Electrochemical investigation of mussel adhesive protein films for corrosion protection.

Diploma work was conducted at Division of surface and corrosion science, Royal Institute of Technology (KTH).

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Abstract

Today there are numerous methods to slow down a corrosion process of metallic materials. However, due to environmental effects and health risk issues, several traditional corrosion inhibitors have to be phased out. Hence, it is of great importance to develop new corrosion inhibitors that are “green”, safe, smart and multifunctional. In this essay, the focus is on mussel adhesive protein (MAP) and its possibility to reduce the rate of the corrosion process. The protein exhibit great adhesive strength and protective properties, allowing it to adhere to a multitude of different surfaces and is therefore of great interest of corrosion science.

The protein Mefp-1, derived from the blue mussel’s foot, had been pre-adsorbed on the carbon steel surface and provided good corrosion inhibition in a basic chloride solution for a short exposure time. The protection was further improved with the assist of iron and ceria ions by formation of protein/ions complexes within the surface films and thus enhanced the corrosion protection for longer exposure time. Ceria nanoparticles were used in order to create a multi-layer composite film with an even higher corrosion protection. The results suggest a denser film compared to previous samples and a more uniform surface.

Key words: Mussel adhesive protein (MAP), corrosion, corrosion protection, electrochemical impedance spectroscopy (EIS), potentiodynamic polarization
Table of Contents

INTRODUCTION .................................................................................................................. 4

1. CONCEPT ...................................................................................................................... 5
   1.1 Metal behavior ........................................................................................................... 5
   1.2 Corrosion in general ................................................................................................. 5
   1.3 Passive layer ............................................................................................................. 6
   1.4 Pourbaix diagram ...................................................................................................... 8

2. DIFFERENT TYPES OF CORROSION ......................................................................... 9
   2.1 Galvanic corrosion ..................................................................................................... 10
   2.2 Erosion corrosion ....................................................................................................... 10
   2.3 Pitting corrosion ....................................................................................................... 11
   2.4 Crevice corrosion ....................................................................................................... 11
   2.5 Dealloying corrosion ................................................................................................. 11
   2.6 Intergranular corrosion ............................................................................................. 12
   2.7 Stress corrosion cracking (SCC) ................................................................................. 12

3. PROTECTION TODAY ..................................................................................................... 12
   3.1 Coatings .................................................................................................................... 12
   3.2 Inhibitors .................................................................................................................. 13
   3.3 Cathodic protection ................................................................................................... 13
   3.4 Anodic protection ...................................................................................................... 13

4. MUSSEL ADHESIVE PROTEIN (MAP) ...................................................................... 13
   4.1 Green inhibitors ....................................................................................................... 14
   4.2 The byssus ............................................................................................................... 15
   4.3 Protein Mefp-1 ......................................................................................................... 15

5. EXPERIMENTAL TECHNIQUES ............................................................................... 17
   5.1 Electrochemical cell used for measurements ............................................................. 18
   5.2 Electrochemical impedance spectroscopy (EIS) ...................................................... 18
5.3 Potentiodynamic polarization (PD) ........................................................................... 22
6. EXPERIMENTAL ......................................................................................................... 24
   6.1 Results – Fe$^{3+}$ effect on pre-adsorbed Mefp-1 film ........................................... 26
       Description: ............................................................................................................. 26
       Results of EIS measurements ................................................................................. 26
       Summary – Fe$^{3+}$ effect on pre-adsorbed Mefp-1 film ......................................... 29
   6.2 Results – creating an iron-protein complex before film formation ....................... 32
       Description: ............................................................................................................. 32
       Results of EIS measurements ................................................................................. 32
       Summary – Fe$^{3+}$ induced Mefp-1 complexation in the bulk solution ................. 33
   6.3 Results – creating ceria (IV) -protein complex before film deposition ............... 36
       Description: ............................................................................................................. 36
       Results of EIS measurements ................................................................................. 36
       Summary – creating ceria(IV) -protein complex before film deposition ............... 37
   6.4 Results – using ceria nanoparticles for film deposition ....................................... 39
       Description: ............................................................................................................. 39
       Results of EIS measurements ................................................................................. 39
       Results – Mefp-1 ceria nanoparticles ..................................................................... 40
       Summary – using ceria nanoparticles for film deposition ....................................... 41
7. DISCUSSION .............................................................................................................. 43
8. CONCLUSIONS ......................................................................................................... 46
9. RECOMMENDATIONS ............................................................................................ 46
10. ACKNOWLEDGEMENTS ....................................................................................... 47
BIBLIOGRAPHY ........................................................................................................ 48
APPENDIX A ........................................................................................................... 51
APPENDIX B ........................................................................................................... 53
BIBLIOGRAPHY ........................................................................................................ 54
INTRODUCTION
Metals are important materials used in numerous applications and constructions. For sustainable development the material need to be durable otherwise it is neither economically nor environmentally friendly. Corrosion is a normal process for metals. However, it is possible to decelerate the degenerative process by suitable strategies. Normally this is achieved by coating or painting the surface in order to shield it from corrosive environment as well as creating a wanted esthetic appearance. Unfortunately, several protections used today are not suitable for the environment when degraded. This is why researches now focus on materials from nature that could replace the traditional resources. The division of surface and corrosion science of the Royal Institute of Technology is performing research in collaboration with Biopolymer of Sweden and Swerea IVF where studies of mussel adhesive protein (Mefp-1) are in focus. This thesis is initiated by the Royal Institute of Technology (KTH) for aiding their research. There are several studies made on mussel adhesive protein for example, Krivosheeva et al (1) found that the initial adsorption rate is dependent on the pH value. Evidently the adsorbed amount increased with the solutions pH. They also concluded that the adsorbed amount of Mefp-1 was dependent on the charge of the surface i.e. electrostatic forces increase the adsorption. Zhang et al (2) found that Mefp-1 functions as a good corrosion inhibitor on carbon steel. The results revealed that, during a short exposure time, the corrosion inhibition was better for samples with high salt concentration although for long term exposure, samples with lower salt concentration showed more promising results. They also found that the protein did not cover the surface completely. Holes were present and near these defects aggregates had formed. For this research the protein-complex with $\text{Fe}^{3+}$ or $\text{Ce}^{4+}$ were induced to study the differences of corrosion protection compared to only using the protein as a film. With electrochemical instruments, Electrochemical Impedance Spectroscopy (EIS) and Potentiodynamic Polarization (PD), measurements were performed based on which the inhibition efficiency of protein films were calculated.
1. **CONCEPT**

In order to understand the difficulty in preventing corrosion one must first know about the material itself and in addition how corrosion in general occurs. A metal can behave in different ways and depending on how a metal acts it can be more or less susceptible to corrosion. The electrochemical cell responsible for the corrosion process will be described as well as a natural passive layer that, to some extent, slows down the corrosion process of certain metals.

1.1 **Metal behavior**

To understand why different metals are more or less exposed to corrosion one must know how the metal will behave in a corrosive environment. A metal can have passive, active or immune behavior. If a metal is *immune* it is thermodynamically stable and no corrosion will occur which is the case for noble metals. However, when a metal exhibits *active behavior*, corrosion occurs and the metal will start to dissolve. In order to slow down the process, some metals have the ability to form a passive layer. This is the *passive behavior* where a thin film consisting of corrosion products is formed on the metal surface. If the layer is damaged or dissolved then the exposed metal will once again become active and the corrosion rate will proceed. (3) (4)

1.2 **Corrosion in general**

The electrochemical cell illustrated in fig. 1 is the reason to why corrosion occurs. The main concept is that electrons are transported from the cathode where reduction occurs to the anode where oxidation takes place. The anode consists of a more active metal since it releases electrons whereas the cathode is a more noble metal in comparison. The electrolyte is a medium which complete the process by allowing transportation of ions. The result is corrosion at the anode which either releases cations to the solution or form oxides on the surface. The electrons released through the oxidation reactions are then transported to the cathode where reduction reactions take place. This limits the corrosion rate since the electrons produced at the anode is controlled by the electron consumption at the cathode. If the reduction process is slow then in turn the oxidation process will be retarded. As mentioned above, some metals can experience passive behavior. Anodic protection is often achieved through passivation of the metal where a potentiostat maintain the potential in the specific region. The oxide layer formed serves as a protection and hence decelerates the corrosion
process. By eliminating one of the objects required in order to create an electrochemical cell, corrosion would be prevented. The problem is how to complete such action. One could try to keep a surface completely dry which would make the ion transport insufficient but in turn the applications would be very limited. Another technique would be to prevent one of the reactions at the anode or cathode, resulting in no electron transport and in turn, no corrosion. Today there are several techniques to slow down the corrosion process which will be described later. For now, the understanding of the corrosion cell is necessary to understand why corrosion can appear in different forms. (3) (5)

![Figure 1 Illustration of electron flow in an electrochemical cell.](image)

### 1.3 Passive layer

The passive layer is a natural protection that covers the metal surface. When using coatings or paint the passive layer will still function as a protection when these are damaged. Depending on metals the layer can have different thickness and withstanding. The passive film can be described as a protective bi-layer. The first layer is a defective oxide barrier closest to the metal surface and the second layer consists of cations remitted from the metal combined with species from the environment. (6) The first layer can grow thicker if the rate of formation exceeds the rate of degradation.

A metal can switch from active to passive state which is best described with an anodic polarization curve where logarithmic current density is plotted against the potential. As fig. 2 illustrates, the metal first experience active state when the potential is low and the current density increases. When the potential increases and the current density decrease the passive state is reached. Here the current density is constant whilst the potential continually increases until transpassive state is initiated. The potential for which the transpassive region starts is
called critical anodic potential, $E_{bd}$ and in this region the metal dissolution increases. An anodic polarization curve only describes which state the metal is in. If combined with a cathodic polarization curve one can get an estimation of the Tafel coefficient and by that the corrosion rate. As illustrated in fig. 3, depending on where the cathodic polarization curves are on the anodic diagram it determines the outcome.

