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# The response of rice seedlings to $O_2$ released from $CaO_2$ in flooded soils

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**Summary** The efficacy of $CaO_2$ as an oxygen-supplying seed coating on the emergence of rice seedlings from flooded soils was studied. Seedling response became increasingly dependent on coating rates of 0–40% (by weight of seed loading with 60% $CaO_2$ material) as the depth of planting increased from 0 – 2.5 cm. No emergence occurred from 5 cm.

The emergence response to 40% $CaO_2$ at 1.5 cm depth of planting was inhibited by the presence of 0.25% finely ground rice straw but was stimulated by increasing pH levels. The organic substrate likely increased microbial competition for available $O_2$ while increasing pH levels improved the rate of $CaO_2$ dissolution during the day 2–8 period of germination. Soil type effects on seedling response were speculated to be due to differences in $O_2$ consumption rates between soils. The addition of CaO to the $CaO_2$ coating improved the emergence response on an unlimed acid soil.

## Introduction

The rapid depletion of $O_2$ from a soil upon submergence inhibits the ability of rice seedlings to emerge from flooded soils. While soil $O_2$ levels drop to near zero within 24 hours of submergence [1,13,17], the demand for $O_2$ by rice seedlings greatly increases two to three days after germination[11] creating a period of critical $O_2$ deficit. Restricted $O_2$ availability has been found to slow rice seedling growth rates [7,18], root and shoot growth [2,19], coleoptile greening [4,5,6], and vertical shoot geotropism[3].

Current seeding methods for lowland rice production in the U.S. ensure an adequate supply of $O_2$ for seedling growth by: (1) drilling the seeds into the soil and delaying permanent flooding for 20–30 days until the seedlings are established, or (2) broadcasting pre-soaked seeds into floodwater where they settle on the aerobic soil surface. Drill seeding presents special problems in weed control and nitrogen fertilization while water seeding exposes the seedlings to drift, insect depradation, fungal attack, and nutrient deficiencies prior to firm anchorage in the soil.

There have been some reports [8,9,12] that calcium peroxide ($CaO_2$) can be used as an $O_2$ source for germinating rice seeds if applied as a seed coating. The $CaO_2$ will undergo hydrolysis as follows:

$$CaO_2 + H_2O \rightarrow CaO + H_2O_2$$

The CaO will have a liming effect by reacting with $CO_2$:

$$CaO + CO_2 \rightarrow CaCO_3$$

The $H_2O_2$ released upon hydrolysis will then decompose:

$$2H_2O_2 \rightarrow 2H_2O + O_2$$

This $O_2$ released is able to meet the rice seedlings' demand while germinating in an otherwise anaerobic flooded soil.

The following investigation was designed to define some of the factors affecting the efficacy of this material. We first investigated the role of planting depth and coating rate on the emergence response of rice seedlings in a flooded soil. We further looked at the influence of soil type and pH and the presence of a readily decomposable organic substrate on both seedling response and the dissolution rate of the $CaO_2$ coating.

## Materials and methods

Rice (*Oryza sativa*, cultivar M-9) seeds were coated at rates of 10, 20, 30, or 40% $CaO_2$ material (60% a.i. $CaO_2$) by placing 100 g samples into a plastic pot attached to an inclined rotator. The seeds were tumbled and sprayed with a 50% aqueous solution of poly-vinyl alcohol until evenly moistened. Weighed portions of the $CaO_2$ material were sprinkled on the seeds until evenly coated and the process repeated until the entire amount of $CaO_2$ required had been applied to the sample.

In the first experiment, designed to delineate the interaction of coating rate and planting depth on emergence response, 60 pots (20 cm dia.) were filled with soil (Yolo fine sandy loam, pH sat. paste 6.9, coarse loamy, mixed, thermic Xerofluvents) and each planted with 25 seeds of one of the coating rates listed above plus an untreated control. Planting depth treatments were surface, 1.3 cm, 2.5 cm, or 5.0 cm. There were four replications of each coating rate × planting depth treatment.

The pots were placed in a fiberglass tank in a greenhouse (27°C) and arranged in a randomized complete block design. The tanks were flooded and water maintained to a depth of 5 cm above the soil surface. On the thirteenth day from seeding, the emerged seedlings in each pot were counted, lifted from the soil, and individual heights (mesocotyl to tip of longest leaf) were measured.

