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Use of hydrogen peroxide as an oxygen source for *in situ* biodegradation
Part II. Laboratory studies

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Abstract

A review of the literature on hydrogen peroxide decomposition and stabilization identified several inorganic and organic compounds that are known to decrease the rate of decomposition of peroxide in simple systems. Phosphate, a commonly used stabilizer in groundwater applications, does not stabilize peroxide in the presence of enzymatic catalysts, primarily catalase, which are the most important catalysts of peroxide decomposition. In laboratory experiments, a variety of other peroxide stabilizers identified in the literature review also did not sufficiently stabilize peroxide. It is concluded that hydrogen peroxide may not be an efficient source of oxygen for *in situ* bioreclamation processes, at the Eglin Air Force Base, Florida site.

Introduction

In situ biodegradation of fuel hydrocarbons by indigenous microbial populations is a promising technique for the remediation of contaminated soils. In field applications of enhanced *in situ* bioreclamation, oxygen supply is the limiting factor. Because of the limited solubility of oxygen in water, hydrogen peroxide is commonly used as an oxygen source. The rate of peroxide decomposition, however, is generally greater than the rate of oxygen utilization by microbial growth [1]. Although the addition of phosphate is known to decrease the rate of peroxide decomposition in aqueous solutions [2], results of a field demonstration of *in situ* biodegradation conducted at the Eglin Air Force Base in northwest Florida [1] demonstrate that phosphate may not sufficiently stabilize hydrogen peroxide in the aquifer, resulting in an inefficient delivery of oxygen.

Because *in situ* biodegradation is potentially an effective technology for re-

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mediating many hydrocarbon-contaminated sites, finding an appropriate additive that will decrease and control the rate of peroxide decomposition would be helpful. Accordingly, we reviewed the literature to understand the mechanism of peroxide decomposition and to identify additives for stabilization. The effectiveness of selected additives for stabilizing hydrogen peroxide in groundwater was then evaluated with laboratory experiments. Results of this investigation are presented below.

Peroxide decomposition

Hydrogen peroxide decomposes to release oxygen and water:

$$H_2O_2 \rightarrow H_2O + \tfrac{1}{2}O_2 \quad (1)$$

Many substances commonly present in groundwater and soils act as catalysts for Reaction (1). Important among these are aqueous species of iron and copper and the enzyme catalase [3]. Several mechanisms for the action of catalysts on peroxide decomposition have been described. The proposed mechanism for the reaction catalyzed by ferric and ferrous iron is as follows [4–6]:

$$Fe^{2+} + H_2O_2 \rightleftharpoons Fe^{3+} + HO^{\cdot} + HO^- \quad (2)$$

$$Fe^{3+} + H_2O_2 \rightleftharpoons Fe^{2+} + HO_2^{\cdot} + H^+ \quad (3)$$

$$HO^{\cdot} + H_2O_2 \rightleftharpoons H_2O + HO_2^{\cdot} \quad (4)$$

$$Fe^{3+} + HO_2^{\cdot} \rightleftharpoons Fe^{2+} + H^+ + O_2^{\cdot} \quad (5)$$

$$Fe^{2+} + HO_2^{\cdot} \rightleftharpoons Fe^{3+} + HO_2^- \quad (6)$$

$$Fe^{2+} + HO^{\cdot} \rightleftharpoons Fe^{3+} + HO^- \quad (7)$$

where Fe^{2+} or Fe^{3+} may be the single ions or complex ionic species of iron. Rate constant data indicate that Reaction (2) controls the overall reaction rate. The mechanism of catalytic action of copper (Cu) is similar to that of iron.

Although inorganic catalysts contribute to the decomposition of peroxide *in situ*, enzymatic catalysts, peroxidase, and catalase may be dominant in soil–groundwater systems. Peroxidases are a group of catalysts that oxidize a substrate in the presence of hydrogen peroxide. Peroxidase activities are greatest in soils containing carbonate and may be a function of the number of soil microorganisms [7]. Peroxidase activity changes with the type of vegetation and with the seasons.