![Anodic polarization curve diagram]

**Figure 2 Illustrating an Anodic polarization curve describing different metal regions.**

The first cathodic slope a) is at the transpassive region where there is a breakdown of the passive layer and consequently, corrosion occurs. The second slope b) is at the passive region. Here the protective layer can repair itself and no corrosion is initiated. The last curve is c) where the metal is in an active state. A relatively high corrosion rate can be presumed since the metal is susceptible to corrosion when there is no passive layer to protect the surface. In order to protect the metal, the potential should not exceed the critical anode potential $E_{bd}$ or decrease lower than the $E_{pp}$ which is the lowest potential for the passive region. (7) (8)
1.4 Pourbaix diagram

In 1938 Marcel Pourbaix developed the potential – pH diagram which today is also known as the Pourbaix diagram. As illustrated in fig. 4 the diagram indicates regions where a metal undergoes different active and passive states. The vertical lines are for pH depending reactions whilst the horizontal lines are for potential depending reactions. In the corrosion region there are often ions that have been dissolved from the metal while in the passive area there are often solid forms of oxides or hydroxides. When the metal is remained in the immune region, no reaction takes place. Pourbaix diagrams are useful to estimate the passive region. The larger the region the better the metal withstands different environments i.e. it can be used for a wider range of applications. However, there are some limitations for Pourbaix diagrams. For example, equilibrium is assumed which may not be the actual condition. The corrosion rate is not given and the diagrams are only for single metals not for alloys. All oxides and hydroxides qualify as passive despite their protective properties. As with any other technique, it is a useful tool providing that the limitations are known. (9) (10)
2. **DIFFERENT TYPES OF CORROSION**

There are two main categories for corrosion; uniform and localized. The latter involves several sublevels which are illustrated by fig. 5. Uniform corrosion could be thought of as several local corrosion cells acting on the complete surface whereas localized corrosion only acts upon a small limited area. It is common that different sorts of corrosion exist on a metal at the same time since one type of corrosion can give rise to another. Different types of localized corrosion will be described more in detail below.

![Pourbaix diagram for iron](image)
2.1 Galvanic corrosion
When two metals with different potential are in contact with each other galvanic corrosion might occur. The noble metal works as a cathode whilst the active metal functions as an anode. The driving force is a potential gradient which induces an electron transport from the anode to the cathode. The result is corrosion at the active metal whereas the noble metal is protected. Since two metals are involved, the given corrosion rate is only for the particular system. If one metal were to be replaced by another, the corrosion rate would be different. With the help of galvanic series one can determine how noble the metals are and thereby predict corrosion. (11)

2.2 Erosion corrosion
This is both a mechanical and chemical corrosion where a moving fluid damages the passive layer. A materials ability to restore the passive layer determines its susceptibility to erosion corrosion. The velocity of the moving fluid is an important factor since a higher velocity causes the fluid itself to erode the metal without corrosion involved. At a lower velocity, suspended particles in the fluid remove protective film and corrosion products which in turn accelerate the corrosion process. (12)
2.3 Pitting corrosion
When defects are present in the passive layer, pitting corrosion can occur. While most of the surface is covered, the exposed area is attacked, which in turn results in a localized corrosion. The driving force is the same as for galvanic corrosion. The exposed area becomes the anode while the unexposed area functions as the cathode. A potential gradient causes anions to travel to the pits where the positive metal ions are. Reduction of oxygen takes place on the cathode surface which sometimes limits corrosion on the surface. The ion flow during pitting corrosion is illustrated in fig. 6. (13)

![Figure 6 Illustrating the ion flow in pitting corrosion.](image)

2.4 Crevice corrosion
Crevice corrosion, as the name suggests, occurs at narrow gaps between either two metals or a metal and a non metal. Within the crevice the electrolyte has a very low concentration of oxygen whilst the electrolyte surrounding the crevice has higher concentration. The result is a concentration gradient where the material within the crevice functions as an anode and the outer surface as a cathode. Inside the crevice the environment becomes very acidic whilst the outer surface can remain neutral. Similar to pitting, the metal ions within the crevice attract negatively charged ions such as chloride ions. When the metal chloride is hydrolyzed the result is hydroxide and hydrochloric acid. The acidic environment is believed to be propagating factor for crevice corrosion. (3)

2.5 Dealloying corrosion
This corrosion process occurs either by dissolving the entire alloy or by dissolving the component more susceptible to corrosion. The result is a porous metal which has
lost/weakened several mechanical properties. The potential difference between the metals determines which will function as an anode and which will function as a cathode i.e. being more susceptible for corrosion. (14)

2.6 Intergranular corrosion
Intergranular corrosion occurs on a microscopic level i.e. impossible to spot without instrument. The grain boundaries functions as anode and the material in between as cathode. Since the corrosive path is very narrow, there is a critical depth where transportation of oxygen and corroding components are impossible and thereby forces the process to halt. (15)

2.7 Stress corrosion cracking (SCC)
As for erosion, stress corrosion is a combination of mechanical and chemical actions. The stress could be applied external although most often, a very small force causes the cracks. Combined whit a corrosive environment, stress corrosion could be devastating. Since it appears on a microscopic level it is hard to detect which could result in failure for the intended purpose. (14)

3. PROTECTION TODAY
There exist numerous methods to slow down corrosion. For example one could select a different material more suited for the given environment. However, this is not always favorable depending on field of application, properties of the metal/alloy and financial reasons. A change of environment could be preferred in some cases, for instance lowering the velocity of a fluid could reduce the corrosion rate in a system. If none of these are preferential, there are other possibilities that will be discussed briefly below.

3.1 Coatings
A desirable coating should serve both for corrosion resistance and for the esthetics although this is not always possible. Good adhesion and impermeability are desired properties as well as toughness required to withstand external force. Coatings exist in many forms and can be either organic (polymer) or inorganic such as ceramic or metallic. It mainly works as a barrier which makes it more difficult for reactants to reach the metal but can also contain inhibitors creating an “active” protection which will be described later. A thicker coating can withstand more external damage and also increases the diffusion path i.e. it takes longer for the reactants
to reach the material. Today there are several coatings to choose from with different properties for different environments. One has to decide which coating is more suitable for the intended environment and purpose. (16)

3.2 Inhibitors
As for coatings, inhibitors depend on both the environment and the material. An inhibitor can either attach to the surface or react in the solution to remove harmful species. When attached to the surface it can either form a protective layer or interfere with the oxidation or the reduction process. There are several demands for a good inhibitor, for example it must be effective at low concentrations, functional for several pH values and temperatures, cost effective and non toxic. One could classify them in relation to the process for which it controls. Anodic inhibitors reduce the amount of metal ions which are transferred to the electrolyte. Cathodic inhibitors suppress the reactions at the cathode by interfering with oxygen reduction. Mixed inhibitors have the ability to suppress both reactions. (17)

3.3 Cathodic protection
Electrons are supplied to the metal through an external source which turns the metal to a cathode. By electrically connecting a sacrificial anode to the metal desired to protect, the first mentioned will oxidize resulting in protection of the desired metal. Another method is to apply a low direct current to the metal where the anode is inert which is more suitable for larger projects. (18)

3.4 Anodic protection
The method is similar to cathodic protection except that the current travels in the opposite direction. The metal is an anode and the electrode potential is adjusted so that it remains in its passive state. This requires great monitoring since it can otherwise increase the corrosion rate if the electrode potential is not confined within the passive region. If the metal becomes too positive, by the loss of electrons, it ends up in a transpassive state. (19)

4. MUSSEL ADHESIVE PROTEIN (MAP)
It is a well-known fact that blue mussels have the ability to adhere to several surfaces as for instance stones, wood and even ship hulls. Also their ability to stick to manmade objects as for instance glass and plastics imply that they have a broad application area. Their remarkable
adhesive property is of great interest in the surface chemistry department and, in the research for green inhibitors, a protein from blue mussels has proven very effective for corrosion protection. In this part, blue mussels will be described briefly and a more detailed description of the protein used in laboratory studies will be given later.

4.1 Green inhibitors

Today we have a completely different outlook on environment friendly approaches compared to several decades ago. We are getting more aware that our actions have consequences and should therefore try to think several steps ahead before introducing new materials. In corrosion science, studies are now made on different plants and animals that might possess properties that could be very useful in surface chemistry. One example is the study on caffeine. According to Antony et al (20) caffeine itself does not support sufficient inhibition but in a complex with Zinc (II) the inhibition efficiency is significantly increased. Many other studies have been made and can be reviewed in short in appendix B. The theory is that since the material would be produced by nature instead of man, it should be environmentally friendly and nontoxic. The problems at hand are that it requires a lot of research which takes time and costs money. Once a suitable material has been found, a way to synthesize it must be developed. This method should be cost-efficient and during the synthesis no environmental harmful material should be used. If the price is wrong, companies in need of the product will probably choose another material. However, human beings can create environmentally friendly materials and also use material from nature without disrupting the fauna. The blue mussel is evidence on the latter mentioned. There is no need to synthesize the protein retrieved from the blue mussels due to an excess of the animal and the possibility of mussel cultivation. On the coasts of Sweden studies are made, funded by the European Union, in order to see if blue mussels could be used as small treatment plants. Several mussel cultivations are distributed and studies have revealed that a blue mussel with a size of 3 cm filters two to three liters of water each hour. When harvesting one ton of blue mussels, approximately 10 kg of nitrogen and 1 kg of phosphorus has been removed from the water. I.e. mussel cultivation brings water treatment at the same time as new mussels are produced. (21)
4.2 The byssus
Adherent properties that blue mussel possesses were found in the byssus which contains several strong threads that enable the mussel to adhere to surfaces. This is illustrated in fig. 7 where the byssus contains plaques, threads and a stem. The adhesive plaques are located at the end of the threads which in turn are connected to the stem and the foot. The initial contact to a surface starts with the foot leaving the shell and connects to the surface. The foot contains a groove that, when pushed down on a surface, it seals from both air and water which enables a good grip on the surface. When the connection is made, a byssus will start to form and the foot is retracted. If the mussel starts to travel on the surface, the byssus is retracted and the foot is extended again and the process starts over. The threads contain several proteins which enables the mussel to stay on the surface when the foot no longer has a grip to it. (22) For this work, the protein of interest can be found on the surface of plaque and threads and functions as a protective coating from sand. This protein is called Mefp-1 and will be described in greater detail below.

![Figure 7 The byssus adheres to the surface while the foot is retracted inside the shell. (22)](image)

4.3 Protein Mefp-1
As mentioned before, there are several proteins that work together to construct the byssus. One of the differences discovered between the proteins is the deviating amount of DOPA. The protein in focus, which also will be used in the experimental studies, is Mefp-1 with a DOPA content of ca. 13mol%. (2) As illustrated in fig. 8 the protein is located at the surface of the threads and plaque. Mefp-1 stands for *Mytilus edulis* foot protein and contains several amino acids which in water have positive charge. Combined with high levels of DOPA this protein is
thought to enable the cross-linking with iron ions and thus, resulting in very good adhesive properties.

