

## Degradation of the Herbicide Butachlor by Laboratory-synthesized Nanoscale Fe<sup>0</sup> in Batch Experiments

Hyang Yeon Kim, In Kyung Kim, Tae-Ho Han, Jae Han Shim and In Seon Kim\*

BK21 Hazard Material Management Group, Graduate School of Department of Agricultural Chemistry,  
Institute of Agricultural Science and Technology,  
College of Agriculture and Life Sciences, Chonnam National University, Gwangju 500-757, Republic of Korea

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**Degradation of the herbicide butachlor was investigated using laboratory-synthesized zerovalent iron (Fe<sup>0</sup>). The synthesized zerovalent iron was determined to be nanoscale powder by scanning electron microscopic analysis. To investigate degradation of butachlor using the synthesized nanoscale zerovalent iron, time-course batch experiments were conducted by treating the solution of butachlor formulation with the iron. More than 90% degradation of butachlor was observed by iron treatment within 24 h. The synthesized nanoscale zerovalent iron showed an increase in particle aggregation in the batch tests. Green rust formation and a pH drop in solutions were observed, suggesting that the oxidation of the iron occurred. When the iron was extracted with dichloromethane, a negligible concentration was found in the extract, suggesting that butachlor did not bind to the iron particles. GC/MS analysis detected the dechlorinated product as a major degradation product of butachlor in the solutions. The data indicate that laboratory-synthesized zerovalent iron functioned as a reductant to remove electron-withdrawing chlorine, giving the dechlorinated product.**

**Key words:** zerovalent iron, butachlor, pesticide, herbicide

Butachlor belongs to the chloroacetanilide class of herbicides used as a pre-emergence pesticide in the control of weeds and grasses. Butachlor has been used extensively in agricultural fields in Korea since 1970. The extensive use of butachlor may be a possible cause of environmental concern to soil and water ecosystems. Butachlor is classified as fish-toxicity level I in Korea due to highly toxic to aquatic organisms. Numerous studies have reported that butachlor is positive in bacterial mutagenicity assays,<sup>1)</sup> sister chromatid exchanges in rat,<sup>2)</sup> chromosomal aberrations in Chinese hamster ovary cells or cultured human lymphocytes.<sup>3)</sup> It is also reported that butachlor induces stomach tumors in rats.<sup>4)</sup> The toxicity of butachlor is due to not only parent compound, but also its degradation product such as dialkylquinoneimine.<sup>5, 6)</sup> Although butachlor are known to bind to soil matrixes as does other acetanilide herbicide,<sup>7,8)</sup> it has been also found in river and ground waters at variable concentrations.<sup>9-10)</sup> Groundwater is the most important source of drinking for human consumption in rural areas in Korea. Considering the possibility that butachlor adsorbed to the soil matrix may ultimately reach the water environment, studies are needed to develop techniques for degradation of this herbicide in water system.

Using zerovalent iron (Fe<sup>0</sup>) has proven a cost-effective method for degradation of pesticides in water samples. Fe<sup>0</sup> increases the dechlorination of halogenated and triazine

herbicides.<sup>11,12)</sup> Zerovalent iron treatment can be also applicable to non-halogenated pesticides such as carbaryl and benomyl.<sup>13,14)</sup> Most of studies on the use of zerovalent iron for degradation of pesticides have performed by treating pure chemicals with the iron. However, the use of zerovalent iron for the treatment of pesticide formulation has not been reported elsewhere. In agricultural fields, pesticides are applied in a type of formulation. Target pesticide would adsorb competitively with chemicals on the iron surface then chemicals in the formulation would affect the reaction of the iron for degradation of target pesticide.

In this study, we examined degradation of butachlor by laboratory-synthesized zerovalent iron. The commercial purchase of zerovalent iron in Korea is somewhat difficult, as most of countries that produce zerovalent iron do not sale to other country. Thus, the preparation of laboratory-synthesized zerovalent iron would be an alternative approach for degradation of pesticides using Fe<sup>0</sup>. Butachlor degradation by the laboratory-synthesized zerovalent iron was conducted in batch experiments by treating butachlor formulation solution with the iron.