Catalases are enzymes that degrade hydrogen peroxide to produce water and molecular oxygen. Catalase activity in soils is associated with high organic matter content. The highest catalase activity is found in litter-accumulating surface layers and in humus-accumulating horizons of the soil column. Catalase activity is also stronger in alkaline and calcareous soils than in acidic soils

TABLE 1

Catalytic activity of several ferric-centered catalysts in the decomposition of hydrogen peroxide (from Reference [8])

Catalyst	Activity (turnover number)[a]
Catalase	9×10^4 (pH 7, 20°C, 0.01 mol peroxide)
Peroxidase	4.0 (pH 7, 20°C, 0.01 mol peroxide)
Fe(III)-TETA	22.0 (pH 7, 25°C, 0.15 mol peroxide)
Fe(II) or Fe(III) ion	1.0 (pH 5, 0°C)

[a]Turnover number is defined as the number of peroxide molecules decomposed per second by each mole of the catalyst.

[7]. The catalytic action of catalase in decomposing peroxide is by far the strongest of all the inorganic and enzymatic catalysts (Table 1).

Catalases are high-spin ferric complexes containing hemin as the prosthetic group; each molecule usually contains four hemin groups. Experimental studies of hydrogen peroxide decomposition by catalase indicate that there is an initial period of rapid decomposition, followed by a slower, steady rate of decomposition. The kinetics of the reaction during the steady rate of decomposition are first order [4,5]. Both the initial and steady rates of decomposition are proportional to catalase activity; variation with peroxide concentration is complex, rising to a maximum at 14,000 mg/L and 2,400 mg/L H_2O_2, respectively, and then decreasing rapidly [4]. This self-inhibitory effect of hydrogen peroxide is not caused by the destruction of catalase; rather, it results from reversible formation of intermediate, enzyme–substrate compounds [4,8,5]. In other words, the loss of catalase activity at high peroxide concentrations will be recovered with a decrease in peroxide concentration. Brown et al. [5] suggested the following scheme for the catalase–peroxide reaction:

$$E + P \rightleftharpoons EP \tag{8}$$

$$EP \rightarrow EP' \tag{9}$$

$$EP' + P \rightarrow E + \text{Products} \tag{10}$$

$$EP' + P \rightarrow \text{Catalase compound II} \tag{11}$$

where E is the enzyme, P is peroxide, and EP and EP′ are intermediate compounds. Catalase compound II has been identified as an inactive species formed in the reaction and may reversibly react with peroxide to form another inactive species, compound III. This reversible formation of compounds II and III accounts for self-inhibition at high peroxide concentrations [5].

Peroxide stabilization

The stability of hydrogen peroxide can be increased by decreasing the avail-

ability of, or by deactivating, the catalysts. Stannate, phosphate, and fluoride are known to deactivate the inorganic catalysts, whereas borate, sulfide, hypophosphite, fluoride, and several organic compounds reduce the catalytic action of catalase.

The rate of peroxide decomposition catalyzed by Fe may be decreased by adding stannate or phosphate to hydrogen peroxide solutions [3]. Adding sodium stannate forms colloidal hydrous stannic oxide, which adsorbs catalytic ions such as Fe^{3+} and improves the stability of hydrogen peroxide.

The role of the phosphate ion is to scavenge the hydroxyl radical HO• produced in Reaction (2), and, thus, stop the chain decomposition reactions. At environmental pH ranges, the species $H_2PO_4^-$ and HPO_4^{2-} predominate. Both phosphate species can act as HO• scavengers. The rate constant of HO• production is 76.5 $M^{-1}\,s^{-1}$ [6] and for HO• scavenging is $5\times10^6\,M^{-1}$ [9], indicating that Reaction (2) controls the overall reaction rate.

Phosphate also impedes peroxide decomposition by lowering the dissolved Fe(II) and Fe(III) concentrations by either precipitating Fe ions as phosphate salts or by complexation as Fe-PO_4 ions. Both orthophosphate and the various polyphosphates act as peroxide stabilizers. Moderate amounts of fluoride ions also inhibit the Fe-catalyzed decomposition of hydrogen peroxide. However, not all ligands that form aqueous complexes with Fe are effective peroxide stabilizers. For example, while phosphate and fluoride complexes of Fe are catalytically inactive, Fe-EDTA or Fe-TETA complexes are stronger catalysts than Fe^{3+} (Table 1).

Stabilization of hydrogen peroxide in the presence of enzymatic catalysts, primarily catalase, is much more complex. Experimental studies of the hydrogen peroxide–catalase system have identified several inorganic and organic species that have an inhibitory effect on catalase activity. The inhibitory effect of these species arises from a reversible or irreversible formation of complexes between the inhibitors and the free enzyme or enzyme–peroxide compounds.