DOPA, the 3,4-dihydroxyphenylalanine is a small molecule and binds to surfaces through its amide and carboxyl group. Compared to other amino acids it can be easily oxidized and binds metal ions well which is thought to be the reason to its cross-linking and complexation abilities. Since DOPA also has the ability to bond to metals, iron ions from the surroundings (i.e. water) may induce metal – protein complexes. Iron ions have the ability to bind up to three DOPA molecules. The resulting structure is illustrated in fig. 9 where the protein chains are excluded for better illustration of DOPA. However, due to the several DOPA molecules on the protein, there is no certainty that there are three different protein chains. Although that is the wanted result, there might actually be only one protein chain with its DOPA groups bonded to the iron ion. The cross-linking is an important factor since it generates cohesive strength between the proteins which solidifies the material. DOPA is also thought to be responsible for the adhesive properties to the surface as it suppress the water from the contact area. Although the mechanism is not fully understood, it is believed that DOPAs ability to form covalent- or hydrogen bonds is the reason for mussels’ ability to adhere to surfaces. Without the adhesive bonding the protein would not be able to stick to the surface, and without the cohesive bonding the threads would not withstand the tension created in order to keep the mussel on a surface. Only cohesive force would result in the protein bonding to itself but not to a surface. If there was only an adhesive force the result would be a thin layer
adhering to the surface without the mussel connected to it since no cohesive strength exist to bind the proteins together. (24) (23)

![Figure 9 Metal-DOPA complex](image)

DOPA is an important functional group for mussel adhesion which is why Mefp-1 was selected for the experimental work. The protein Mefp-1 containing high levels of DOPA creates a very attractive protein for corrosion protection especially in the search for green inhibitors. There are a lot of studies made on the subject, trying to figure out the mechanism behind mussel adhesion. For instance, the company Biopolymer Technology of Sweden AB is researching applications for biomedicine for example using MAP based glue for biomedical purpose. Its affiliated company Biopolymer Products of Sweden AB is carrying out research for surface protection on metals in collaboration with the department of corrosion science at the Royal Institute of Technology (KTH). (25) In order to use the protein in the real applications, studies must first reveal that the protein has the desired properties. Better understanding of the proteins properties will be provided by using the experimental techniques described in section 5.

### 5. EXPERIMENTAL TECHNIQUES

For the experimental studies Potentiodynamic polarization (PD) and electrochemical impedance spectroscopy (EIS) techniques were used to determine the carbon steel resistance to corrosion before and after the adsorption of Mefp-1 on the surface, respectively. This section will describe the two techniques in detail and the results of the measurements will be handled in section 6.
5.1 Electrochemical cell used for measurements

This section explains the electrochemical cell used for both techniques described later in this chapter. The electrochemical cell contains a small circular hole in the middle. By applying a rubber ring above the hole one can adjust the contact area to which the metal is exposed on and it also seals the edges so no electrolyte can leak. A plate of carbon steel used as working electrode (we) is placed on top of the rubber ring and the exposed area for this work is approximately $1 \text{ cm}^2$. The counter electrode (ce) consists of platinum and the reference electrode (re) is silver/silver chloride electrode that is saturated with potassium chloride. The net reaction is:

$$\text{Ag} + \text{Cl}^- \leftrightarrow \text{AgCl} + e^-$$

The lid is used to keep the electrodes separate and to some extent to prevent the solution from evaporating.

![Figure 10 An illustration of a electrochemical cell used for measurements.](image)

5.2 Electrochemical impedance spectroscopy (EIS)

Electrochemical impedance spectroscopy is a very useful instrument since it is both quick and nondestructive. Resistance - a measurement of a material’s ability to resist current flow is described by Ohm’s law:

$$R = \frac{E}{I}$$

where $R$ is the resistance, $E$ the voltage and $I$ the current. Unfortunately it is restricted in use since the resistor needs to follow Ohm’s law at all levels of current and voltage and it should be independent of frequency. This is not always the case why impedance is a better unit to
As resistance, impedance is also a measurement of a material’s ability to resist current flow, but it does not have the limitations that resistance measurements experience. The potential applied is a small AC signal with amplitude of 5 to 10 mV. If a sinus wave potential is applied, a sinus wave current will be the responding signal. Fig. 11 illustrates an AC (alternating current) potential applied to an electrochemical cell and the response is an AC current although shifted in phase.

The applied voltage as a function of time can be written as:

\[
E_t = E_0 \sin(\omega t)
\]  
(2)

where \(E_t\) is the potential at the time \(t\), \(E_0\) is the amplitude and \(\omega\) the radial frequency. The latter can in turn be expressed as:

\[
\omega = 2\pi f
\]  
(3)

where \(f\) is the frequency.

The responding signal, i.e. the sinus wave of the current is described as:

\[
I_t = I_0 \sin(\omega t + \phi)
\]  
(4)

where \(I_t\) is the responding signal at the time, \(I_0\) is the amplitude and \(\phi\) is the phase shift.

Recall the equation for Ohm’s law (1), the expression for impedance is quite similar. It is a ratio between potential and current although the expression is more complex.

\[
Z = \frac{E_t}{I_t} = \frac{E_0 \sin(\omega t)}{I_0 \sin(\omega t + \phi)} = Z_0 \frac{\sin(\omega t)}{\sin(\omega t + \phi)}
\]  
(5)

By using Euler’s relationship below, the impedance can be expressed as a complex function:

\[
\exp(i\phi) = \cos(\phi) + i\sin(\phi)
\]  
(6)

If this equation is applied to equation (2) and (4) the resulting expression for impedance is:

\[
Z(\omega) = \frac{E}{I} = Z_0 \exp(i\phi) = Z_0(\cos(\phi) + i\sin(\phi))
\]  
(7)
The equation contains one imaginary part and one real part. By plotting the real part on the x-axis and the imaginary part on the y-axis a Nyquist plot is derived. This is illustrated in fig. 12 where each point represents the impedance at one frequency. The y-axis is negative and the frequency increases from left to right. The vector represents the impedance and the angle between the vector and the x-axis is $\phi$. Unfortunately the Nyquist plot does not reveal which frequency is used at different points, to retrieve that data, a Bode plot is utilized.

![Nyquist plot](image)

Figure 12 Illustrating the Nyquist plot where Rs and Rp are solution resistance and polarizations resistance respectively. (26)

In a Bode plot, the logarithmic frequency is plotted on the x-axis and both the absolute value of impedance and phase shift on the y-axis. This is illustrated with two plots in fig. 13. In order to interpret the data it is necessary to match it to an equivalent electrical circuit which in turn describes the chemical phenomenon related to the system. Most often it is a combination of different circuits that describe the system. In order to retrieve the polarization resistance one subtracts the lowest value on the y-axis from the highest value.
A few examples of the different electrical circuits to match with are:

Impedance of a resistor: \( Z = R \) independent of frequency

Impedance of capacitor: \( Z = \frac{1}{i\omega C} \) phase shift \(-90^\circ\)

Impedance of inductor: \( Z = i\omega L \) phase shift \(90^\circ\)

The impedance of a resistor contains only a real component. From the equations above, one can see that if the frequency increases, the impedance of an inductor increases whilst the impedance for a capacitor decreases. Both inductor and capacitors only have imaginary components. Not only is the system often described by different circuits but also if they are serial or parallel coupled. If there is a serial combination the impedance is described by:

\[ Z = Z_1 + Z_2 \]  \hspace{1cm} (8)

If there is a parallel combination then the impedance can be written as:

\[ Z = \frac{1}{\frac{1}{Z_1} + \frac{1}{Z_2}} \]  \hspace{1cm} (9)

Several conditions affect the impedance as for example diffusion, double layer capacitance, resistance in electrolyte et al. One of the most common cell models is called a Randles cell illustrated in fig. 14. It includes a double layer capacitor, solution resistance and a polarization resistance (or charge transfer resistance). On the Nyquist plot, the solution resistance can be found at high frequencies at the real axis. Since the sum of polarization and solution resistance can be found at low frequencies the diameter of the semicircle equals the
polarization resistance. I.e. a larger diameter on the Nyquist plot illustrates great polarization resistance. The electrical double layer exists on the surface of an electronic conductor surrounded by an electrolyte. It originates from ions in the solution that attracts to the electrode surface hence shielding the electrode. This is called the inner Helmholtz plane (IHP) which is then surrounded by an outer Helmholtz plane (OHP), i.e. a layer of ions with opposite charge to the first layer. As a result the electrical double layer functions as a capacitor and, in series with the electrolyte functioning as a resistor, is thereby easy for EIS to calculate. The problem with EIS is to find the right model that is suitable for the results, given that several systems can compose a perfect fit.

![Figure 14 Randles cell where RS is the solution resistance, Cdl the double layer capacitance and Rp the polarization resistance.](image)

5.3 Potentiodynamic polarization (PD)

Whilst EIS measurements can be performed several times, potentiodynamic polarization alters the surface only allowing for one test. The system consists of a potentiostat and three electrodes. The potentiostat is able to control the potential as well as measuring the current resulting from the system. The three electrodes are reference electrode, working electrode and counter electrode (often platinum or graphite). The corrosion rate is determined by both anodic and cathodic reaction. The anodic reaction results in oxidation i.e. electrons are released into the metal. The cathodic reaction affects the solution through reduction which results in electrons being removed from the metal. During equilibrium, there is no net flow of electrons and hence no electrical current. Consider the graph illustrated in fig. 15 where potential is plotted on the y-axis and logarithmic current on the x-axis. The filled line illustrates the current that is measured and the sharp point is where the current change direction.
There is no way to measure the corrosion current directly nevertheless, the Tafel equation (eq. 10) can be used to approximate it:

\[ I = I_0 e^{(2.3(E-E^0)/\beta)} \]  

(10)

where \( I \) is the resulting current from the reaction, \( I_0 \) the exchange current, \( E \) the electrode potential, \( E^0 \) the equilibrium potential and \( \beta \) the Tafel constant for the reaction. However, this reaction only describes one isolated reaction why the Butler-Volmer reaction below is used to describe both the anodic and cathodic reaction.

\[ I = I_a + I_c = I_{corr} e^{(2.3(E-E_{oc})/\beta_a)} - e^{(-2.3(E-E_{oc})/\beta_c)} \]  

(11)

where \( I \) is the measured current, \( I_{corr} \) the corrosion current, \( E_{oc} \) the corrosion potential and \( \beta_a \) and \( \beta_c \) are Tafel constants for anodic and cathodic polarization respectively. The first exponential describes the metal dissolution i.e. the anodic reaction, and the latter the metal deposition i.e. the cathodic reaction. At the corrosion potential, the exponential terms equals one and hence, the cell current is zero. Moving away from the corrosion potential results to either anodic or cathodic dominants and consequently, one of the exponential terms can be ignored. If the logarithmic current is plotted versus the potential the result is a straight line as illustrated in fig. 15. By drawing linear slopes for both the anodic and cathodic curves results in an intersect from where the corrosion current can be approximated. (26)

A lot of techniques have limitations why one must know the boundaries for the instrument in use. In the list below, assumptions and complications when using the polarization technique are listed.

Complications:

- Passivation from oxides may alter the surface i.e. might have other constants than the original surface.
× If one component in an alloy is more likely to dissolve than the other, there will be changes in the surface.
× If more than one cathodic or anodic reaction occurs simultaneously, for instance reduction of oxygen and hydrogen ions, there will be difficulties.
× The solution resistance cannot be too high since it might affect the kinetic model.

