

The Effect of *Ralstonia pickettii* on Environmental Fatigue Crack Growth of 7xxx Series Aluminum Alloys

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Abstract. As more industries and nations focus on environmental protection, the desire to develop non-toxic, sustainable coatings to protect against corrosion becomes a primary focus. One of the areas on the cutting edge of new coating development is the use of bacteria in corrosion prevention coatings. For many years the focus in the corrosion community was on microbial influenced corrosion with the assumption that all bacteria had negative consequences for corrosion and corrosion fatigue. More recently it has been documented that a variety of bacteria can protect against general surface corrosion. None of the work to date on bacteria preventing general corrosion has shown that the inhibitive effects could also be applied to corrosion fatigue. Researchers at the United States Air Force Academy discovered that a bacteria, *Ralstonia pickettii*, is capable of reducing the fatigue crack growth rate of AA7075-T651 and AA7475-T7351 in 0.06M NaCl to near that of chromate. In cycles to failure testing in 0.06M NaCl the sample life was extended approximately 5 to 6 times depending on the fatigue loading variables compared to samples in 0.06M NaCl without the bacteria. The mechanism behind the corrosion fatigue protection is being investigated in hopes that it could lead to the development of new coatings to reduce corrosion fatigue. The current theories behind how the bacteria slows corrosion fatigue crack propagation are (1) presence of a biofilm, (2) metal sequestration and replating on the crack surface, (3) desalination of the test solution, and (4) oxide layer development and repair.

Introduction

Historically researchers have observed that microbial induced corrosion was detrimental to the metal and accelerated the corrosion process. More recent research has started to look at the ability of microbe to protect aluminum against corrosive effects [1]. Given that bacteria are diverse living organisms with many reactions and processes available depending on the genus and species, it is likely that the reason for corrosion protection or acceleration can be material, bacteria and/or environment dependent.

While there is some research looking at the effect of bacteria on pitting and general corrosion very little work has been completed looking at the effect of bacteria on corrosion fatigue inhibition [1-3]. The Center for Aircraft Structural Life Extension (CAStLE) at the United States Air Force Academy (USAF) has discovered that a bacteria *Ralstonia pickettii* (generally thought to be of limited

occupational concern for otherwise healthy individuals) slows the fatigue crack growth rates in 7xxx series aluminum alloys [4].

Experimental Procedure

In 2011, stress life corrosion fatigue tests were being completed at varying stress levels and stress ratios (R) of either 0.1 or 0.65 to examine the effect of a modern alloy and temper (AA7475-T7351) on the initiation of a crack from corrosion damage (pit) compared to a legacy alloy and temper (AA7075-T651) at a frequency (f) of 1 Hz. Figure 1 shows the sample and test chamber used for this testing. Bacteria grew on some of the samples changing the fatigue life.

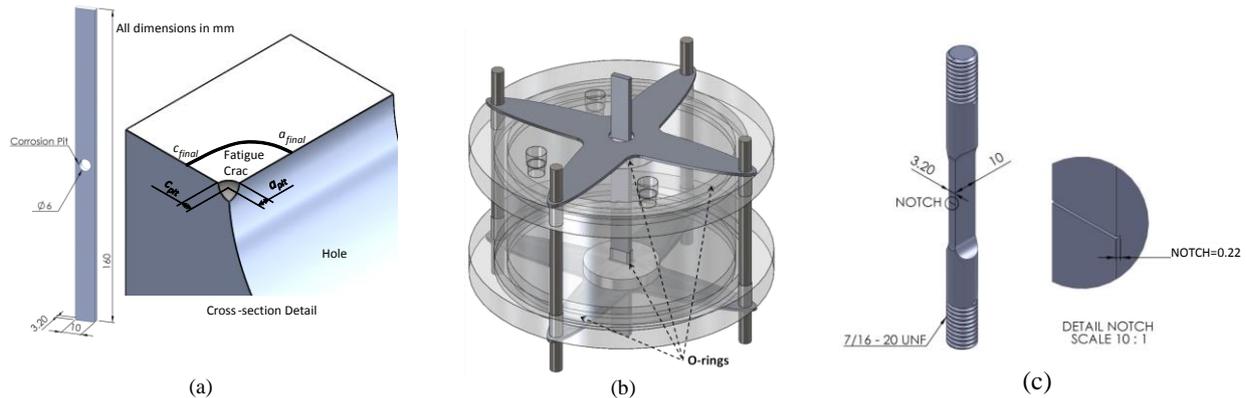


Figure 1: (a) Fatigue sample with a preferential pit for crack initiation used in stress life corrosion fatigue testing. (b) Environmental test chamber used for corrosion fatigue testing. (c) Single edge notch (SEN) specimens were made of 7xxx series aluminum alloy. All dimensions are in millimeters.