The second experiment was designed to investigate the effects of soil type, soil pH, and organic amendment (rice straw) on both the seedling emergence response and the dissolution rate of $CaO_2$ at the 40% coating level. The three soils used were: Yolo fine sandy loam (pH sat. paste 6.9, coarse loamy, mixed, thermic Mollic Xerofluvents), Meyers clay (pH sat. paste 7.0, fine, montmorillonitic, thermic Entic Chromoxererts), and Stockton clay (pH sat. paste 5.4, fine, montmorillonitic, thermic Typic Pelloxererts). Each soil was adjusted to pH levels of 5.4, 7.0 and 8.1 through stepwise equilibration with CaO or 3*N* $H_2SO_4$. The soils were moistened after the addition of each material and the pH level measured after a three day period. The process was repeated until the designated pH levels had been attained.

Three plastic flats (50 × 25 × 6 cm) were filled with each soil type × pH treatment to a 3-cm depth. A divider was placed across the center of each flat and finely ground rice straw added at the rate of 0.25%

to one side. Sixty seeds coated with 40% $CaO_2$ were placed in three rows 3 cm apart in each side of the flats.

Coated seeds were prepared for periodic removal and residual $CaO_2$ assay by enclosing groups of ten seeds in polyester coarse-mesh fabric. The coated seeds were placed in a row on strips (2.5 × 10 cm) of fabric, the fabric then folded over the seeds and sealed with a hot iron. Four groups of these fabric-enclosed seeds were placed on each side of each flat. All flats then had an additional 1.5 cm of soil added over the seeds.

The flats were arranged in fiberglass tanks in a greenhouse (27°C) in a randomized complete block design with three soil types, three pH levels, and two organic amendment treatments factorially applied and replicated three times. The tanks were flooded and water maintained 5 cm above the soil level.

Daily counts were made of coleoptile emergence from each treatment. At days 2, 3, 5, and 8 after planting, a group of fabric-enclosed seeds was removed from each treatment, the fabric cut open, and the samples placed in a 125-ml erlemeyer flask containing 0.3 $N$ $H_2SO_4$. The peroxide equivalence of the residual $CaO_2$ on the group of seeds was measured by titration with 0.1 $N$ $KMnO_4$ and reported as g $CaO_2$/10 seeds.

In the third experiment, additional liming materials were added to the $CaO_2$ seed coating to test their efficacy in enhancing the emergence response in an acid soil. Seeds coated with 40% $CaO_2$ were left further untreated or were additionally coated with 20% $CaCO_3$ or CaO. Forty seeds of each treatment were planted (1.5 cm deep) in two rows replicated three times in flats filled with untreated Stockton clay soil. The flats were flooded as previously described and emergence counted at daily intervals.

## Results and discussion

### *Coating rate and depth of planting*

The emergence and plant height responses of rice seedlings to coating rates of $CaO_2$ and depths of planting are shown in Table 1. Both of these indicators of seedling vigor were stimulated by increasing rates of $CaO_2$ coating and the optimal rate of coating increased with increasing depth of planting. While seedling emergence decreased with depth of planting, seedling height was greater for the 1.3 cm depth than for the surface planting. Of agronomic interest is that an optimal coating of $CaO_2$ affected comparable emergence and greater seedling height for seeds planted 1.3 cm deep in a flooded soil as compared to untreated, surface planted seeds.

It is apparent that seedling emergence was restricted at the 2.5 cm depth of planting, requiring a 40% $CaO_2$ coating for maximum emergence in this trial. Linear regression of seedling emergence against level of coating at this depth of planting ($r = 0.97$) indicates that a coating rate of 55% $CaO_2$ would be necessary for an emergence percentage similar to the untreated surface planted treatment. Additionally, no emergence was evidenced from the 5.0 cm planting depth. Depth of planting obviously places a restriction on the efficacy of this seeding technique.

### *Soil factors*

The effects of various soil factors on the emergence response are summarized

Table 1. Rice seedling emergence and height measured 13 days after planting in a flooded soil as affected by coating rate of $CaO_2$ and depth of planting

| Depth of planting | $CaO_2$* loading rates on rice seed – % by weight<br>0 | 10 | 20 | 30 | 40 |
|---|---|---|---|---|---|
| *Seedling emergence (of 25 seeds)*** | | | | | |
| Surface | 17.5 a | 22.3 b | 23.0 b | 23.0 b | 22.3 b |
| 1.3 cm | 2.8 a | 14.8 b | 18.3 c | 18.0 c | 17.0 bc |
| 2.5 cm | 1.0 a | 5.5 b | 9.0 c | 9.3 c | 13.3 d |
| *Seedling height (cm)*** | | | | | |
| Surface | 16.96 a | 20.75 b | 20.91 b | 20.44 b | 21.39 b |
| 1.3 cm | 17.27 a | 20.86 b | 23.84 c | 24.93 c | 23.10 bc |
| 2.5 cm | 8.48 a | 11.92 b | 15.24 c | 18.09 d | 21.66 e |

* $CaO_2$ material was 60% purity.