Assumptions:
× The values of the Tafel constants are known.
× The material exhibits uniform corrosion.
× There is only one anodic and cathodic reaction.
× The solution resistance is negligible i.e. solutions should have very high conductivity.

Potentiodynamic polarization is relatively fast, accurate and inexpensive hence, attractive equipment for corrosion studies. However, the technique alters the surface i.e. it is only possible to perform the measurement at the end of the testing period.

6. EXPERIMENTAL

The results were divided in to four different parts where the film deposition has been altered. The experimental techniques were the same for all parts and every test was repeated three times. The metal used for the experimental studies was carbon steel due to its broad field of applications not least in structures. (27) The protein used was pure Mefp-1 which was provided by Biopolymer AB (Gothenburg, Sweden). The carbon steel samples were wet ground with SiC grinding paper successively from 120 (roughest) to 1200 (smoothest) grits in order to receive a relatively smooth surface and to remove organic and corrosive particles. During the polishing process the sample was kept in isopropyl in order to prevent corrosion of the sample. Afterwards an ultrasonic cleaning is performed for 5 minutes in order to remove organic contaminant that was adhering to the surface. Polished carbon steel samples were then kept in a sodium hydroxide solution at pH 14 in order to form a relatively protective oxide layer. Before immersing the carbon steel into the protein solution the metal was rinsed with ethanol to remove the sodium hydroxide. The _Mefp-1_ solution was delivered as a 27 mg/ml aqueous solution containing 1 wt % citric acid and stored as received in the dark at +4 °C before use. A solution containing 1wt % citric acid, at pH 9-10 was prepared as the buffer solution, and the pH was adjusted by sodium hydroxide solution. Just 3 minutes prior to measurements, _Mefp-1_ was added from its stock solution to the buffer solution to give a concentration of 0.1 mg/ml. The protein film was formed on the steel surface by immersing
the carbon steel into the Mefp-1 solution for 30 minutes. After film deposition a gentle stream of nitrogen gas was used for surface drying. The electrochemical cell was then assembled and the sodium chloride solution with the concentration of approximately 0.1 M was used as electrolyte. All the samples had a contact area of approximately 1 cm² to the electrolyte.

To interpret the EIS data an equivalent circuit was used for curves fitting by employing the software Zview. The equivalent circuit that fitted the data was very similar to Randles cell although, instead of a double layer capacitance there was a constant phase element (CPE). CPE gave a better approximation due to capacitive response not always being ideal at the steel/solution interface. The equivalent circuit is illustrated in fig. 16 where \( R_s \) is the solution resistance and \( R_p \) is the polarization resistance. The instrument used to perform the measurements was a Solartron electrochemical interface 1287 coupled with a Solartron frequency response analyzer 1250. The frequency range for EIS measurements was from \( 10^4 \) to \( 10^2 \) Hz with a perturbation amplitude of 10 mV. All measurements were carried out at room temperature.

![Equivalent Circuit](image)

**Figure 16** Illustrates the equivalent circuit used to describe the interface of the carbon steel in order to interpret data from EIS measurements.

At the last day of the exposure potentiodynamic polarization was performed with a scan rate of 10 mV/min from the cathodic to the anodic direction. In order to analysis data from potentiodynamic polarization, Tafel curve fitting was performed with the Corrview software. The corrosion current was calculated using curve fitting and eq. 11. By using the Levenburg – Marquardt (LEV) method in the Corrview software the values for \( i_{corr} \), \( E_{corr} \) and the Tafel coefficients were determined and the error was minimized:

\[
\chi^2 = \sum_{i=1}^{n} \left[ \frac{l_i - l_i'}{\sigma_i} \right]^2
\]

(12)

where \( l_i \) is the current at data point \( i \), \( l_i' \) the calculated current and \( \sigma_i \) the weighting factor. Since the Tafel coefficients are fitted in this equation as well, the error from using the default Stern – Gary constant is eliminated.
6.1 Results – Fe³⁺ effect on pre-adsorbed Mefp-1 film

Description:
A polished carbon steel specimen was directly used for the control test. A carbon steel sample was immersed into the Mefp-1 solution at pH about 9.5 for 30 minutes to form a Mefp-1 film on the surface. In order to form a complexed protein film, another carbon steel sample was alternately immersed into the Mefp-1 solution and 10 µM FeCl₃ solution at pH 9 respectively. The immersion time for each solution was 10 minutes and the process was repeated three times. Consequently three different surfaces were formed:
- one bare carbon steel surface (referred as control sample)
- one with the pre-formed Mefp-1 film (referred as Mefp-1 sample)
- one with the Fe³⁺ complexed Mefp-1 film. (referred as Mefp-1/Fe sample)

Results of EIS measurements
Fig. 17 below illustrates the Nyquist and Bode plots for three control samples. As mentioned in section 5.2, the wider the diameter of the semicircle the better the polarization resistance. The Nyquist plot reveals that the polarization resistance increased with longer exposure time. During the immersion to the NaCl solution corrosion products were formed, creating a protective layer on the surface of the carbon steel. The layer continued to grow throughout the measuring period which explains the appearance of the curves. The Bode plot of modulus provides similar information as the Nyquist plot however, it also reveals specific frequencies for which the measurement data were detected, which are not presented in the latter. The Bode plot of phase angle revealed that there was only one time constant, therefore the equivalent circuit in fig. 16 was used for the experiments.
Illustrated in fig. 18 is the carbon steel coated with the Mefp-1 film. After 30 minutes of exposure to NaCl solution a very high polarization resistance was observed which after 3 hours decreased remarkably. This is probably due to the pre-adsorbed protein film being broken down. Afterwards the polarization resistance starts to increase although with Mefp-1 it does so very assembled i.e. compared to control samples, there is no large variation in polarization resistance after longer exposure times. This stability could be the result of the protein covering the surface which makes it more uniform, i.e. the “invisible defects” from the polishing process become unapparent. The increase in polarization resistance was most likely due to corrosion products and/or iron ions released from the dissolving metal complex the protein. The Bode plot of the phase angle revealed that there was only one time constant therefore the equivalent circuit in fig. 16 was used for the data fitting presented later.
Figure 18 Illustrates the EIS spectra for carbon steel coated with Mefp-1 in 0.1 M NaCl solution at pH 10 during one measurement period of 3 days in total.

The results for Mefp-1/Fe 10 μM are illustrated in fig. 19. Compared to control and Mefp-1 samples, the 30 minutes exposure time does not present a high polarization resistance i.e. the surface is slightly more stable, missing the dramatic decrease at short exposure time. Instead the samples start with relatively low corrosion resistance and then increases with increased exposure time. However, after one day the increase slows down i.e. the fluctuation in polarization resistance is very small. Samples coated with Mefp-1 presented the same behavior that is, the protein created a more uniform surface and the polarization resistance became more stable. Nevertheless the fluctuation was much smaller for Mefp-1/Fe which could be due to the amount of iron which created a denser surface by complexation.

Figure 19 Illustrates the EIS spectra for carbon steel coated with Mefp-1/Fe 10 μM in 0.1 M NaCl solution at pH 10 during one measurement period of 3 days in total.
**Summary – Fe$^{3+}$ effect on pre-adsorbed Mefp-1 film**

Compared to control samples, both Mefp-1 and Mefp-1/Fe samples revealed much more stable behavior after longer exposure time. The results from task 1 can be found in table 1. As mentioned in the beginning of section 6, there is a specific electric equivalent circuit used to fit the data obtained from EIS measurements. The fit factor n is associated with the interface heterogeneity, i.e. the larger the deviation from 1 the more heterogeneous is the interface. The inhibition efficiency, $\eta_z$, is calculated using eq. (21) below:

$$\eta_z = \frac{R_p^{(inhib)} - R_p}{R_p^{(inhib)}} \times 100\%$$  \hspace{1cm} (21)

Although it is called the inhibition efficiency it is more a measure of the change in corrosion resistance. A higher value indicates a better protection. For instance, Mefp-1 samples reveal that the protein does increase the corrosion resistance. However, the longer the exposure time the less effective the protein film becomes. Mefp-1/Fe complex film also exhibit protection although it varies over time. It might be due to an excess amount of iron ions on the surface which dissolves instead of forming a complex with the protein. However, the film is also very thin which might have caused lower polarization resistance than the oxide layer at the beginning of the measuring period. From table 1 it was noted after 3 hours of exposure that Mefp-1/Fe exhibits a lower capacitance, Y0, and a higher n value combined with a higher polarization resistance. The fit parameter, exponential factor n, is often associated to the degree of heterogeneity of the interface and/or surface film hence, the larger the deviation from 1, the more heterogeneous surface layer. (28) The results indicated that the film was denser than samples coated with only Mefp-1 film. The inhibition efficiency in fig. 20 is consistent with those results.

![Figure 20 Illustrating the inhibition efficiency over time for Mefp-1 and Mefp-1/Fe in 0.1 M NaCl.](attachment:image.png)
Table 1 reveals a large deflection at 30 minutes of exposure which might be due to an initial oxide layer formed during the pre-treatment process which was later broken down. Since this was more or less present in the samples the standard deviation was very high. Since relatively rough grinding papers were used in the polishing process, the surfaces might have different irregularities. I.e. there are scratches, metal inclusions, which was invisible for the naked eye. Compared to control samples, the standard deviation is much lower for both Mefp-1 film and Mefp-1/Fe film. This might be due to the protein covering the surface which reduced the polishing factor and to some extent blocked the dissolution of the carbon steel.

Since the polarization resistance technique damages the surface of the sample the measurement was carried out at the last day of every test period. Fig. 21 show polarization curves for all the samples after three days of exposure to 0.1 M NaCl. Mefp-1 sample clearly illustrates higher corrosion potential than control sample consistent with EIS measurements. However, Mefp-1/Fe complex and control sample are hard to distinguish without calculations.
Figure 21 Illustrates the potentiodynamic polarization curves for control, Mefp-1 and Mefp-1/Fe 10 μM in 0.1 M NaCl at pH 10 after three days of exposure.

In order to obtain the corrosion rate from polarization curves the Levenburg – Marquardt model was used for curve fitting. In this way, \( b_a \) and \( b_c \) are also fitted quantities hence, the error by using the default Stern – Gary constant in the software is avoided. This is particularly important for measurements of aged samples. (29) The average results can be viewed in table 2, where \( E_{corr} \) is the corrosion potential, \( I_{corr} \) the corrosion current, \( S_G \) the Stern – Gary constant and \( b_a \) and \( b_c \) the Tafel coefficients. \( \eta_p \) is the inhibition efficiency which reveals the reduction in corrosion rate. It is calculated with the equation:

\[
\eta_p = \left(1 - \frac{i_{inh}b}{i_{corr}}\right) \cdot 100\%
\]

Table 2 Results obtained from data fitting of polarization measurements.

