Diffusion of Gaseous Sulfur Dioxide into Corn Grain

S. R. ECKHOFF and M. R. OKOS

ABSTRACT

The diffusion path of gaseous sulfur dioxide into sound yellow dent corn was explored. The iodine-staining procedure used gave qualitative indication that the SO₂ enters primarily through the tip cap in whole intact kernels. The SO₂ enters at the tip cap, moves up through the area between the pericarp and seed coat, and then diffuses into the endosperm. A second pathway up through the germ into the endosperm was also observed. Complete penetration of gaseous SO₂ into the kernel can be achieved in about 2 min. Damage to the pericarp increases the amount of SO₂ taken up by the grain.

The diffusion path of gaseous or liquid materials into and through cereal grains is complex, as would be predicted based upon the structure of cereal grains. Although many researchers have modeled diffusion in cereal grains using a spherical diffusion mode (Fan et al. 1965, Park and Kyle 1975, Hsu 1984), Cox et al. (1944) and Wolf et al. (1952) showed that liquids tend preferentially to enter the tip cap or attachment point of the grain, diffuse up through the space between the pericarp and seed coat, and then diffuse into the endosperm of the grain. The seed coat is semipermeable to organic and inorganic materials, allowing water to diffuse through but inhibiting flow of other liquids until structural changes are induced in the seed coat by the diffusing chemical (Beesko 1924). Gaseous SO₂ has been shown to be effective in controlling microbial growth during the low-temperature drying of corn (Eckhoff et al. 1983, 1985; VanCawenberge et al. 1979). Microbial infection of grain is not homogeneous, and the location of the organisms depends, among other factors, upon the particular invading organism (Semeniuk 1954). The efficacy of SO₂ treatment could be totally dependent upon the location of the organisms and the ability of the SO₂ to diffuse to those locations. This research was done to determine the diffusion path of gaseous SO₂ into corn grains in order to better understand the diffusion process.

Procedures

Yellow dent corn, Cargill 1967, was hand-harvested at 30% moisture content and hand shelled. Samples were stored at −16°C until the test was run. Grains that appeared sound and showed no visible cracks or defects were selected for the experiment. The grains were exposed to a stream of pure gaseous sulfur dioxide at room temperature for 30, 60, and 120 sec. After treatment, the kernels were removed and sliced into sections using a razor blade. The sliced kernels were dipped into an iodine solution (0.002N). Sliced sections were then blotted on tissue to remove excess iodine, and the exposed endosperm and germ were photographed using 35 mm color film. Because iodine reacts with sulfur dioxide to form a colorless compound (Schroeter 1966), the presence of SO₂ in sufficient quantity to react with the iodine taken up by the grain during the dipping results in normal coloring of the kernel. Excess or unreacted iodine reacts with the starch in the kernel to produce a dark colored complex, thus, this staining procedure is a qualitative rather than quantitative measure of the presence of SO₂.

The entire procedure (treating, slicing, dipping in iodine, and photographing) was carried out at timed intervals to insure reproducibility. Ten seconds after the kernel was removed from the gas stream it was dipped into the iodine for 10 sec. The blotting of the excess iodine and positioning took an additional 10 sec, so that the kernels were positioned and ready to be photographed 30 sec after being removed from the gas stream. When multiple kernels were to be photographed at the same time, the length of time from removal from the gas stream to dipping increased to 30 sec and positioning time for the photograph also increased so that total time increased to 1.5–2 min. Photographs of the same kernels taken at different times after removal from the SO₂ gas stream were achieved by shutting the lights off on the photographic stand and

1Associate professor, Agricultural Engineering Department, University of Illinois, Urbana 61801.
2Professor, Agricultural Engineering Department, Purdue University, West Lafayette, IN 47907.

©1989 American Association of Cereal Chemists, Inc.
leaving the kernels on the surface.

To help study the entrance point of SO$_2$ into corn kernels, some kernels were dipped into liquid paraffin 24 hr prior to being exposed to SO$_2$. The kernels were dipped in approximately one third of their length such that the tip cap was completely covered and approximately one half of the germ face was covered. Also, certain kernels not coated with paraffin were cut with a razor blade deep into the endosperm area of the kernel. The cut was to show the effect of cracked or damaged kernels on the rate of SO$_2$ sorption.

**RESULTS AND DISCUSSION**

Figure 1 shows four kernels exposed to pure SO$_2$ gas for different lengths of time. The exposure times were (from left to right) 120, 60, and 30 sec, or no exposure. There are visible differences between the kernels due to exposure time. The untreated kernel was totally dark as expected, and as exposure time increased, the amount of area lightened by the SO$_2$ increased until after only 2 min the SO$_2$ had penetrated well into the kernel. This rapid transfer of pure SO$_2$ into the grain suggested that long exposure to pure SO$_2$ may be undesirable and inefficient.

Figure 1 also displays the same four kernels photographed at different times after being removed from the SO$_2$ gas. Figure 1A was taken 2 min after the iodine was applied, Figure 1B was taken after 5 min, and Figure 1C after 8 min. This series of photographs shows several interesting phenomena. This is demonstrated most clearly in the 60-sec exposure kernel between Figures 1A and 1B. The increased time allows the SO$_2$ in the kernel to diffuse further, since immediately after removal from the gas stream there are large SO$_2$ gradients established in the kernel. During the cutting, dipping, and positioning of the kernel for photography, some of the gradient is dissipated as the SO$_2$ diffuses further into the kernel, vaporizes from the kernel, or reacts with corn constituents. However, some SO$_2$ gradients remain, and the movement of SO$_2$

---

**Fig. 1.** Kernels exposed to sulfur dioxide for different times (from left to right: 120, 60, and 30 sec exposure, and no exposure) and dipped in iodine to show path of diffusion. Darkened areas indicate low or no sulfur dioxide. A, Photo taken 2 min after iodine was applied. B, Photo taken 5 min after iodine was applied. C, Photo taken 8 min after iodine was applied.