### Materials and Methods

**Chemicals.** Reference standard of butachlor (purity 99%) was purchased from Dr. Ehrenstorfer GmbH (Augsburg, Germany). All solvents used in this study were HPLC grade and purchased from Fisher Scientific (Pittsburgh, PA, USA). Butachlor formulation was commercially available in Korea.

\*Corresponding author

Phone: +82-62-530-2131; Fax: +82-62-530-2139

E-mail: mindzero@jnu.ac.kr

All other chemicals were of analytical grade and commercially available, unless otherwise stated.

**Synthesis of zerovalent iron.** Zerovalent iron was prepared by dropwise addition of 1.6 M  $\text{NaBH}_4$  solution to 1 M  $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$  solution at an ambient temperature with magnetic stirring as previously described.<sup>15)</sup> The solution was centrifuged at  $8\,000 \times g$  for 10 min to obtain the  $\text{Fe}^0$  precipitates. The  $\text{Fe}^0$  precipitates were then washed twice with acetone and dried at  $110^\circ\text{C}$  for 6 h under a gentle stream of  $\text{N}_2$ . The resultant black zerovalent iron powder was then stored in  $\text{N}_2$  purged desiccators until use.

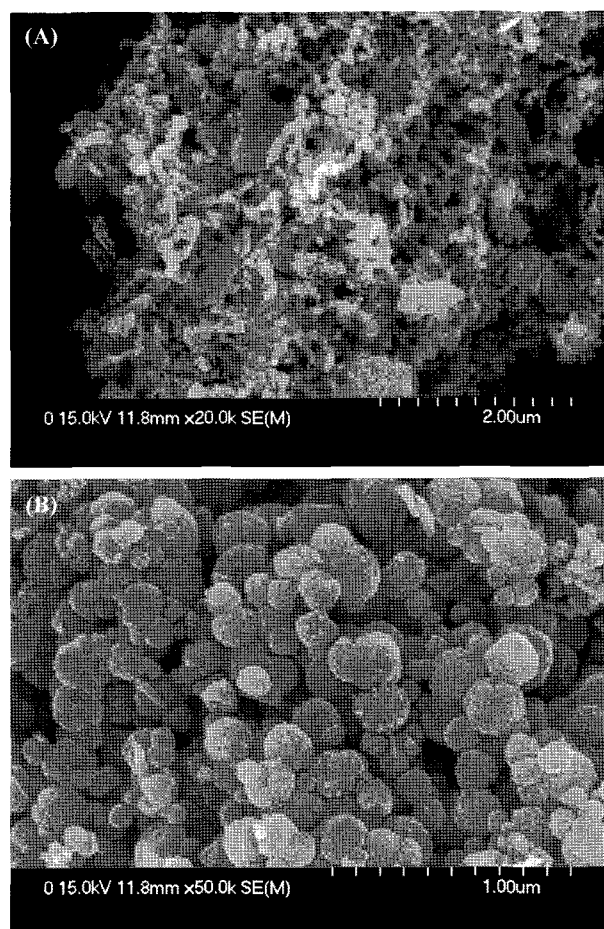
**Batch experiments.** Batch experiments were performed by incubating butachlor formulation solution with the synthesized zerovalent iron. About 0.5 g of the iron was added to triplicate 50 ml centrifuge tubes containing 30 ml of pesticide-formulation solution, Macet EC (58.5%), at the rate of  $20 \text{ mg L}^{-1}$ . Optimum amount of zerovalent iron for reacting with 30 ml of the butachlor solution was determined to be about 0.5 g in preliminary experiment. The centrifuge tubes were capped tightly and incubated at ambient temperature on a shaker at 150 rpm. Control experiments were conducted without  $\text{Fe}^0$  or the herbicide solution. Triplicate tubes were taken periodically and centrifuged at  $8\,000 \times g$  for 10 min. The supernatants were transferred to 100 ml-separatory funnels and extracted with 50 ml of dichloromethane twice. The organic phase was collected and dehydrated over anhydrous sodium sulfate. The extracts were then evaporated on a vacuum evaporator at  $40^\circ\text{C}$ . The residues were dissolved finally in 2 ml of methanol and analyzed by gas chromatograph. The concentrations of butachlor in the samples were determined quantitatively on the basis of calibration curve.