Inorganic inhibitors of catalase activity include fluoride, borate, sulfide, and hypophosphite (H_3PO_2) [10,11]. Organic inhibitors include acetate, citrate, pyrogallol, hydroquinone, catechol, and several other compounds [3,12]. Of these, only citrate is known to have been used previously in nutrient formulations for *in situ* bioreclamation. The stated purpose of using citrate is to reduce the iron precipitation potential, although it may also have some impact on hydrogen peroxide stability.

Ascorbate also inhibits catalase activity [13]. Ascorbate inhibition of catalase is reversible, but may be enhanced and made irreversible in the presence of either oxygen or organic complexes of copper. However, the presence of both oxygen and copper apparently decreases the irreversible inhibition of catalase by ascorbate [13].

The inhibitory effect of borate on catalase activity has not been studied directly. Jones and Suggett [14] observed that the activity of catalase decreased

in aqueous solutions containing the borax–sodium carbonate buffer. In a more recent study, Kelly and King [15] also suggest inhibitory action of borate on catalase based on their experiments in a system containing iron(II)-porphyrin complexes and hydrogen peroxide.

An additional mechanism in the stabilizing action of borate may involve the formation of aqueous perborate (H_4BO_5) complexes. Thermodynamic studies of the boric acid–hydrogen peroxide system [16] indicate that perborate species are dominant in the pH range of 7.5 to 13. Boric acid is dominant at pH <7.5 and the borate ion at pH >13. Thus, perborates may provide an alternative source of oxygen in the pH range of their stability if direct catalase–perborate interactions are not significant.

For a stabilizer to be effective in an *in situ* bioreclamation process, it must provide sufficient stabilization to allow significant movement of hydrogen peroxide into the aquifer. That is, the stabilized hydrogen peroxide must have a half-life that is a substantial fraction of the travel time from groundwater injection to withdrawal. In a typical field application, this would require a half-life of several hours to several days. The stabilizer itself must not create toxicity problems or be a problematic contaminant. For example, using borate and fluoride as stabilizers would probably result in concentrations in groundwater that exceed U.S. drinking water standards. Organic stabilizers would only be effective if non-toxic and if they do not add significant oxygen demand to the injection water.

A successful stabilizer must also be sufficiently soluble and not be removed from injected groundwater by precipitation, ion exchange, or adsorption. For example, the soluble phosphate ion which is known to stabilize hydrogen peroxide is relatively insoluble in the presence of calcium, common in many groundwaters. The result is that phosphate added to groundwater to stabilize hydrogen peroxide may be precipitated on contact with soils and may reduce aquifer permeability [17].

Experimental design

Experiments to study hydrogen peroxide stabilization were conducted using a combination of batch and flow-through experiments. Batch experiments were conducted in a column packed with 15 to 25 grams of soil. The soils were pretreated for 30 to 60 minutes with a solution containing inorganic nutrients (phosphate and nitrogen) and a hydrogen peroxide-stabilizing additive. The hydrogen peroxide-spiked nutrient solution was then introduced into the bottom of the columns in an upflow configuration for 60 to 80 min (>50 pore volumes). Flow was then stopped and the column sealed to allow batch reaction for 60 to 90 min. Pump tubings were flushed and a sample was taken from the bottom end of the column. Dissolved hydrogen peroxide concentrations were measured by a ceric acid titration procedure using a HACH® kit.

Flow-through experiments were conducted to study on a larger scale the expected performance of hydrogen peroxide stabilizers in the field. A flow-