** Values followed by the same letter at each planting depth are not significantly different at the .05 level of probability according to DMRT.

in Figs. 1–3. Firstly, the presence of 0.25% rice straw was found to restrict the emergence of rice seedlings in each soil and at each pH level. This inhibitory effect of rice straw was probably due to a stimulation of microbial activity which acted to increase the competition for $O_2$ released upon decomposition of $CaO_2$. This would naturally lower the $O_2$ supply available for the germinating rice seedlings. Although rice straw additions are known to stimulate microbial production of organic acids and carbon dioxide in flooded soils[15], and these products are sometimes deleterious to the growth of young rice seedlings[14], the inhibition of seedling emergence was evidenced here much too soon after flooding to be accounted for by these factors.

The level of soil pH also had a strong influence on emergence response (Figs. 1–3). Higher pH consistently enabled the seedlings to exhibit a greater emergence response. This effect is shown most clearly in Fig. 1 where, on the Meyers clay, increasing pH is shown to improve the emergence response regardless of the rice straw treatment. Of particular significance is the effectiveness of increasing pH levels in overcoming the inhibitory effects of straw additions.

The stimulatory effect of pH may be a direct effect on seedling growth, as has previously been found for rice seedlings[14], but this is difficult to assess due to the influence that the $CaO_2$ coating would have on the effective pH in the soil solution surrounding the germinating seedlings. While a direct effect cannot be discounted, we must consider the effect of pH on the performance of $CaO_2$ and

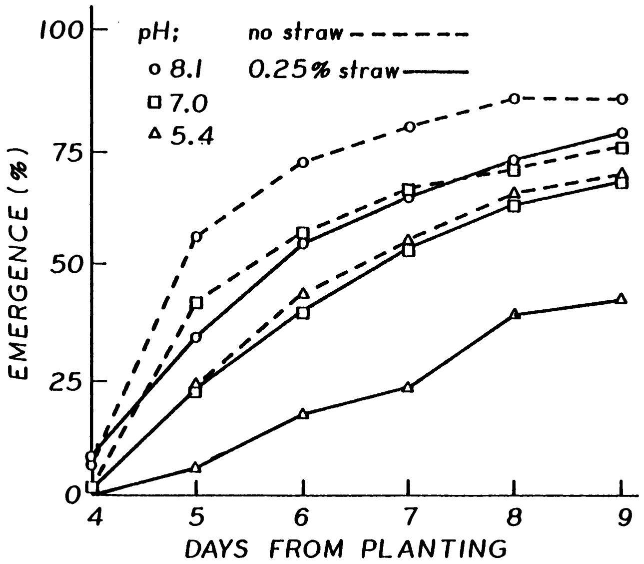

Fig. 1. The emergence response of rice seeds to 40% $CaO_2$ coating in a flooded Meyers clay at 3 pH levels, with and without straw added.

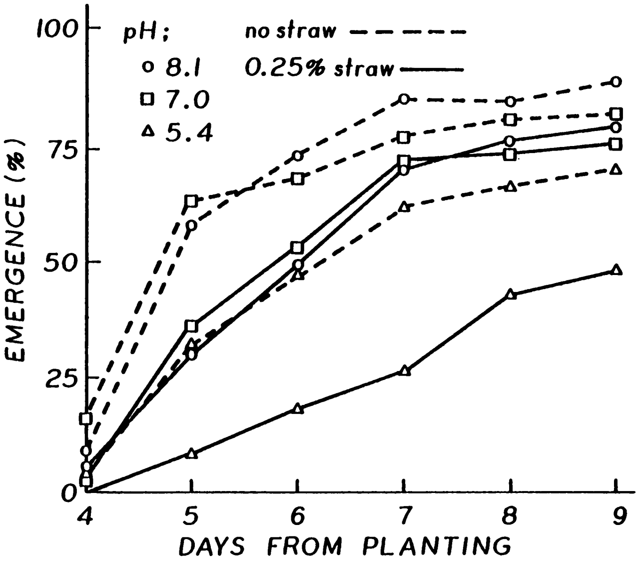

Fig. 2. The emergence response of rice seeds to 40% $CaO_2$ coating in a flooded Yolo fine sandy loam at 3 pH levels, with and without straw added.

how the supply of $O_2$ might be affected by various pH levels. These factors will be discussed in the next section.