**Analytical methods.** Gas chromatographic analyses were conducted using a Hewlett Packard 6890 model gas chromatograph (GC) equipped with  $^{63}\text{Ni}$  electron capture detector (ECD). The column was an HP-5 capillary column (0.25 mm i.d.  $\times$  30 m length,  $1.0 \mu\text{m}$  film thickness) and the carrier gas was helium at the flow rate of 1 ml/min. Injection was made in split mode of 50 : 1. The column temperature was hold at  $100^\circ\text{C}$  for 2 min and increased to  $280^\circ\text{C}$  by the rate of  $10^\circ\text{C}/\text{min}$ . The injection port and detector temperatures were 250 and  $300^\circ\text{C}$ , respectively. To determine the degradation products of butachlor, gas chromatograph-mass spectrometry (GC/MS) analysis was conducted using an Agilent model 5973N system with HP-5MS capillary column (0.25 mm i.d.  $\times$  30 m length,  $1.0 \mu\text{m}$  film thickness). GC conditions for mass spectrometric analysis were the same as described above. The mass spectrometer was run in electron ionization (EI) mode at 70 eV electron. Mass value was scanned from 50 to 500 amu at 2.94 sec per scan with mass resolution 10,000. The scanning electron microscopy analysis was conducted using a JEOL model JSM-840A SEM (Japan) as a modified method from the previous study.<sup>16)</sup>

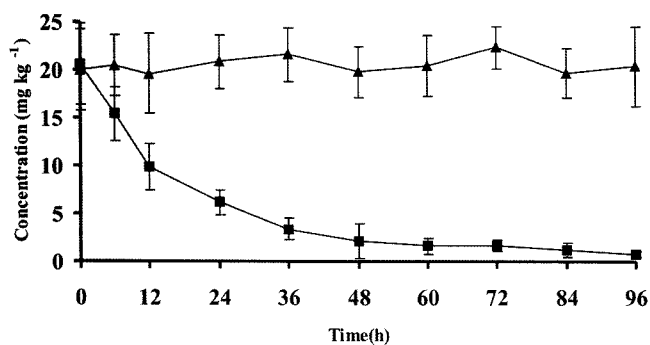
## Results and Discussion

The scanning electron microscopy (SEM) data of zerovalent iron synthesized in this study are presented in Fig. 1. The SEM analysis shows that the synthesized zerovalent iron particles are in a size range of 10-100 nm (Fig. 1A), which suggests that our zerovalent iron is nanoscale iron particles. An increase in particle aggregation of the iron was observed after reaction with the butachlor formulation solution (Fig. 1B). This increase in the particle aggregation was suggested to be due to the formation of iron (III) oxide/hydroxide in the reaction solution, as demonstrated in other studies.<sup>17-20)</sup> The oxidation of zerovalent iron in the aqueous solution produces ferrous iron, and the resulting ferrous ion reacts to give magnetite ( $\text{Fe}_3\text{O}_4$ ), ferrous hydroxide ( $\text{Fe}(\text{OH})_2$ ) and ferric hydroxide ( $\text{Fe}(\text{OH})_3$ ).<sup>21)</sup> It is, therefore, suggested that the aggregation of the iron was resulted from oxidation in the reaction solution.

