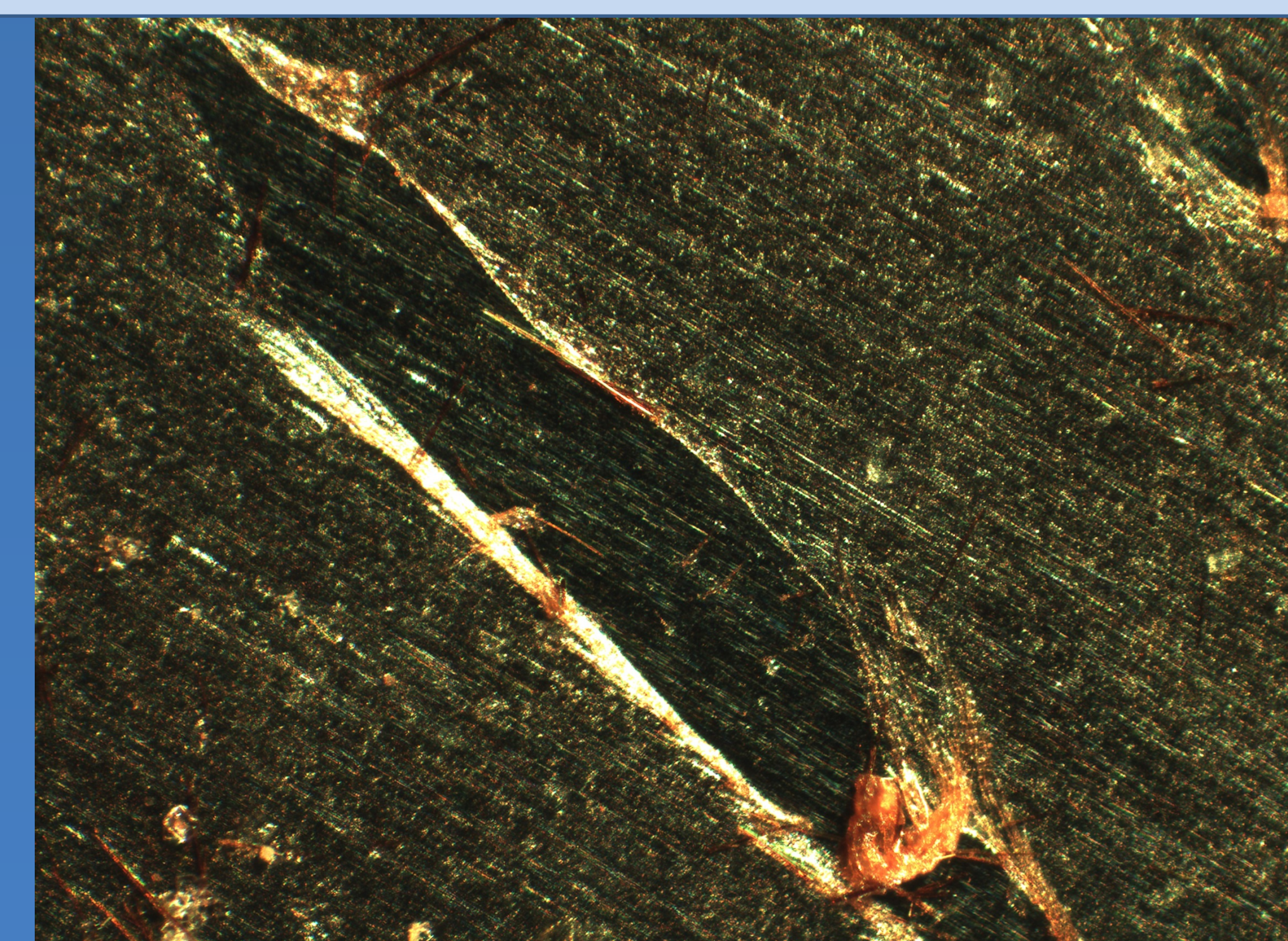


Figure 4-Dyed treated surface with enhancement



The Congo red dye sticks to B-pleated sheets (in the structure of the spider silk protein). In the microscopic image in Figure 3 you see the original edge of the treated stainless steel coating treated with dye. In Figure 4 you see this edge after the enhancing process treated with the same dye again. The brighter red color shows that the enhancing process aids the formation of B-pleated sheets in the protein structure making a stronger coating. This also is further evidence that the coating of spider silk exists on the steel since it is hardly noticeable to the naked eye.



On the non-treated stainless steel surface holes in the coating occur like the one shown in Figure 5 (also seen using the Congo red dye). These are not found on the treated surface.