After the stress life testing was completed, the bacterial effect was examined using the direct current potential drop (dcPD) method to measure the effect on the crack growth rate. All fatigue crack growth rate testing was completed using a single edge notch specimen, shown in Figure 1. The fatigue test was completed at a constant ΔK ($6 \text{ MPa}\sqrt{\text{m}}$) over a range of frequencies (0.02-20 Hz) and at a stress ratio (R) of either 0.10 or 0.65. The test parameters were selected to compare the crack growth rates to published inhibited corrosion fatigue crack growth rates using chromate and molybdate [5-7]. The corrosive environment was 0.06 M NaCl. The bacteria were allowed to grow naturally or intentionally added to solution to quantify the effect. To add bacteria to solution, colonies of *R. pickettii* were grown on R2A agar and single colonies were added to the 0.06 M NaCl solution. Figure 2 shows the growth of the bacteria on the aluminum sample. *R. pickettii* is known to build biofilms and the growth is thought to be a biofilm [4,8-10]. Figure 2b gives a schematic of biofilm development [8].

Results

The results from the stress life fatigue testing showed that the presence of the bacteria during the tests completed with an $R=0.1$ saw approximately 5-fold increase in fatigue life. The samples tested with an $R=0.65$ saw a 6-fold increase in fatigue life. Figure 3 shows the comparison of high and low amounts of *R. pickettii* when added to a 0.6 M NaCl solution to a known corrosion fatigue inhibitor chromate (Na_2CrO_4) at $f = 1 \text{ Hz}$ [5]. The addition of 0.3 M of chromate lowers the fatigue crack growth rate for frequencies just below 1 Hz; when 0.5 M chromate is added, the inhibition occurs over the entire frequency range (0.1-70 Hz) [5]. The amount of bacteria was quantified visually, based on the amount of “fuzz” seen. For crack lengths of 2-4 mm both the high and low bacteria

concentrations slow the crack growth rates to that of high amounts of inhibiting pigments [5]. For the 6-6.5 mm data the inhibition appears to be less effective particularly for the low bacteria case. However it should be noted that the presence of the bacteria can cause the dcPD system to under-measure the crack length (increases conductivity of the sample) which means for a constant ΔK test, where the load is being constantly adjusted, an overload is applied. Based on post-test analysis of the crack length, the 6-6.5 mm crack length data is likely between $\Delta K=10-14 \text{ MPa}\sqrt{\text{m}}$, meaning that inhibition is still occurring even though the figure suggests it is not.