The data for time-course degradation of butachlor by the laboratory-synthesized nanoscale zerovalent iron are presented



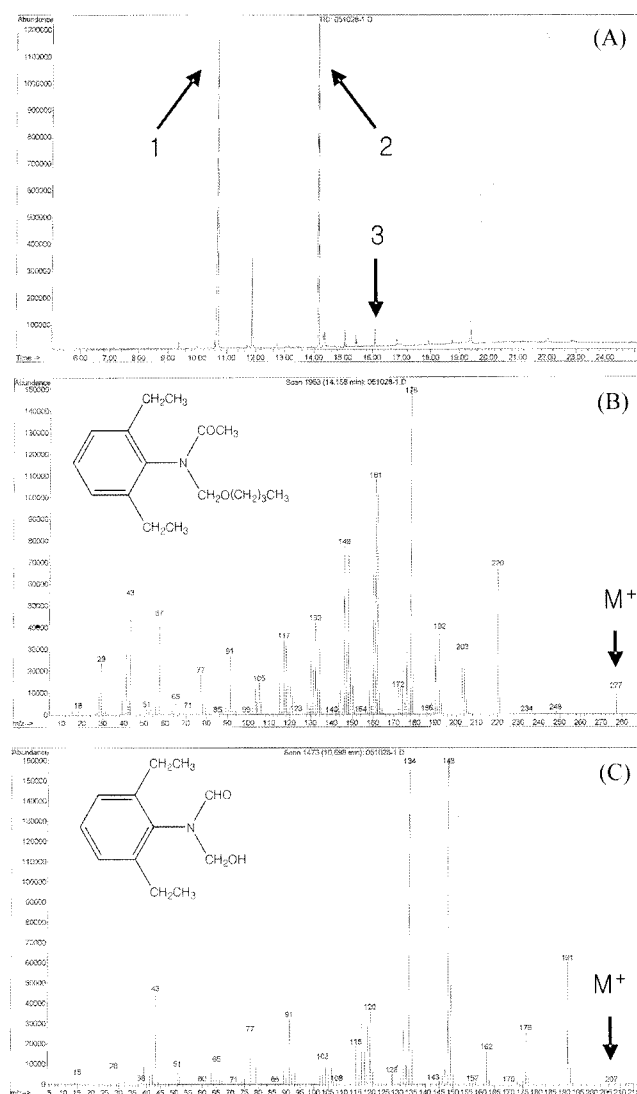
**Fig. 1.** Scanning electron microscopy image of laboratory-synthesized zerovalent iron particles before (A) and after reaction with the pesticide solutions for 24 h (B).



**Fig. 2.** Time-course degradation of butachlor in the pesticide solutions treated without (▲) and with (◁) zerovalent iron. Each value represents mean  $\pm$  SD ( $n = 3$ ).

in Fig. 2. Significant degradation of butachlor was observed in the reaction solutions treated with zerovalent iron. Butachlor was degraded continuously with time by iron treatment, giving more than 95% degradation in a 60 hr-reaction period. In contrast, the control samples without iron treatment showed a negligible degradation of butachlor throughout experiments. These data indicate that butachlor degradation was due to zerovalent iron reaction in the solutions. The reaction solutions became black during incubation. The pH of the reaction solution was decreased from approximately 6.5 to 4.3 and the formation of green rust in the solution was observed, demonstrating that the oxidation of zerovalent iron occurred. The pH drop and green rust formation in the zerovalent iron solution have been also well established in other studies.<sup>11, 22</sup> Previous study indicated that the formation of green rust by zerovalent iron treatment enhanced the reduction of chlorine-containing herbicides.<sup>23</sup> The data in this study, therefore, could demonstrate that the synthesized iron functioned as a reductant to produce magnetite (Fe<sub>3</sub>O<sub>4</sub>) and green rust.

Someone may wonder if the time-course corresponds to degradation of butachlor by zerovalent iron treatment are results of butachlor adsorption to the surfaces of reaction tubes and zerovalent iron. However, concentrations of butachlor in the control samples were constant with no significant fluctuation, which suggested that butachlor dissolved in the solutions did not bind to the surfaces of reaction tubes and zerovalent iron. In this study, butachlor was treated in a type of formulation at the rate of water solubility. The added butachlor would become more water soluble due to the presence of surfactant in the solutions. The presence of surfactants in the solution would allow butachlor concentration to maintain the aqueous solubility. To investigate if butachlor bound to zerovalent iron, the iron was extracted with organic solvent and then the extract was subjected to gas chromatographic analysis. A negligible butachlor concentration less than 0.05 mg/kg was detected in the extract of the iron particles, suggesting that adsorption/desorption behavior of the herbicide did not affect its degradation. Our main emphasis in this study is that zerovalent iron treatment resulted in a significant degradation of butachlor in aqueous solutions.