<table>
<thead>
<tr>
<th>Solution</th>
<th>( E_{corr} ) vs. SCE (mV)</th>
<th>( b_a )</th>
<th>( b_c )</th>
<th>( S_G )</th>
<th>( I_{corr} ) (μA/cm²)</th>
<th>( \eta_p ) (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control pH 10</td>
<td>-674</td>
<td>75</td>
<td>245</td>
<td>24</td>
<td>2.5 ± 1.055</td>
<td>-</td>
</tr>
<tr>
<td>Mefp-1</td>
<td>-647</td>
<td>71</td>
<td>250</td>
<td>24</td>
<td>3.3 ± 0.719</td>
<td>-31</td>
</tr>
<tr>
<td>Mefp-1/Fe 10 μM</td>
<td>-699</td>
<td>82</td>
<td>285</td>
<td>26</td>
<td>2.4 ± 0.986</td>
<td>3</td>
</tr>
</tbody>
</table>

From table 2 it is apparent that Mefp-1/Fe sample shows a lower corrosion current density than that of the Mefp-1 sample. Recall from description of task 1 above, the carbon steel sample was first immersed into Mefp-1 for ten minutes and afterwards dipped in FeCl₃ and the alternating immersion were repeated three times. This might have resulted in several layers of the films which also could make up for the protection presented. Mefp-1 experienced an increase of the corrosion rate with approximately 31%, hence iron complexed Mefp-1 film did improve the protection with nearly 35%
6.2 Results – creating an iron-protein complex before film formation.

Description:
The Mefp-1 solution used was the same as in Part 1, however the concentration of FeCl₃ solution was decreased to 1 μM and 0.1 μM in order to study the relationship between the concentration of iron ions and DOPA content. Different from previous samples, Mefp-1 and FeCl₃ solution were mixed before the film adsorption. The test will reveal if there was a difference between complexation of the Mefp-1 before and after film formation, as well as the concentration dependence of iron ions for the Mefp-1 complexation. The samples were then compared with samples retrieved from previous measurements.

Results of EIS measurements
The iron ions concentration was only a tenth of the concentration used in task 1 and the results can be viewed in fig. 22. As illustrated, the polarization resistance was very stable within 3 days of exposure. These results are consistent with Mefp-1 and Mefp-1/Fe 10μM, hence it is reasonable to conclude that Fe³⁺ complexed protein creates a more uniform surface which stabilizes the polarization resistance of the carbon steel.

Figure 22 Illustrates the EIS spectra for carbon steel coated with Mefp-1/Fe 1 μM in 0.1 M NaCl solution at pH 10 during one measurement period of 3 days in total.

The concentration of Fe³⁺ for this test was very low in order to obtain the critical amount of iron ions needed for a sufficient protein complexation. Too large amount of Fe³⁺ might induce desorption of Mefp-1 from steel surface and in turn, the coverage of the film will not be satisfactory. However, too small amount of Fe³⁺ might not create fully complexation of Mefp-1 and in turn, the film will not be dense enough. Results are illustrated in fig. 23 which reveals
a steady increase of the polarization resistance, although values were lower compared to samples with 1 µM Fe³⁺. This might be the result of insufficient complexation of the Mefp-1 and in turn, less dense surface film. The steady increase might be the result of corrosion products and/or increased complexation with iron ions dissolved from the steel surface.

Summary – Fe³⁺ induced Mefp-1 complexation in the bulk solution

Fig. 24 illustrates the inhibition efficiency for Mefp-1/Fe 0.1 µM and 1 µM respectively. Similar to Mefp-1/Fe 10 µM, none of these samples experienced as good inhibition efficiency as Mefp-1 alone after 30 minutes of exposure to 0.1 M NaCl. However, both Mefp-1/Fe 0.1 µM and Mefp-1/Fe 1 µM respectively exhibited an increase of the inhibition efficiency within one day of exposure, probably due to cooperation of corrosion products with the protein film. At the third day of exposure both samples displayed same behavior as Mefp-1/Fe 10 µM which shown a decrease in the inhibition efficiency probably the result of film degradation.
Figure 24 Illustrates the inhibition efficiency over time for Mefp-1/Fe 0.1 μM and Mefp-1/Fe 1 μM respectively in 0.1 M NaCl at pH 10.

The results retrieved from data fitting can be viewed in table 3. Comparing the results with previous samples suggested that the Mefp-1/Fe 1 μM had denser film than Mefp-1 film alone. For Mefp-1/Fe 0.1 μM no such conclusions could be drawn which could be the result of too small amount of Fe3+ not fully creating a complexation with the protein. The higher inhibition efficiency was therefore probably due to corrosion products and/or that iron ions released from the dissolving metal complex the protein.

Table 3 Results obtained from data fitting of EIS spectra.

<table>
<thead>
<tr>
<th>Solution</th>
<th>Time</th>
<th>$R_s$ (Ω cm$^2$)</th>
<th>$Y_0$ (Ω$^{-1}$ cm$^2$)</th>
<th>n</th>
<th>$R_p$ (Ω cm$^2$)</th>
<th>$n_z$ (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control NaCl</td>
<td>30 min</td>
<td>142</td>
<td>4.0 x 10^{-4}</td>
<td>0.9</td>
<td>(3.0 ± 0.85) x 10^{3}</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>157</td>
<td>7.1 x 10^{-4}</td>
<td>0.8</td>
<td>(2.3 ± 0.60) x 10^{3}</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>145</td>
<td>7.4 x 10^{-4}</td>
<td>0.8</td>
<td>(2.5 ± 0.30) x 10^{3}</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>142</td>
<td>7.4 x 10^{-4}</td>
<td>0.8</td>
<td>(3.3 ± 0.57) x 10^{3}</td>
<td>-</td>
</tr>
<tr>
<td>Mefp-1/Fe 1 μM</td>
<td>30 min</td>
<td>133</td>
<td>6.7 x 10^{-4}</td>
<td>0.8</td>
<td>(3.5 ± 0.41) x 10^{3}</td>
<td>13</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>129</td>
<td>1.1 x 10^{-3}</td>
<td>0.8</td>
<td>(2.7 ± 0.50) x 10^{3}</td>
<td>14</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>127</td>
<td>1.1 x 10^{-3}</td>
<td>0.8</td>
<td>(3.3 ± 0.18) x 10^{3}</td>
<td>24</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>126</td>
<td>1.1 x 10^{-3}</td>
<td>0.8</td>
<td>(3.0 ± 0.34) x 10^{3}</td>
<td>24</td>
</tr>
<tr>
<td>Mefp-1/Fe 0.1 μM</td>
<td>30 min</td>
<td>135</td>
<td>1.0 x 10^{-3}</td>
<td>0.8</td>
<td>(2.8 ± 0.56) x 10^{3}</td>
<td>-9</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>134</td>
<td>1.3 x 10^{-3}</td>
<td>0.7</td>
<td>(2.7 ± 0.24) x 10^{3}</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>139</td>
<td>1.2 x 10^{-3}</td>
<td>0.7</td>
<td>(3.3 ± 0.32) x 10^{3}</td>
<td>24</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>135</td>
<td>1.2 x 10^{-3}</td>
<td>0.7</td>
<td>(3.8 ± 0.39) x 10^{3}</td>
<td>13</td>
</tr>
</tbody>
</table>

Compared to Mefp-1/Fe 10 μM the corrosion resistance is higher for Mefp-1/Fe 1 μM and Mefp-1/Fe 0.1 μM. As mentioned above, too high concentration of FeCl$_3$ induces formation of di- or mono- DOPA/Fe$^{3+}$ complex and hence, the film will become less compact. (30) The constant increase in the polarization resistance could be due to corrosion products that
complement the surface protection or the further complexation of the Mefp-1 film with surface released iron ions.

Fig. 25 illustrates the polarization curves of the potentiodynamic measurements for all three samples. It is evident that Mefp-1/Fe 0.1μM had low corrosion current resulting in improved protection compared to only the protein film covering the surface. For Mefp-1/Fe 1μM the results are similar to data obtained from EIS measurements above i.e. the relatively high corrosion current results in poor protection.

Figure 25 Illustrates the potentiodynamic polarization curves for control, Mefp-1/Fe 1 μM and Mefp-1/Fe 0.1 μM in 0.1 M NaCl at pH 10 after three days of exposure.

Table 4 contains results obtained from the polarization measurements. These results are coherent with previous data obtained from EIS measurements. Mefp-1/Fe 0.1 μM has an inhibition efficiency of 26 % while Mefp-1/Fe 1 μM produced no corrosion protection. Considering that Mefp-1/Fe 0.1 μM has a less dense surface film, there are probably corrosion products causing this positive effect. The standard deviation is less than 0.5 % which is a very promising result for potentiodynamic measurements.

Table 4 Results obtained from data fitting of polarization measurements

<table>
<thead>
<tr>
<th>Solution</th>
<th>$E_{corr}$ vs. SCE (mV)</th>
<th>$b_a$</th>
<th>$b_c$</th>
<th>SG</th>
<th>$I_{corr}$ (μA/cm²)</th>
<th>$\eta_p$ (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control pH 10</td>
<td>-674</td>
<td>75</td>
<td>245</td>
<td>24</td>
<td>2.5 ± 1.055</td>
<td>-</td>
</tr>
<tr>
<td>Mefp-1/Fe 1 μM</td>
<td>-642</td>
<td>76</td>
<td>244</td>
<td>25</td>
<td>2.6 ± 1.105</td>
<td>-4</td>
</tr>
<tr>
<td>Mefp-1/Fe 0.1 μM</td>
<td>-700</td>
<td>76</td>
<td>190</td>
<td>24</td>
<td>1.8 ± 0.009</td>
<td>26</td>
</tr>
</tbody>
</table>
6.3 Results – creating ceria (IV) -protein complex before film deposition

Description:
For this test Ce(NH₄)₂(NO₃)₆ was used instead of FeCl₃ in order to create complexes before film deposition. The thought was that ceria (IV) should be able to create bigger Me fp-1 complexes than iron(III). Consequently, this would also result in a denser film and therefore greater protection. The concentration of ceria (IV) used was 5 μM mixed with 0.1 mg/ml Me fp-1 solution. Similar to task 2, the complexes were formed in the bulk solution first and afterwards the protein film was adsorbed on the carbon steel surface.

Results of EIS measurements
The EIS results for Me fp-1/ceria (IV) film were illustrated in fig. 26. The polarization resistance was relatively stable throughout the measurement period. Similar observations were noticed in previous samples with the protein suggesting a more uniform surface than control sample and hence, a more stable polarization resistance. However, the variation in polarization resistance was much smaller for Me fp-1/ceria (IV) which could be due to ceria (IV) creating a denser surface by complexation. Me fp-1/Ce⁴⁺ complex also have the possibility to be further complexed with iron ions dissolved from the metal. However, this could also desorb the protein film if the degree of the complexation is too high.