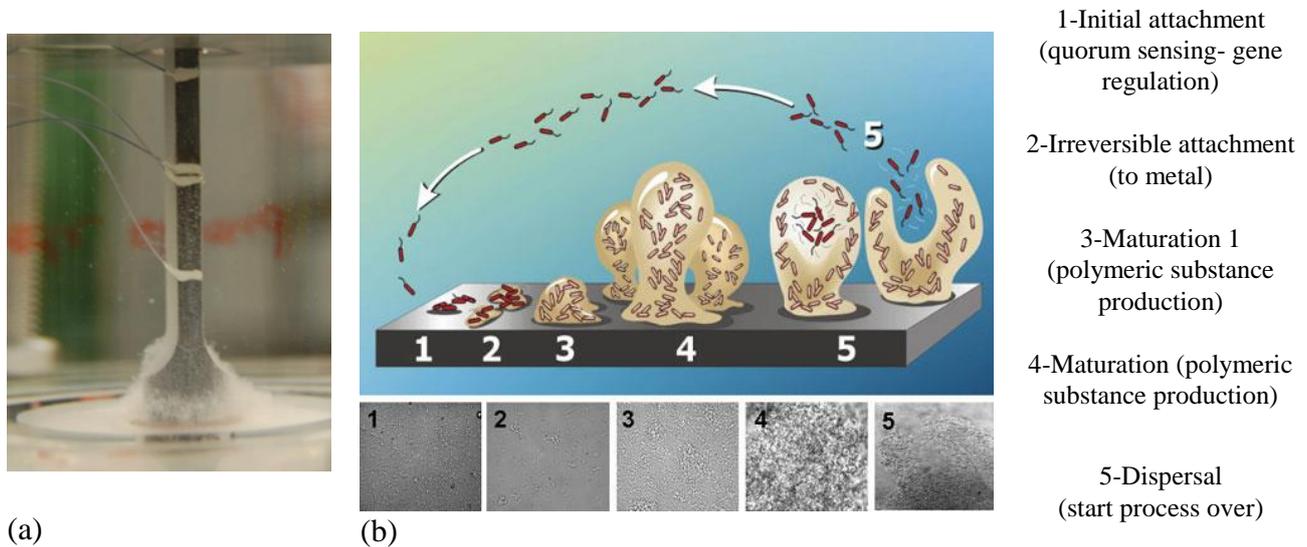


Figure 2: (a) Bacterial growth ("fuzz" at bottom of samples) on the 7xxx series aluminum alloy in 0.06M NaCl solution. The white on the dcPD wires is a protective coating. The fuzz continues up the entire length of the sample. (b) Biofilm development [8].

A large database of inhibition of corrosion fatigue crack growth by a corrosion replacement inhibitor, molybdate, has been published [6,7]. That work found that with inhibitive salts thought to form a passive film at the crack tip testing at an $R=0.65$ produced better inhibition so a comparison with *R. pickettii* was completed over a range of frequencies. Figure 4 shows the comparison between *R. pickettii* and molybdate [6,7]. Again in this figure the progression of inhibition by increasing (0.1 mM to 0.6 M) levels of molybdate can be noted, with the 0.6 M molybdate slowing the crack growth rate to that of vacuum at 0.02 Hz. The tests were run as frequency scans, starting at either the high (20 Hz) or low (0.02 Hz) frequency and then progressing in the order of the arrows. Each frequency scan would take on the order of two weeks to complete. The results showed that if a test is started before the bacteria has had time to become visible ("fuzzy") from Figure 2 then inhibition of the crack growth rate does not occur, however once the fuzz is visible then inhibition can occur even up to frequencies of 20 Hz. It should also be noted that the reduced flushing of the crack tip caused by the increased stress ratio does not increase the ability of the bacteria to inhibit corrosion fatigue when compared to the testing completed in comparison with chromate shown in Figure 3 [11].

Discussion and Conclusions

R. pickettii consistently shows inhibition on the order of known corrosion inhibitors as shown in Figures 3 and 4 [5-7]. The reason for this inhibition is currently unknown, however there are three leading theories being investigated: (1) *R. pickettii* is sequestering metal (copper) into its cell wall which is stabilizing the passive film at the crack tip [3,10,11], (2) the bacteria is producing oxygen which is causing an oxide layer to develop and protect the crack tip, (3) the polymeric biofilm the bacteria produces allows for protection of the crack through an inhibitive species or coating [14]. *R.*

pickettii has been shown to uptake certain metals into its cell wall to protect itself from hazardous environments [12,13]. Based on aluminum corrosion fatigue inhibition research and *R. pickettii* metal uptake literature, copper and silicon were loaded into R2A agar and *R. pickettii* colonies grown on the agar were analyzed by scanning electron microscopy and electron dispersive spectroscopy (EDS) [6, 12, 13]. The results did not show evidence of metal uptake. The ability of bacteria to help repair the oxide layer is supported by the literature and could possibly be occurring in this case [1]. When the aluminum samples with bacterial growth were analyzed using EDS, the areas with biofilm have large amounts of oxygen present, suggesting a heavy oxide layer.