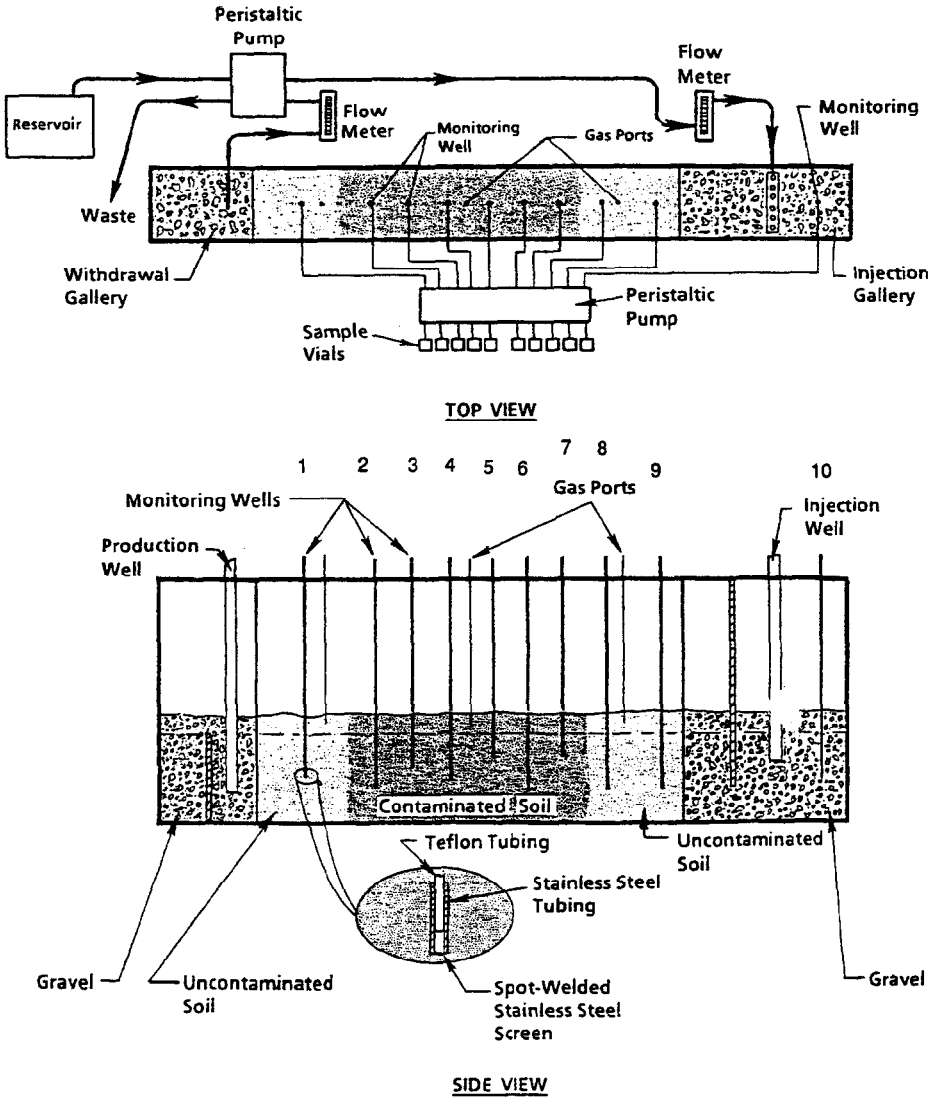

Fig. 1. Two views of the aquifer simulator. Monitoring wells are numbered consecutively with Well 10 near the injection well and Well 1 near the production well. The distances between the injection gallery and the wells are as follows: 9 = 5 cm; 8 = 20 cm,, 7 = 30 cm, 6 = 40 cm, 5 = 52 cm, 4 = 60 cm, 3 = 72 cm, 2 = 82 cm, and 1 = 102 cm.

through aquifer simulator was constructed using a glass tank measuring 183 cm × 76 cm × 18 cm (Fig. 1). Infiltration and extraction galleries (~30 cm each) were created by placing gravel to a height of 25 cm on the two ends of the tank. The ~122-cm space between the gravel zones was filled with zones of uncontaminated soil sandwiching a zone of contaminated soil. The soil was

saturated to a height of ~20 cm and the tank was covered with a Plexiglass® lid. Nutrient solutions were injected into the injection gallery using of a peristaltic pump and the injection rates were monitored with a flow meter. A similar setup was used for pumping solutions from the extraction gallery. Baffles were placed in both the injection and extraction galleries to avoid channeling. The seepage velocity in the soil zone was ~1.25 m/day and the dispersion coefficient was ~0.08 m^2/day [18]. These values are similar to those observed for unconsolidated sandy aquifers [19].

Nine "monitoring wells" were placed in the soil zone and one in the infiltration gallery. The wells were constructed by inserting a 7-cm (internal diameter) stainless steel tubing into the soil or gravel. A stainless steel screen was spot-welded on the lower end of the tube to prevent the flow of wet soil into the well. Teflon® tubing was placed into the well and connected to a peristaltic pump for drawing samples.

Soil samples used for peroxide experiments were collected at Eglin Air Force Base. The contaminated soil was a composite of soils from a depth of 0.3 to 1.2 m collected from the vicinity of the *in situ* hydrogen peroxide studies described by Hinchee et al. [1,20]. Similar soils were collected from a nearby uncontaminated location. The soils consisted primarily of fine- to medium-grained quartz sand.