**Fig. 3.** Total ion chromatogram (A) of the extract obtained from the pesticide solution treated with laboratory-synthesized zerovalent iron for 24 h and GC/MS spectra of dealkylated (B) and dechlorinated product (C) of butachlor. The chromatograms in Fig. 1A represent dealkylated product (1), dechlorinated product (2) and butachlor (3), respectively.

To analyze the degradation products of butachlor in the reaction solutions, the solutions were extracted with dichloromethane and analyzed by GC/MS. M<sup>+</sup> peak at  $m/z$  277 was identified as a major degradation product of butachlor by GC/MS analyses (Fig. 3B), which showed one chlorine difference compared to the ion spectrum of butachlor ( $m/z$  311). Main fragment ion spectrums of the peak at  $m/z$  277 were  $m/z$  220, 178, 161, 146 and 131. The base peak at  $m/z$  178 was due to the loss of  $-\text{CO}(\text{CH}_2)_2$  from  $m/z$  277. The fragment ion spectrum of  $m/z$  220 was identified by its  $(\text{M}-\text{C}_4\text{H}_8)^+$ . The fragment ion spectrum of  $m/z$  161 was due to the loss of an OH from  $m/z$  178. These observations suggested that the peak at  $m/z$  277 is a dechlorinated compound of butachlor, suggesting that dehalogenation mechanism of butachlor was occurred by zerovalent iron treatment. Thus, it was suggested

that our zerovalent iron functioned well as a reductant to give dechlorination in the butachlor structure. The oxidation of laboratory-synthesized zerovalent iron generated iron oxide form (Fig. 1). The data demonstrate that electrons would transfer from the iron surface to butachlor to give the dechlorinated product. GC/MS analyses also identified an  $M^+$  peak at  $m/z$  207 as a degradation product of butachlor (Fig. 3C). Main fragment ion spectrums of  $m/z$  207 were  $m/z$  191, 148, 134 and 120. The peak at  $m/z$  207 showed five  $CH_2$  differences compared to the peak at  $m/z$  277, suggesting that five methyl groups were removed from  $m/z$  277. The peak at  $m/z$  191 was due to the loss of oxygen from  $m/z$  207, and the peaks at  $m/z$  148 and 134 were due to the losses of  $-COCH_2O$  and  $-NCOCH_2O$ , respectively, from  $m/z$  207. These observations identified as dealkylated metabolite probably resulted from an abiotic degradation of butachlor after dechlorination.

Researchers have reported enhanced mineralization of the nitroaromatic compound after zerovalent iron treatment,<sup>24-26)</sup> suggesting that zerovalent iron may affect biodegradation processes. Our laboratory-synthesized zerovalent iron may similarly produce more biodegradable products of butachlor, since zerovalent iron removed electron-withdrawing chlorine to give dechlorinated products and a dealkylated metabolite. Numerous studies have reported that butachlor and its metabolites are toxic to organisms.<sup>1-6)</sup> The toxicities of butachlor and its metabolites may be removed by treatment of zerovalent iron. The use of  $Fe^0$  for remediating ground water and soil that has been contaminated with nitroaromatic or organochlorine chemicals have been demonstrated largely.<sup>25-29)</sup> From these studies,  $Fe^0$  proved effective in removing these chemicals from waters and soils. Those studies were carried out by using pure chemicals. In the present study, we examined degradation of butachlor by treating its formulation with laboratory-synthesized nanoscale  $Fe^0$ . Many studies have reported the detection of variable concentrations of acetanilide herbicides such as butachlor in river and ground waters.<sup>30-33)</sup> Our zerovalent iron may be applicable for the remediation of waters contaminated with butachlor.

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