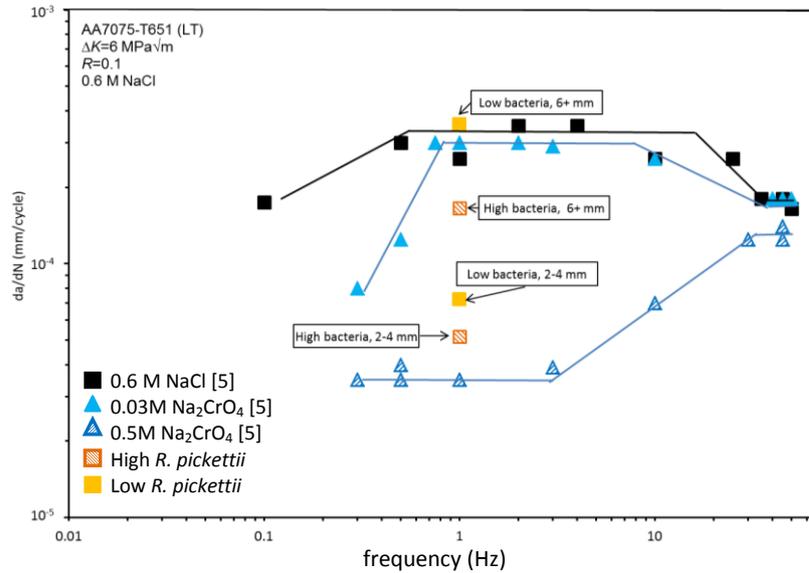


Figure 3: Comparison of the effect of *R. pickettii* on fatigue crack growth rates on AA7075-T651 to a known corrosion fatigue inhibitor chromate at $\Delta K=6 \text{ MPa}\sqrt{\text{m}}$; $R=0.1$, $f=1 \text{ Hz}$ in 0.6 M NaCl [5].

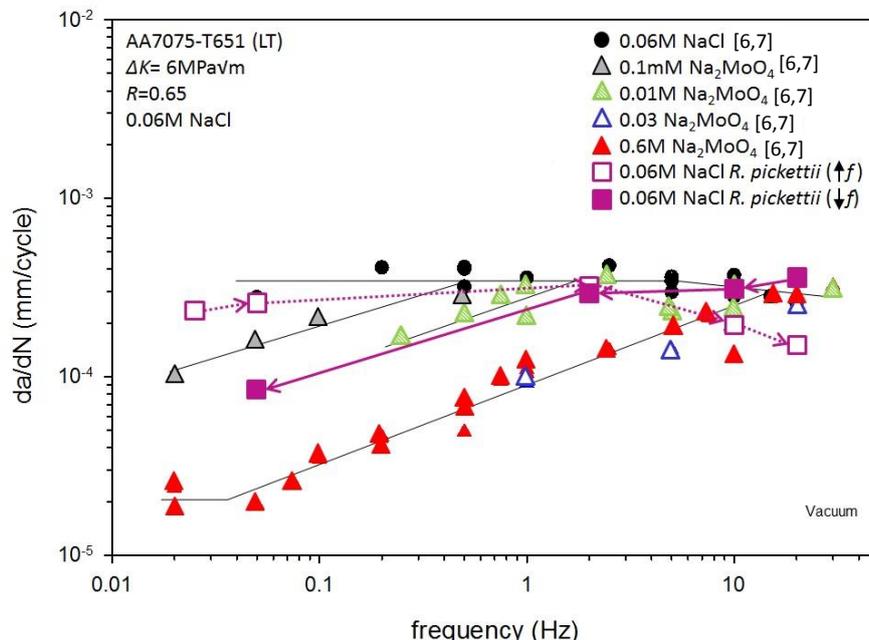
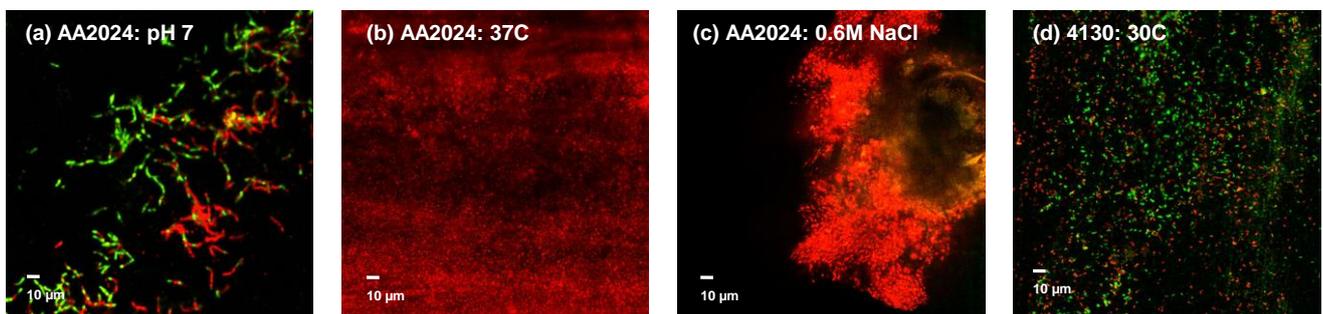


Figure 4: Comparison of *R. pickettii* inhibition to a known corrosion fatigue inhibitor, molybdate, at a constant ΔK of $6 \text{ MPa}\sqrt{\text{m}}$, R of 0.65 over frequencies from 0.02 to 20 Hz [6,7]. The arrows denote the order of testing.