Results and discussion

Batch studies

Results of batch peroxide stability experiments with uncontaminated soil (Table 2) indicate that the natural peroxide-decomposing capacity of the soil is significantly high. When peroxide solution reacted with soil without any additive (Runs 89-1 and 91-2), up to 85% of the peroxide decomposed in 90 min. Treatment with trimetaphosphate (TMP) only marginally increased the stability (75% decomposition in 90 min). This slight increase in stability probably was because of the effect of TMP on inorganic catalysts, as discussed later in this section.

Adding fluoride, boric acid, or hypophosphite did not increase the stability of peroxide. As noted earlier, perborate may be an alternative source of peroxide. Perborate salts dissolve in aqueous solutions to produce hydrogen peroxide. The aqueous state of this peroxide may be as free-H_2O_2 or a complex perborate ion, depending on pH. Several batch experiments were conducted in which the nutrient solutions were spiked with sodium perborate instead of hydrogen peroxide. The stability of peroxide in these experiments did not improve significantly, with an average decomposition of 56% in 90 min and a range of 44 to 72%.

Finally, three organic additives designed to enhance peroxide stabilization were evaluated. Ascorbic acid (176 and 352 mg/L) or catechol (11 mg/L) did not significantly enhance the stability of peroxide. Using different concentra-

TABLE 2

Decomposition of hydrogen peroxide in uncontaminated Eglin soil

Run #	Additive concentration (mg/L)						Peroxide decomposed after 90 min (%)	Half-life[a] (min)
	TMP	Citric acid	Perborate	Boric acid	Catechol	Fluoride		
89-1							65	22
91-2							85	10
91-3	306						70	19
92-1	306			62			75	16
94-1	306	206					35	41
98-1	306	100		31			75	21
98-2	306					10	80	13
100-1	306		1,170				44	35
100-2	31		1,170				72	17
120-1	306	206					82	11
125-1	306	21					73	17
125-2	31	21					80	13
127-1	306				11		79	13
127-2	31				11		93	5.2

Run	Additive concentration (mg/L)				Peroxide decomposed after 90 min (%)	Half-life[a] (min)
	TMP	Ascorbic acid	Hypophosphite	Restore® 375		
130-1	31		66		99	0.6
130-2	306		66		81	12
131-1	31		6.6		85	9.6
131-2	306		6.6		85	9.6
135-1	306	176				
135-2	306	352			83	11
128-1				800	86	9.0
128-2				800	86	9.0

[a]Calculated with the assumption that reaction rates are first order.

tions of TMP with the organic additives had no effect on peroxide stability. Citric acid at a concentration of 206 mg/L or higher decreased the rate of peroxide decomposition to ~40% in 90 min. At a lower concentration (20 to 100 mg/L), citric acid did not stabilize the peroxide.

Soil samples were taken from the citric acid experiment and agar plated to determine the total microbial population. The microbial populations in three samples from the citric acid experiment (3.9 to 9.5×10^6 colony-forming units [CFU] per gram) were greater than those in the soil before treatment (2×10^4

CFU/g). This indicates that the increase in peroxide stability in the citric acid experiments was not detrimental to microbial growth.

The rate of hydrogen peroxide decomposition was lower in the contaminated soil (Table 3) than in the uncontaminated soil. Peroxide in untreated, contaminated soil without any additives showed a 45% decomposition in 90 min. When perborate was used, 60% of the peroxide decomposed in 60 min. Note that the pretreatment microbial population in the contaminated soil (2×10^4 CFU/g) was two orders of magnitude lower than in the uncontaminated soil (3×10^6 CFU/g).

Adding TMP to boric acid–peroxide solutions or perborate solutions did not significantly decrease the rate of peroxide decomposition in contaminated soil. Citric acid, at a concentration of 100 mg/L or higher, seems to have stabilized peroxide. This concentration is slightly lower than that observed for uncontaminated soil, perhaps because of the lower microbial activity in contaminated soil.