![Figure 26 Illustrates the EIS spectra for carbon steel coated with Me fp-1/Ceria (IV) 5 μM in 0.1 M NaCl solution at pH 10 during one measurement period of 3days in total](image-url)
Summary – creating ceria(IV) -protein complex before film deposition

Mefp-1/ceria (IV) film demonstrates a very stable behavior in polarization resistance compared to both control and Mefp-1. At the initial exposure the inhibition efficiency was very low, probably due to the film was very thin and hence presented a lower polarization resistance compared with the air-formed oxide layer of the control sample. However, the air-formed oxide layer was very fragile and could easily be dissolved during initial exposure whereas the Mefp-1/ceria (IV) could cooperate with corrosion products throughout the measuring period. This could explain the increase detected after one day of exposure. The decrease of the corrosion inhibition efficiency after 3 days was induced by the increase of the resistance of the control samples due to the growth of the corrosion products, however, the resistance of the protein film remained stable judged from the EIS results in fig. 26.

The results from EIS measurements are shown in table 5. Compared to Mefp-1/Fe samples, Mefp-1/ceria (IV) does not meet the expectations. This was probably due to the concentration of the Ce⁴⁺ was too high which induced a relatively high degree of complexation of the Mefp-1. Hence, the adsorption of Mefp-1 onto the carbon steel was limited. After 1 day the Mefp-1/ceria (IV) reduced the corrosion rate with 25 %. However, compared to Mefp-1/Fe samples the Ce⁴⁺/protein complexed film did not create as sufficient protection as expected. Unfortunately there is no possibility to determine whether or not the ceria ions created more than one cross-link by the equipment used. However, the results suggested that Mefp-1/Fe samples all provided better protection than Mefp-1/ceria (IV) which was probably due to the high concentration of Ce(NH₄)₂(NO₃)₆ used. Compared to Mefp-1, the Ce⁴⁺/protein
complexed film did provide better protection than the protein alone and results indicated a denser film after one day.

Table 5 Results obtained from data fitting of EIS spectra.

<table>
<thead>
<tr>
<th>Solution</th>
<th>Time</th>
<th>$R_s$ ($\Omega$ cm$^2$)</th>
<th>$Y_0$ ($\Omega$ cm$^2$)</th>
<th>$n$</th>
<th>$R_p$ ($\Omega$ cm$^2$)</th>
<th>$n_z$ (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control NaCl pH=10</td>
<td>30 min</td>
<td>142</td>
<td>4.0 x 10$^{-4}$</td>
<td>0.9</td>
<td>(3.0 ± 0.85) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>157</td>
<td>7.1 x 10$^{-4}$</td>
<td>0.8</td>
<td>(2.3 ± 0.60) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>145</td>
<td>7.4 x 10$^{-4}$</td>
<td>0.8</td>
<td>(2.5 ± 0.30) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>142</td>
<td>7.4 x 10$^{-4}$</td>
<td>0.8</td>
<td>(3.3 ± 0.57) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td>Mefp-1</td>
<td>30 min</td>
<td>129</td>
<td>6.8 x 10$^{-4}$</td>
<td>0.8</td>
<td>(3.8 ± 0.34) x 10$^3$</td>
<td>21</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>129</td>
<td>1.1 x 10$^{-3}$</td>
<td>0.7</td>
<td>(2.6 ± 0.35) x 10$^3$</td>
<td>12</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>130</td>
<td>1.1 x 10$^{-3}$</td>
<td>0.7</td>
<td>(2.8 ± 0.35) x 10$^3$</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>131</td>
<td>1.0 x 10$^{-3}$</td>
<td>0.7</td>
<td>(2.4 ± 0.42) x 10$^3$</td>
<td>-36</td>
</tr>
<tr>
<td>Mefp-1/ceria (IV) 5 μM</td>
<td>30 min</td>
<td>135</td>
<td>7.0 x 10$^{-4}$</td>
<td>0.8</td>
<td>(2.1 ± 0.56) x 10$^3$</td>
<td>-43</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>135</td>
<td>9.5 x 10$^{-4}$</td>
<td>0.7</td>
<td>(3.1 ± 0.27) x 10$^3$</td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>134</td>
<td>1.0 x 10$^{-3}$</td>
<td>0.7</td>
<td>(2.9 ± 0.41) x 10$^3$</td>
<td>12</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>134</td>
<td>1.1 x 10$^{-3}$</td>
<td>0.7</td>
<td>(3.0 ± 0.18) x 10$^3$</td>
<td>-13</td>
</tr>
</tbody>
</table>

The polarization curves retrieved from potentiodynamic measurements are illustrated in fig. 28. The corrosion potential were very similar however, the corrosion currents differed greatly. The protein complex experienced a higher corrosion current and hence increased the corrosion rate with 14%. The results from the potentiodynamic measurements were consistent with the EIS measurements in table 5 which at the third day indicated that the ceria complex had negative effect to the corrosion resistance.

![Figure 28](image-url)

Figure 28 Illustrates the potentiodynamic polarization curves for control and Mefp-1/Ce4+ 5 μM in 0.1 M NaCl at pH 10 after three days of exposure.

Table 6 reveals results obtained from data fitting of potentiodynamic measurements. Mefp-1/ceria (IV) increased the corrosion rate with approximately 14% however, compared to the
protein alone with an increase of nearly 31% the ceria complexed protein film did improve the protection.

Table 6 Results obtained from data fitting of polarization measurements.

<table>
<thead>
<tr>
<th>Solution</th>
<th>$E_{corr}$ vs. SCE (mV)</th>
<th>$b_a$</th>
<th>$b_c$</th>
<th>$SG$</th>
<th>$I_{corr}$ ($\mu$A/cm$^2$)</th>
<th>$\eta_p$ (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control pH 10</td>
<td>-674</td>
<td>75</td>
<td>245</td>
<td>24</td>
<td>2.5 ± 1.055</td>
<td>-</td>
</tr>
<tr>
<td>Mefp-1</td>
<td>-647</td>
<td>71</td>
<td>250</td>
<td>24</td>
<td>3.3 ± 0.719</td>
<td>-31</td>
</tr>
<tr>
<td>Mefp-1/Ce$^{3+}$ 5 μM</td>
<td>-685</td>
<td>73</td>
<td>234</td>
<td>24</td>
<td>2.8 ± 0.340</td>
<td>-14</td>
</tr>
</tbody>
</table>

6.4 Results – using ceria nanoparticles for film deposition.

Description:
For this test Ce(NH$_4$)$_2$(NO$_3$)$_6$ was replaced with ceria nanoparticles with a size of approximately 10 nm. The nanoparticles may fill voids in-between the Mefp-1 film and allow for further adsorption of the protein by changing the surface charge from positive to negative. 40 μL Mefp-1 was aided to 10 ml of 1% citric acid containing 50 mM Nitrilotri where the latter was used in order to create a more hydrophobic protein. The metals were first immersed in the protein solution for 30 minutes and afterwards placed in a solution containing 500 ppm ceria nanoparticles. This was repeated 4 times and as the last step, the samples were immersed into sodiumperiodate with a concentration of 10 mM. Between every step, the metals were rinsed with MiliQ water. The electrolyte for corrosion test was 0.1 M sodium chloride solution with pH 4.6.

Results of EIS measurements

Illustrated in fig. 29 are EIS results of the control samples in 0.1 M NaCl solution at pH 4.6. The Nyquist plot revealed that the polarization resistance increased with time. After one day the results are relatively stable compared to control samples exposed to electrolyte of pH 10. This might be the result of a more corrosive environment causing more iron ions to dissolve from the metal and hence, increased the protection and thus the polarization resistance. However, although it was more stable, the samples did not achieve as high polarization resistance as in a solution of pH 10. The Pourbaix diagram in fig. 4 explained the differences between the solutions. At alkaline pH, the corrosion products formed (Fe$_2$O$_3$/Fe$_3$O$_4$) were oxides with protective properties. Magnetite (Fe$_3$O$_4$) created insoluble surface oxides whereas hematite (Fe$_2$O$_3$) produced a thin film adhering to the surface. However, at pH 4.6 ferrous iron ions were formed instead which do not experience protective properties. (10) Hence the
polarization resistance for control sample at pH 4.6 was not as large as control sample at pH 10.

Figure 29 Illustrates the EIS spectra for carbon steel in 0.1 M NaCl solution at pH 4.6 during one measurement period of 3 days in total.

Results – Mefp-1 ceria nanoparticles
A bulk material can have different properties compared to its nanoparticles why the latter was used in order to see if the film could become denser. The EIS results for Mefp-1 and ceria nanoparticles composite film are shown in fig. 30. Evident in the Nyquist plot is the high polarization resistance after 30 minutes of exposure. After one day’s exposure, the film has been broken down and the polarization resistance was evidently lower. The increase throughout the remaining measurement period was most probably due to the growth of corrosion products.
Figure 30 Illustrates the EIS spectra for carbon steel coated with Mefp-1 ceria nanoparticles in 0.1 M NaCl solution at pH 10 during one measurement period of 3 days in total.

Summary – using ceria nanoparticles for film deposition.

As illustrated above the polarization resistance was very large after 30 minutes of exposure which in turn resulted in good inhibition efficiency shown in fig. 31. When the film was broken down, the inhibition rapidly decreased and after one day of exposure the inhibition efficiency was actually better for the control sample. The increase throughout the remaining measurement period was most probably due to growth of corrosion products.

Figure 31 Illustrates the inhibition efficiency over time for Mefp-1 with ceria nanoparticles composite film.
The results for task 4 can be viewed in table 7. After short exposure times, the film gives a corrosion inhibition efficiency of approximately 33%. However, with the increasing exposure time the film was degraded very fast and after 1 day of exposure, negative corrosion inhibition was observed. Remember that the solution was much more acidic compared to previous samples. Nevertheless, Mefp-1 ceria nanoparticles still reveal very good results. Comparisons with the all the samples in table 9 (in appendix A) reveal that Mefp-1 ceria nanoparticles have lower capacitance, higher n value and polarization resistance. The results suggests a fairly high homogeneity compared to previous samples which might be due to the fact that the film was more compact and the nanoparticles can reach defects on the surface that might be difficult for regular material. And it can be concluded that the composite film could actually be thicker or denser than all of the other samples.