The soil types were found to differ significantly in terms of seedling response. Most obvious is the poorer performance on Stockton clay (Fig. 3) as compared to the other two soils. Seedling emergence was especially sensitive to the presence of

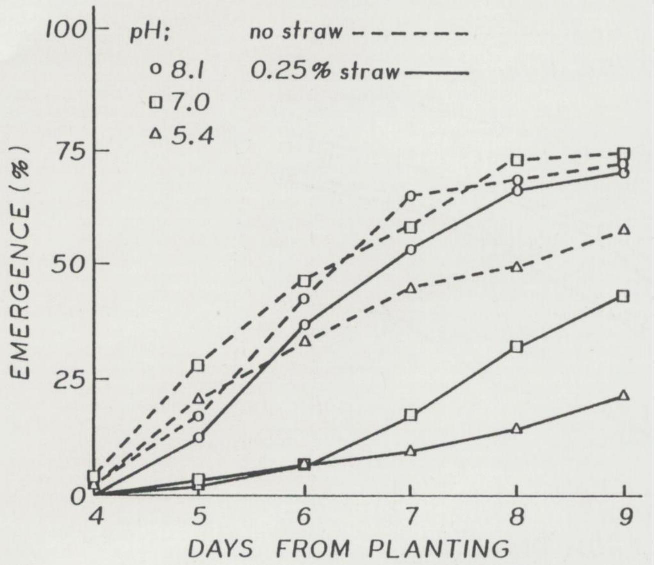

Fig. 3. The emergence response of rice seeds to 40% $CaO_2$ coating in a flooded Stockton clay at 3 pH levels, with and without straw added.

straw on this soil and was inhibited across all pH and straw treatments when compared to the Meyers and Yolo soils. It is possible that the rate of $O_2$ consumption in the flooded Stockton soil was greater than for the other two soils, leading to a greater competition for $O_2$ released from the seed coatings. Although not measured here, such processes as microbial respiration and the chemical oxidation of $Fe^{+2}$ would be factors in such an effect[16].

*Dissolution of $CaO_2$*

The dissolution of $CaO_2$ from the seed coatings was found to occur very rapidly, with less than 50% of the original material remaining after three days of flooding (Fig. 4). There were highly significant differences between pH levels in the initial rate of dissolution with decreasing pH levels affecting greater rates. By day 8, however, the level of $CaO_2$ remaining undissolved on the seeds was unaffected by pH.

In terms of the germinating seed, the period of interest would most logically be that occurring from the time that its demand for $O_2$ becomes significant, 2–3 days after germination begins[11], to the time that the coleoptile has emerged and greening is evident. In this regard, it is seen in Fig. 4 that the amount of $CaO_2$ dissolved from day 2 through day 8 was greater with increasing pH levels. It is speculated, then, that the effect of pH on the emergence response of $CaO_2$ coated seeds is due to an effect on the dissolution of $CaO_2$ during this critical period.

Comparisons are made between emergence response and $CaO_2$ dissolution from day 2 through day 8 for each unamended soil in Table 2. For the Stockton

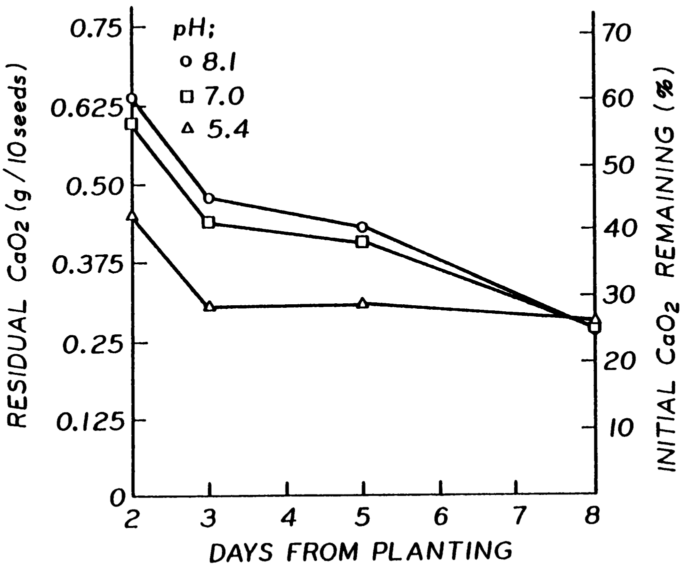

Fig. 4. Residual $CaO_2$ as a function of time on rice seeds planted in flooded soils at 3 pH levels (initial $CaO_2$ coating rate at 40% of seed weight).