During the corrosion fatigue testing it has been shown that *R. pickettii* can desalinate the sodium chloride test solution. The salinity is expected to drop by approximately half in 48 hours. For aluminum alloys the presence of chloride can be detrimental to corrosion fatigue but is not a primary driving force as the presence of water vapor is sufficient to raise crack growth rates over

that of dry air or inert environments [15]. This suggests that the desalination of the test solution is not the primary cause of the corrosion fatigue inhibition.

Work is underway to analyze the structure of the biofilm. The early research has looked at how the biofilm develops under varying conditions in an effort to understand why the biofilm sometimes appears to be produced faster and thicker than at other times [16]. Figure 5 shows the development of the *R. pickettii* biofilm using a live/dead stain with confocal microscopy under varying temperatures, pH, and salinities and with various alloys. The amount of growth and living bacteria is rated from no growth/living (-) to heavy biofilm/all living (++++). It appears that a neutral to slightly basic pH (7-8) allows for string-like bacteria formations (5a). Higher temperatures (30 and 37 °C) (5b) and low salt content (0.06 to 0.6 M NaCl) (5c), or neutral to slightly basic pH (7-8) cause thick biofilm formation. Formation on 4130 steel is scattered but not stringy (5d). Interestingly these are conditions very close to what exist in the corrosion fatigue test cell.



24 Hr Biofilm Development

Alloy	Exposure Parameters	Growth Amount	Amount Living	Morphology
AA2024	10C	+	++	Scattered
AA2024	30C	+++	-	Fuzzy
AA2024	37C	++	+	Fuzzy
AA2024	Broth	++++	+	Fuzzy
AA2024	0.03 M NaCl	++++	+	Fuzzy
AA2024	0.06M NaCl	++++	+	Fuzzy
AA2024	0.6M NaCl	++++	+	Fuzzy
AA2024	pH 6	+	++	Scattered
AA2024	pH 7	++	+++	Stringy
AA2024	pH 8	++++	++++	Fuzzy
4130	30C	+++	++	Scattered
4130	37C	++	+	Scattered
AA7075	37C	+++	+	Scattered
AA2024	Mn	+++	-	Stringy
AA2024	Fe	+++	-	Fuzzy
AA2024	Cu	++	++	Scattered
AA2024	Ni	+	-	Fuzzy

48 Hr Biofilm Development

Alloy	Exposure Parameters	Growth Amount	Amount Living	Morphology
AA2024*	10C	-	-	N/A
AA2024	30C	+++	-	Fuzzy
AA2024	37C	++++	-	Fuzzy
AA2024	Broth	+++	+	Fuzzy
AA2024	0.03 M NaCl	+++	+	Fuzzy
AA2024*	0.06M NaCl	-	-	N/A
AA2024	0.6M NaCl	++++	-	Fuzzy
AA2024	pH 6	+	+	Scattered
AA2024	pH 7	+++	++	Stringy
AA2024	pH 8	++++	++	Fuzzy
AA2024	30C	+	+	Scattered
AA7075	30C	+	+	Scattered
AA2024	Ni	-	-	N/A
AA2024	Mn	++	-	Stringy
AA2024	Fe	+++	-	Fuzzy
AA2024	Cu	++++	-	Fuzzy

Figure 5: Development of a *R. pickettii* biofilm under varying conditions; pictures and charts [16].

Over the course of this project, research has also been conducted into how difficult *R. pickettii* is to kill. This is particularly important because if bacteria were to be developed into a natural protective coating for any sort of military or commercial application it needs to be able to withstand a variety of environmental conditions. To date the bacteria has proven to be extremely resistant to most forms of disinfection including, heat, freezing, bleach solutions, heavy metal exposure, hydrogen peroxide and UV light exposure. The ability of these bacteria to resist disinfection suggests that the processes of living bacteria could be placed into a coating to make a non-toxic, environmentally friendly coating with corrosion fatigue protection on the order of chromate.

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