<table>
<thead>
<tr>
<th>Solution</th>
<th>Time</th>
<th>Rs (Ω cm²)</th>
<th>Y₀ (Ω-1cm-2)</th>
<th>n</th>
<th>Rp (Ω cm²)</th>
<th>nz(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Control NaCl pH≈4.6</strong></td>
<td>30 min</td>
<td>125</td>
<td>8.3 x 10⁻⁴</td>
<td>0.8</td>
<td>(2.5 ± 0.42) x 10⁻³</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>128</td>
<td>8.8 x 10⁻⁴</td>
<td>0.8</td>
<td>(3.0 ± 0.55) x 10⁻³</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>104</td>
<td>1.2 x 10⁻³</td>
<td>0.8</td>
<td>(3.3 ± 0.39) x 10⁻³</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>7 d</td>
<td>110</td>
<td>1.1 x 10⁻³</td>
<td>0.8</td>
<td>(3.0 ± 0.32) x 10⁻³</td>
<td>-</td>
</tr>
<tr>
<td><strong>Mefp-1 ceria nanoparticles</strong></td>
<td>30 min</td>
<td>136</td>
<td>4.1 x 10⁻⁴</td>
<td>0.8</td>
<td>(3.7 ± 1.00) x 10⁻³</td>
<td>33</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>134</td>
<td>7.3 x 10⁻⁴</td>
<td>0.8</td>
<td>(2.9 ± 0.16) x 10⁻³</td>
<td>-3</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>131</td>
<td>8.2 x 10⁻⁴</td>
<td>0.8</td>
<td>(3.3 ± 0.28) x 10⁻³</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>7 d</td>
<td>129</td>
<td>8.9 x 10⁻⁴</td>
<td>0.7</td>
<td>(3.4 ± 0.20) x 10⁻³</td>
<td>12</td>
</tr>
</tbody>
</table>

Comparing the surface layer of the control sample with Mefp-1 ceria nanoparticles composite film sample suggest that the latter was denser after 30 minutes which explained the high inhibition efficiency. Equivalent results were noticed after 3 and 7 days of exposure.

In fig. 32 are the polarization curves for both control and Mefp-1 ceria nanoparticles retrieved from potentiodynamic measurements. It is obvious that samples with the composite film had a lower corrosion potential than the control sample however, the corrosion current may not be as easy to determine. A closer look revealed that the cathodic slope was somewhat steeper for the control sample resulting in higher corrosion current. However, the corrosion current for control sample used at pH 10 was higher revealing that there are more corrosion products protecting the control sample at higher pH.
Figure 32 Illustrates the potentiodynamic polarization curves for control and Mefp-1 ceria nanoparticles composite film in 0.1 M NaCl at pH 4.6 after seven days of exposure.

From table 8 below it is apparent that Mefp-1 ceria nanoparticles reduced the corrosion rate with approximately 17% which was consistent with results above. However, there are samples that produced even better results which are summarized in table 10 in appendix A. Nevertheless, the electrolyte used in this test was much more acidic and through comparison, the lowest corrosion potential and corrosion current were evident for Mefp-1 ceria nanoparticles. The standard deviation for the complex was acceptable concluding the reproducibility feasible.

Table 8 Results obtained from data fitting of polarization measurements.

<table>
<thead>
<tr>
<th>Solution</th>
<th>$E_{corr}$ vs. SCE (mV)</th>
<th>$b_a$</th>
<th>$b_c$</th>
<th>SG</th>
<th>$I_{corr}$ ($\mu A/cm^2$)</th>
<th>$\eta_p$ (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control pH=4.6</td>
<td>-680</td>
<td>60</td>
<td>183</td>
<td>20</td>
<td>1.8 ± 0.216</td>
<td>-</td>
</tr>
<tr>
<td>Mefp-1 Ce nanop.</td>
<td>-699</td>
<td>74</td>
<td>154</td>
<td>22</td>
<td>1.5 ± 0.258</td>
<td>17</td>
</tr>
</tbody>
</table>

7. DISCUSSION

The project’s main focus was to determine if metal ions could improve the corrosion protection of the protein Mefp-1 on mild carbon steel. After 30 minutes the Mefp-1 film was broken down and further increase in polarization resistance was probably due to corrosion products and/or that iron ions released from the dissolving metal complexed the protein. Zhang et al showed that the protein Mefp-1 formed aggregates resulting in a non-uniform surface. This could explain the results suggested that Mefp-1 had lowest homogeneity which is in accordance with Zhang’s studies. (30)
It is evident that metal ions aided to the protein do decrease the corrosion process. Compared to Mefp-1, all protein metal complexed samples provided better protection. After 3 hours, EIS results suggested that Mefp-1/Fe 10 μM was denser than the protein film alone providing higher inhibition efficiency. Hence, the iron ions improved the corrosion protection through complexation. The iron concentration was decreased to 1 μM and 0.1 μM respectively in order to investigate the iron ion dependency of the protein. No conclusion about the films density could be drawn from the data fitting results of EIS measurements due to similarities of the samples. This could be due to the amount of Fe$^{3+}$ was too small resulting in an uncompleted complexation of Mefp-1 hence, the film was not dense enough. However, the polarization resistance increased with decreased amount of iron, which was unexpected since the concentration of iron determined the amount of complexed protein. This could be the result of iron ions dissolved from the metal and hence increased the resistance through increased complexation i.e. the protection is improved by corroding sample. According to Zeng et al, to high iron concentration might occupy the binding sites on the protein and as a result there were less complexation than at lower iron concentrations. Zeng et al also proved that crosslinks are reversible for iron ions where the reversibility increases with contact time and iron concentration up to 10 μM. (31) This could explain why 10 μM has lower polarization resistance. During the measurement period the sample releases more iron to the surface which combined with bonding reversibility might result in less complexation. Since the samples show large variation between each other and over time, the reversibility could explain the standard deviation.

Ce$^{4+}$ was used in order to determine if the ceria ions could further complexate the protein film compared to Fe$^{3+}$. After 3 hours the results suggested a denser film than only Mefp-1 protecting the surface and after 1 day Mefp-1/Ce$^{4+}$ were denser than all Mefp-1/Fe samples. This could be due to Mefp-1/Ce$^{4+}$ had the possibility of further complexation with iron ions dissolved from the carbon steel. However, if the degree of complexation is too high it could desorb the protein film which would explain the decrease in inhibition efficiency at longer exposure time.

During the sample preparation of Mefp-1 ceria nanoparticles, the object was to create a four layer film. The results suggest that the film could be thicker or denser than all the other samples. A fairly high homogeneity compared to previous samples suggests that the film was more compact and the nanoparticles could reach defects on the surface that might be difficult
for regular material to reach. Sababi et al showed that nanoparticles produced a denser film compared to only the protein as a coating. (32) The results above suggested similar behavior although surface studies are needed to confirm the results. This might also explain why the $n$-value is high for nanoparticles compared to previous measurements.

Sources of error:
There were some variations between the samples which could be due to several factors. The control samples were not prepared with any solution i.e. the difference must be due to the polishing process. When rough grinding papers were used, damage might occur to the surface not visible to the naked eye. The results could have been scratches unable to remove with smoother grinding paper, caused by inability to reach the dept. Hence, the surface was not uniform and the results differ between the samples.

When the protective film covered the surface, it was dependent of the preparation of the film and the metal surface to which it was supposed to adhere to. When a protein is used, there is always a possibility for aggregation. Preparing one solution and dividing it into three beakers might result in different amount of protein in the beakers. The problem could be minimized by preparing three different solutions, hence containing the same amount of protein. However, the polishing processes do still affect the outcome. If there were scratches on the surface the protein did not provide uniform coverage causing corrosive agents to attack the sample.

The last source of error was the drying process for which the metal was blow-dried with nitrogen. Although the risk was small it is worth mentioning. When blowing nitrogen gas on the samples there was a risk that the film was harmed i.e. the protein chains were moved. This could force the protein closer to the edges of the sample and hence, away from the area later used in the measurements. However this should have been minimized since the pressure was rather low and the distance between the nitrogen “pistol” and the samples were fairly large.
8. CONCLUSIONS

The study focused on corrosion inhibition using film formation of Mefp-1 by itself and in combination with iron or ceria. The conclusions drawn are listed below:

× The protein Mefp-1 do serve as a protection however, the film was broken down early in the measuring period resulting in poor long term protection. The stable increase in polarization resistance was probably due to corrosion products serving as a protection.

× Mefp-1/Fe complex improved the protection of the carbon steel and showed higher stability of the iron complexed film. The negative inhibition efficiency after 30 minutes was probably due to sample preparation not allowing enough time for the protein and metal ions to interact.

× Results received from the EIS do imply that the film was denser for Mefp-1 Fe 10 μM compared to only the protein as protection. This was probably due to complexation between the protein and Fe³⁺.

× The results for Mefp-1/Ce⁴⁺ suggested denser film than Mefp-1 after 3 hours and all Mefp-1/Fe samples after 1 day. However, the high concentration of ceria salt used combined with dissolving iron ions could have desorbed the film hence explaining the decreased inhibition efficiency after longer exposure time.

× The results from the EIS and PD suggest that Mefp-1 ceria nanoparticles had thicker or denser film than the other samples which has been proven elsewhere. Evidently the surface had relatively high homogeneity compared to previous samples suggesting less aggregation and improved coverage of the surface.

9. RECOMMENDATIONS

Even though data retrieved above suggest that a film should be thicker or denser, surface studies have to be preformed to actually confirm the theory as for instance Atomic Force Microscopy (AFM) or Scanning Electron Microscope (SEM). These measurements would also reveal if the protein is aggregated or uniform throughout the surface.

Theoretically one metal ion should be able to complexate to an amount of protein chains although there was no way of telling from the results above. In order to see how many crosslinks there actually were in the sample, IR or Raman should be used. This would also confirm which metal, ceria or iron, had larger affinity of conducting crosslinks.
10. ACKNOWLEDGEMENTS

The diploma work was carried out at the Division of Surface and Corrosion Science, Royal Institute of Technology (KTH), Stockholm, Sweden. The thesis would not have been possible without the guidance and knowledge from several individuals involved.

First I would like to express my deep gratitude to my supervisor Professor Jinshan Pan for trusting me with this project and providing guidance and encouragements.

I would like to express my utmost appreciation to my co-supervisor Fan Zhang who has dedicated a generously amount of time for this project as well as contributing to constructive dialogs and laboratory guidance.

A special thanks to Majid Sababi for taking time sharing his expertise in the instruments used for the study.

I would like to acknowledge the researchers at the department who took their time discussing and casting an eye on the essay.

I would like to thank Biopolymer of Sweden for providing the mussel protein making the project feasible. The protein costs a lot which could have been a large limitation in the several studies made.

Lastly, I would like to thank my family for their support and encouragements.
BIBLIOGRAPHY


APPENDIX A

Table 9 below shows a summary of all the results received after electrochemical impedance spectroscopy.

Table 9 Results obtained from data fitting of EIS spectra.