Effect of additives on inorganic and enzymatic catalysts

Decomposition of hydrogen peroxide was examined in sterilized, uncontaminated Eglin soil to estimate the relative activities and inhibition of enzymatic and inorganic catalysts. Uncontaminated soil was sterilized by treatment with a 1 mg/L $HgCl_2$ solution for 16 hours. This treatment decreased the rate of

TABLE 3

Decomposition of hydrogen peroxide in contaminated Eglin soil

Run #	Additive concentration (mg/L)				Peroxide decomposed (%)		Half-life[a] (min)
	TMP	Citric	Perborate	Boric	60 min	90 min	
79-2	306			620		38	38
80-1			78		60		17
82-2	306		78		33		28
83-1	306		6,200		1		41
83-2	306		620		20		33
85-1	306		62		31		29
85-2	2,060				11		37
86-1	306	206			15		35
88-1	306					15	53
94-2						45	34
95-1	306					45	34
95-2	306		62			45	34
96-1	306	206				17	52
96-2	306	100	31			8	58

[a]Calculated with the assumption that reaction rates are first order.

peroxide decomposition in uncontaminated soil to ~60% in 60 min, compared with 85% in 90 min in untreated soil (Table 4). Trimetaphosphate did not increase the stability of peroxide in sterilized soil while citric acid significantly increased the stability of peroxide (28% decomposition in 90 min).

Treatment with $HgCl_2$ inhibits microbial growth, but very likely does not destroy extracellular enzymes. In fact, the treatment may increase the enzymatic activity in the soil by exposing intercellular enzymes to microbial destruction. Therefore, sterilization was achieved also by autoclaving a sample of soil at 120°C for 30 min. The destruction of enzymatic activity by autoclaving was confirmed by using pure catalase as a control.

The rate of peroxide decomposition in autoclaved soil (Table 4) is ~10% in 90 min, with or without additives. This rate of decomposition is significantly lower than that observed for soil treated with mercuric chloride, probably because of the destruction of microbial and enzymatic activities upon autoclaving. The lower rate of peroxide decomposition in autoclaved soils suggests that enzymatic catalysts are dominant in the Eglin soil.

However, it is possible that the activity of the inorganic catalysts was reduced as a result of autoclaving. To clarify the effect of autoclaving on inorganic catalysts of peroxide decomposition in the soil, several experiments were conducted with Fe-enriched solutions. In these experiments, peroxide was added to 100 mL of $FeSO_4$ or $FeCl_3$ solution spiked with other additives, and the pH of the solution was adjusted to 7. The aqueous concentration of hydrogen peroxide was monitored for 24 h. The effect of heating during sterilization on peroxide decomposition in the soil was evaluated by autoclaving the solutions before spiking with peroxide and other additives.

Results of the aqueous batch experiments are given in Table 5. Nearly complete decomposition of peroxide occurred within 12 h in solutions with no additives. Sodium phosphate, TMP, and citric acid inhibited peroxide decom-

TABLE 4

Hydrogen peroxide decomposition in abiotic Eglin soil

Method of sterilization	Active (concentration, mg/L)	Peroxide decomposed after 90 min (%)	Half-life[a] (min)
$HgCl_2$	TMP (306)	60	25
$HgCl_2$	TMP (306)	59	26
$HgCl_2$	TMP (306)+Citric (206)	28	45
Autoclave		15	53
Autoclave	Citric (206)+TMP (306)	9	57

[a]Calculated with the assumption that reaction rates are first order.

TABLE 5

Decomposition of hydrogen peroxide in aqueous Fe solutions

Fe concentration (mg/L)		Fe-salt	Autoclaved	Additive (Conc., mg/L)	Peroxide conc. (mg/L)	Decomposed (%)	
Total	Dissolved[a]					12 h	24 h
57	~52	$FeSO_4$	No	-	11000	81	~100
36	<1	$FeSO_4$	No	-	1400	26	27
36	<1	$FeSO_4$	Yes	-	1400	25	25
32	21	$FeCl_3$	No	-	1800	78	86
36	<1	$FeCl_3$	Yes	-	1800	25	25
57	~52	$FeSO_4$	No	Na_3PO_4 (1640)	12000	25	27
57	~52	$FeSO_4$	No	TMP (306)	13000	60	70
57	~52	$FeSO_4$	No	Citric acid (206)	12000	73	85
36	<1	$FeSO_4$	Yes	Na_3PO_4 (1640)	1800	25	25
36	30	$FeSO_4$	No	Na_3PO_4 (1640)	1100	38	39

[a]Concentration in solution after filtration through 0.22-μm membrane filter.

position to varying extents, depending on the degree of Fe-complexation by the additive.