Table 2. The relationship between $CaO_2$ dissolved from day 2–8 of germination and emergence of $CaO_2$ coated rice seedlings at day 8 in three unamended flooded soils at three pH levels

| Soil | pH | $CaO_2$ dissolved from day 2–8 (g/10 seeds) | Emerged seedlings day 8 (out of 60 seeds) | Regression equation for each soil y = emergence; x = $CaO_2$ dissolved | $r^2$ values |
|---|---|---|---|---|---|
| Stockton | 5.4 | .24 | 30 | | |
| clay | 7.0 | .43 | 42.7 | y = 65.3x + 14.28 | .997** |
| | 8.1 | .41 | 40.7 | | |
| Yolo fine | 5.4 | .10 | 40.7 | | |
| sand loam | 7.0 | .30 | 48.3 | y = 21.9x + 39.6 | .863** |
| | 8.1 | .52 | 50.0 | | |
| Meyers clay | 5.4 | .19 | 40.3 | | |
| | 7.0 | .30 | 43.3 | y = 11.8x + 42.0 | $.014^{ns}$ |
| | 8.1 | .23 | 51.0 | | |

clay and Yolo fine sandy loam, a significant positive relationship was found between these two variables. This relationship was not significant on the Meyers clay. The high $r^2$ value and steep slope of regression equation for the Stockton clay indicate that seedlings were especially sensitive to the dynamics of $O_2$ availability in this soil.

*Addition of lime to the coating*

When CaO was added as a final coating to the $CaO_2$ coated seeds, the seedling emergence response was significantly improved on the unlimed Stockton clay over that for seeds coated with $CaO_2$ alone or supplemented with $CaCO_3$ (Fig. 5). Evidence presented here indicates that the CaO probably acted by raising the pH in the soil solution surrounding the seed and thus affected a greater availability of $O_2$ for the germinating seeds during the period of greatest need. The CaO treatment was more efficacious than was the $CaCO_3$, possibly because of a greater liming potential. The addition of CaO to $CaO_2$ coatings for rice seed may prove beneficial in applying this seeding technique to acid soils or situations where microbial respiration is high.

The results of this investigation indicate a clear need for an $O_2$ extender such as $CaO_2$ if rice seedlings are to emerge from a planted depth in a flooded soil. Certain constraints exist which may limit the use of this technique, however. Planting depth was found to be critical, with efficacy falling off sharply below 1.3 cm. Optimal coating rates were found to increase with increased planting depth as well. In applying this technique to an agronomic situation, the accuracy with which coated seeds can be planted at a certain depth and the cost of the $CaO_2$ material should prove to significantly interact on its feasibility.

Additionally, factors such as soil type and pH and the presence of readily decomposable organic materials will place limits on the use of $CaO_2$. We've shown that these factors interact with one another and that the dynamics of when the $O_2$ is made available to the seed plays an important role. There is further

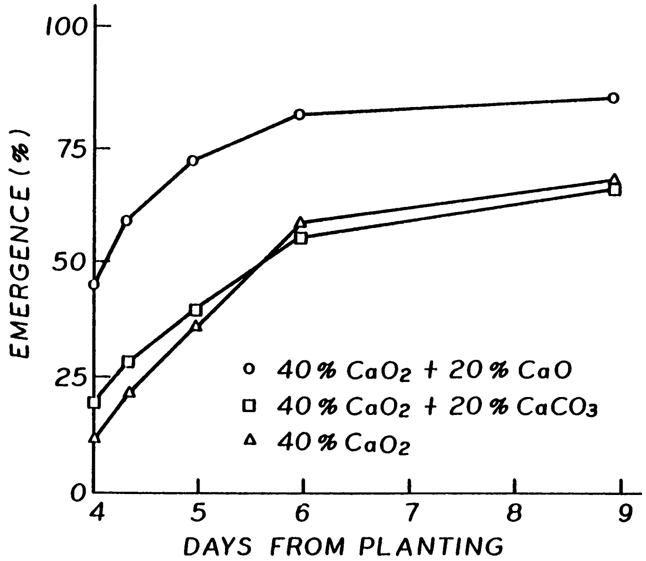

Fig. 5. Emergence response of rice seeds to 40% $CaO_2$ coating in a flooded Stockton clay (pH 5.4) as affected by the addition of liming materials to the coating.

indication that these factors can be manipulated to increase the seedling response.

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