<table>
<thead>
<tr>
<th>Solution</th>
<th>Time</th>
<th>$R_s$ (Ω cm$^2$)</th>
<th>$Y_0$ (Ω$^{-1}$cm$^2$)</th>
<th>n</th>
<th>$R_p$ (Ω cm$^2$)</th>
<th>$n_z$ (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control NaCl pH=10</td>
<td>30 min</td>
<td>142</td>
<td>4.0 x 10$^{-4}$</td>
<td>0.9</td>
<td>(3.0 ± 0.85) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td>(Task 1)</td>
<td>3 h</td>
<td>143</td>
<td>6.1 x 10$^{-4}$</td>
<td>0.8</td>
<td>(1.9 ± 0.36) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>157</td>
<td>7.1 x 10$^{-4}$</td>
<td>0.8</td>
<td>(2.3 ± 0.60) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>145</td>
<td>7.4 x 10$^{-4}$</td>
<td>0.8</td>
<td>(2.5 ± 0.30) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>142</td>
<td>7.4 x 10$^{-4}$</td>
<td>0.8</td>
<td>(3.3 ± 0.57) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td>Mefp-1</td>
<td>30 min</td>
<td>129</td>
<td>6.8 x 10$^{-3}$</td>
<td>0.8</td>
<td>(3.8 ± 0.34) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>3 h</td>
<td>126</td>
<td>1.0 x 10$^{-3}$</td>
<td>0.8</td>
<td>(2.3 ± 0.10) x 10$^3$</td>
<td>21</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>129</td>
<td>1.1 x 10$^{-3}$</td>
<td>0.7</td>
<td>(2.6 ± 0.35) x 10$^3$</td>
<td>14</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>130</td>
<td>1.1 x 10$^{-3}$</td>
<td>0.7</td>
<td>(2.8 ± 0.35) x 10$^3$</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>131</td>
<td>1.0 x 10$^{-3}$</td>
<td>0.7</td>
<td>(2.4 ± 0.42) x 10$^3$</td>
<td>-36</td>
</tr>
<tr>
<td>Mefp-1 Fe 10 μM</td>
<td>30 min</td>
<td>128</td>
<td>5.5 x 10$^{-4}$</td>
<td>0.8</td>
<td>(2.4 ± 0.19) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>3 h</td>
<td>126</td>
<td>7.5 x 10$^{-4}$</td>
<td>0.8</td>
<td>(2.5 ± 0.23) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>125</td>
<td>8.5 x 10$^{-4}$</td>
<td>0.7</td>
<td>(2.6 ± 0.26) x 10$^3$</td>
<td>24</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>147</td>
<td>1.0 x 10$^{-3}$</td>
<td>0.8</td>
<td>(2.9 ± 0.27) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>129</td>
<td>1.1 x 10$^{-3}$</td>
<td>0.7</td>
<td>(3.3 ± 0.26) x 10$^3$</td>
<td>-2</td>
</tr>
<tr>
<td>Mefp-1 Fe 1 μM</td>
<td>30 min</td>
<td>133</td>
<td>6.7 x 10$^{-4}$</td>
<td>0.8</td>
<td>(3.5 ± 0.41) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td>(Task 2)</td>
<td>1 d</td>
<td>129</td>
<td>1.1 x 10$^{-3}$</td>
<td>0.8</td>
<td>(2.7 ± 0.50) x 10$^3$</td>
<td>13</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>127</td>
<td>1.1 x 10$^{-3}$</td>
<td>0.8</td>
<td>(3.3 ± 0.18) x 10$^3$</td>
<td>14</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>126</td>
<td>1.1 x 10$^{-3}$</td>
<td>0.8</td>
<td>(3.0 ± 0.34) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td>Mefp-1 Fe 0.1 μM</td>
<td>30 min</td>
<td>135</td>
<td>1.0 x 10$^{-3}$</td>
<td>0.8</td>
<td>(2.8 ± 0.56) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>134</td>
<td>1.3 x 10$^{-3}$</td>
<td>0.7</td>
<td>(2.7 ± 0.24) x 10$^3$</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>139</td>
<td>1.2 x 10$^{-3}$</td>
<td>0.7</td>
<td>(3.3 ± 0.32) x 10$^3$</td>
<td>24</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>135</td>
<td>1.2 x 10$^{-3}$</td>
<td>0.7</td>
<td>(3.8 ± 0.39) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td>Mefp-1 ceria salt 5 μM</td>
<td>30 min</td>
<td>135</td>
<td>7.0 x 10$^{-4}$</td>
<td>0.8</td>
<td>(2.1 ± 0.56) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td>(Task 3)</td>
<td>1 d</td>
<td>135</td>
<td>9.5 x 10$^{-4}$</td>
<td>0.7</td>
<td>(3.1 ± 0.27) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>134</td>
<td>1.0 x 10$^{-3}$</td>
<td>0.7</td>
<td>(2.9 ± 0.41) x 10$^3$</td>
<td>13</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>134</td>
<td>1.1 x 10$^{-3}$</td>
<td>0.7</td>
<td>(3.0 ± 0.18) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td>Control NaCl pH=4.6</td>
<td>30 min</td>
<td>125</td>
<td>8.3 x 10$^{-4}$</td>
<td>0.8</td>
<td>(2.5 ± 0.42) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td>(Task 4)</td>
<td>1 d</td>
<td>128</td>
<td>8.8 x 10$^{-4}$</td>
<td>0.8</td>
<td>(3.0 ± 0.55) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>104</td>
<td>1.2 x 10$^{-3}$</td>
<td>0.8</td>
<td>(3.3 ± 0.39) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>7 d</td>
<td>110</td>
<td>1.1 x 10$^{-3}$</td>
<td>0.8</td>
<td>(3.0 ± 0.32) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td>Mefp-1 Ce nanopart.</td>
<td>30 min</td>
<td>136</td>
<td>4.1 x 10$^{-4}$</td>
<td>0.8</td>
<td>(3.7 ± 1.00) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>1 d</td>
<td>134</td>
<td>7.3 x 10$^{-4}$</td>
<td>0.8</td>
<td>(2.9 ± 0.16) x 10$^3$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>3 d</td>
<td>131</td>
<td>8.2 x 10$^{-4}$</td>
<td>0.8</td>
<td>(3.3 ± 0.28) x 10$^3$</td>
<td>33</td>
</tr>
<tr>
<td></td>
<td>7 d</td>
<td>129</td>
<td>8.9 x 10$^{-4}$</td>
<td>0.7</td>
<td>(3.4 ± 0.20) x 10$^3$</td>
<td>-</td>
</tr>
</tbody>
</table>
Table 10 below illustrates the results obtained from polarization measurements.

**Table 10 Results obtained from data fitting of polarization measurements.**

<table>
<thead>
<tr>
<th>Solution</th>
<th>$E_{corr}$ vs. SCE (mV)</th>
<th>$b_a$</th>
<th>$b_c$</th>
<th>SG</th>
<th>$I_{corr}$ ($\mu$A/cm$^2$)</th>
<th>$\eta_p$ (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control pH 10</td>
<td>-674</td>
<td>75</td>
<td>245</td>
<td>24</td>
<td>2.5 $\pm$ 1.055</td>
<td>-</td>
</tr>
<tr>
<td>Mefp-1</td>
<td>-647</td>
<td>71</td>
<td>250</td>
<td>24</td>
<td>3.3 $\pm$ 0.719</td>
<td>-31</td>
</tr>
<tr>
<td>Mefp-1 Fe 10 μM</td>
<td>-699</td>
<td>82</td>
<td>285</td>
<td>26</td>
<td>2.4 $\pm$ 0.986</td>
<td>3</td>
</tr>
<tr>
<td>Mefp-1 Fe 1 μM</td>
<td>-642</td>
<td>76</td>
<td>244</td>
<td>25</td>
<td>2.6 $\pm$ 1.105</td>
<td>-4</td>
</tr>
<tr>
<td>Mefp-1 Fe 0.1 μM</td>
<td>-700</td>
<td>76</td>
<td>190</td>
<td>24</td>
<td>1.8 $\pm$ 0.009</td>
<td>26</td>
</tr>
<tr>
<td>Mefp-1 ceria 5 μM</td>
<td>-687</td>
<td>85</td>
<td>297</td>
<td>28</td>
<td>3.7 $\pm$ 0.869</td>
<td>-12</td>
</tr>
<tr>
<td>Control pH=4.6</td>
<td>-680</td>
<td>60</td>
<td>183</td>
<td>20</td>
<td>1.8 $\pm$ 0.216</td>
<td>-</td>
</tr>
<tr>
<td>Mefp-1 Ce nanop.</td>
<td>-699</td>
<td>74</td>
<td>154</td>
<td>22</td>
<td>1.5 $\pm$ 0.258</td>
<td>17</td>
</tr>
</tbody>
</table>
APPENDIX B

Several studies have been made in the search for natural corrosion inhibitors. It ranges from plants to animals and some studies are showing promising results. Here there will be a short summary of the several research areas today and their result in the hunt for a new “green” corrosion inhibitor.

Black pepper has proved to be useful as corrosion inhibitor. According to Raja et al the efficiency is depending of the concentration i.e. more substrate can adsorb to the surface and thereby shield the metal. The presence of piperine, the chemical that gives black pepper its spiciness, was the reason to its inhibition efficiency. [1]

Another concentration dependent inhibitor is natural honey. El-Etre et al demonstrated that the inhibition efficiency was dependent on iron ions dissolved from the sample i.e. some corrosion products are needed in order to build up an efficient protection. They also found that after a certain exposure time, fungi started to grow and thereby reducing the inhibition efficiency. However, when using high saline water this problem was reduced markedly. [2]

Abdel-Gaber et al found that olive leaf extract could be used as a good corrosion inhibitor and also as an antiscalant i.e. able to interfere with precipitation. As previous mentioned, the inhibition efficiency is dependent of the extract concentration. [3]

Negm et al modified vanillin in order to receive cationic vanillin. The results revealed that cationic vanillin had wanted properties and that it functioned as a mixed inhibitor. They also found that an increase of the alkyl chain attached to the inhibitor resulted in an increase of the inhibition efficiency. [4]

Loto et al used Kola Tree and Tobacco extracts in order to slow down corrosion. They found that the best inhibition was when using kola leaf and kola bark with a inhibition efficiency of 67,2% and 66,67% respectively. Kola nut and kola bark combined barely reached half the inhibition efficiency of the previous mentioned which was also the case for kola leaf and tobacco leaf combined. After 21 days only kola nut and kola bark proved inhibition. [5]

Umoren et al found that Raphia hookeri exudates gum can function as an inhibitior for aluminum. An increase in concentration also increased the inhibition efficiency although an increase in temperature had the opposite effect. [6]
Solid waste (Genus Musa, Genus Saccharum and Citrullus Lanatus) as corrosion inhibitor was investigated by Ismail et al. The results revealed good inhibition ranging from approximately 60 – 70% where Genus Saccharum had the best efficiency and Genus Musa the lowest. [7]

**BIBLIOGRAPHY**