In solutions subjected to autoclaving, the decomposition of peroxide decreased to ~25% over 24 h. The precipitation of Fe-oxide/hydroxide as a result of heating may have removed most of the ion from solution and, therefore, prevented the reaction with peroxide. Analysis of a filtered (0.22-μm) sample indicated <1 mg/L dissolved Fe, compared with an Fe concentration of 36 mg/L in the stock solution. A new $FeSO_4$ solution was, therefore, prepared and a precipitate was formed by increasing the pH to ~10. The solution was thoroughly mixed to maximize precipitation and the pH was adjusted slowly to ~7. The stability of hydrogen peroxide in this experiment was similar to that in the autoclaved $FeSO_4$ solution. Analysis of dissolved Fe concentration again indicated <1 mg/L Fe.

Based on the above experiments, it can be concluded that the inorganic catalysts of peroxide decomposition are most effective in their dissolved form. Once the dissolved catalysts are removed from solution by precipitation or are complexed with ligands such as phosphate, TMP, and citrate, the rate of peroxide decomposition decreases significantly. The extent of complexation of the catalysts by the ligands determines the degree of increased peroxide stabilization.

Aquifer simulator experiments

Based on the batch studies, citrate and TMP together appear to deactivate significantly the enzymatic and inorganic catalysts of peroxide decomposition. Thus, citric acid was the primary additive tested in the aquifer simulator ex-

TABLE 6

Input compositions of nutrient solutions used in the aquifer simulator experiment

Time (h)	Solution composition[a] (mg/L)			
	TMP	Citric acid	Na_3PO_4	Peroxide
0	306	206		
52	300	206		900
59	97	67	28	650
123	97	67	28	650
145	97	206	28	650
197[b]	97	-	28	650
218	injection of nutrients stopped for 50 h			
291	306	206	28	1200
323	800 mg/L RESTORE® 375 Formulation			

[a]Other additives (mg/L): KCl - 20; NH_4Cl - 50.
[b]Instead of citric acid, ascorbic acid (352 mg/L) and $CuSO_4 \cdot 5H_2O$ (2.5 mg/L) were added as peroxide stabilizers.

periments (see Fig. 1). The RESTORE 375® formulation was tested to confirm the results obtained from the field demonstration at Eglin Air Force Base [2]. In addition, ascorbic acid was also tested as a peroxide stabilizer.

Nutrient solutions in the aquifer simulator experiments were injected continuously for ~450 h, with a 50-h break after 218 h. The compositions of the input solutions at various times are given in Table 6. Peroxide was almost completely stable over an extended period of time in the input gallery. However, peroxide was never detected even in the first monitoring well only ~6 cm into the soil zone, corresponding to a travel time of ~1.5 h. Measurements of dissolved oxygen indicated that oxygen was consumed rapidly in the contaminated soil because of microbial growth. The presence of citrate may have added to the increased oxygen uptake by providing an additional microbial substrate.

The results of the aquifer simulator experiment are not consistent with those obtained from the batch experiments. The flow-through experiments are likely to be more reliable because in the batch experiments, rapid decomposition of peroxide resulted in bubble formation which displaced the peroxide solution in the soil column, preventing complete soil–solution reaction. Thus the actual reaction time in the batch experiments probably was much lower than the recorded reaction time.

Summary and conclusions

Controlling the rate of hydrogen peroxide decomposition is essential to its

efficient use in the *in situ* bioreclamation process. A substantial body of literature exists on hydrogen peroxide decomposition and stabilizers under a variety of laboratory conditions. However, much less information exists on the behavior of peroxide stabilizers in groundwater. The most common approach to stabilizing hydrogen peroxide injected into groundwater is by using phosphate. Hinchee et al. [1,20], however, report a failure of this approach at a JP-4 contaminated site on Eglin Air Force Base, Florida.

The laboratory studies reported in this paper used batch experiments to screen a variety of known hydrogen peroxide stabilizers as possible additives to reduce the rate of decomposition in soils from the Eglin site. Results of the batch experiments led to large-scale aquifer simulation testing where primarily citric acid was used to reduce the rate of hydrogen peroxide decomposition to acceptable levels. These tests were unsuccessful and hydrogen peroxide decomposition was complete in less than 6 cm of the soil zone. These results suggest that the efficient use of hydrogen peroxide as an oxygen source for *in situ* bioremediation in soils from the Eglin site does not appear possible using a wide variety of known stabilizers.

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