**Fig. 2.** Penetration of sulfur dioxide into kernels treated to show diffusional barrier or pericarp. Darkened areas indicate low or no sulfur dioxide: A, no exposure to sulfur dioxide; B, 60-sec exposure; C, kernel with deep endosperm cut exposed to sulfur dioxide for 60 sec; and D, kernel with tip cap coated with paraffin and exposed to sulfur dioxide for 60 sec.
can be observed. This rate of lightening of the kernel can serve as only a rough qualitative measure of the direction and magnitude of diffusion occurring within the kernel, because the slicing of the kernel has modified gradients within the kernel and may allow for other transfer mechanisms to become important. A secondary reaction with iodine also occurs that causes it to lose its color with time, as can be observed in the untreated kernel. The area near the center of the kernel by the germ lightened with time, as can be seen in Figure 1C. This reaction is considerably slower than the change observed from diffusion.

Figure 1 also shows the pathway that SO$_2$ follows as it penetrates the kernel. The SO$_2$ appears to enter primarily through the tip cap, shown by the lack of coloration in the area near the tip cap. As Hoseney et al. (1983) showed for popcorn, the pericarp has a higher resistance to diffusion than does the endosperm area. Although popcorn is unusual in experiencing a very short period of high temperature, the data still indicate the relatively higher resistance of the pericarp. SO$_2$ also appears to diffuse rapidly around the outer area of the endosperm, in agreement with the findings of Cox et al. (1944); using liquid demonstrated that sulfuric acid enters the tip cap area and diffuses around to the top of the kernel before starting to penetrate through the aleurone layer into the endosperm. The lightened (reacted) area between the endosperm and the pericarp appears to be larger than could be accounted for by just the area between the seed coat and the pericarp as suggested by Cox, Cox et al. (1944), Beeskos (1924), and Freyberg (1931) indicated that seed coats are semipermeable such that water is allowed to pass through but other inorganic or organic substances are denied passage. Our findings indicate that the seed coat offers some increased resistance to gaseous SO$_2$ transport but does not act as a true semipermeable membrane.

The increased resistance of the seed coat may result from the obvious difference in cell density of the areas. The cross and tube cells are large, loosely packed, thin-walled cells that are designed to facilitate the movement of material through them. The aleurone layer has no large intercellular spaces and thus offers greater resistance to diffusion (Wolf et al. 1952). Another factor that helps to explain the apparent semipermeability is the difference in composition and reaction of the two areas. The aleurone layer is high in protein and fat, whereas the bran is relatively low in protein but higher in ash and fiber (Kent 1975). If there is binding or reaction between the diffusing material and the substrate (i.e., the protein or fat), then the apparent diffusivity or movement of the material into the kernel depends upon the rate of reaction relative to the rate of diffusion. In such a case, if the SO$_2$ rapidly reacts with the aleurone layer, the SO$_2$ will not appear to be diffusing into the endosperm. Only when all the reaction sites have been used with the SO$_2$ species appear to diffuse into the endosperm.

To confirm that SO$_2$ enters the kernel through the tip cap, tip caps were covered with paraffin and exposed to SO$_2$ for 60 sec. The paraffin acted as a diffusion barrier over the tip cap area. Three kernels exposed to SO$_2$ for 60 sec are shown in Figure 2 along with an unexposed kernel. Paraffin on the tip cap of the kernel in Figure 2D showed the uptake of SO$_2$, as can be seen by comparing Figure 2D with 2B. The dark area in the germ is characteristic of paraffin-

covered kernels and is presumably due to the reduced diffusion of SO$_2$ through the tip cap. Some SO$_2$ has obviously entered the kernel, as shown by the light areas on the upper germ and on the back side of the endosperm. The pericarp is thinnest over the germ, which would reduce the diffusion resistance at the point. The light area on the back side of the kernel is probably due to a crack in the pericarp.

The kernel in Figure 2C was cut deeply into the endosperm with a razor blade prior to exposure to the SO$_2$. The cut acted as a second entrance point for the sulfur dioxide, and a 60-sec exposure allowed the SO$_2$ to permeate the kernel completely. This adds support to the conclusion that the tip cap is the primary entrance (compare Fig. 2B and C).

Not all paraffin-coated kernels showed the same effect as that shown in Figure 2D. Some kernels showed larger areas of lightness, particularly in the endosperm. The darkened lower germ area covered by paraffin was characteristic of all the kernels. But, as shown in Figure 2D, fissures or cracks in the pericarp would increase the amount of SO$_2$ taken up by the kernel, and it is assumed that such cracks were the cause of the light areas in the endosperm.

Figure 3 shows a kernel exposed to SO$_2$ for 60 sec and then cross-sectioned. The right section in the photograph is from the lower end of the kernel near the tip cap. As expected, high levels of SO$_2$ were present as shown by the lack of color. The middle section taken near the top of the germ shows an interesting effect. The light area in the center of the endosperm indicates that the SO$_2$ had penetrated to that point. It apparently came up into the endosperm by way of the germ. The SO$_2$ is seen entering the germ, and then within the aleurone layer into the endosperm. This is reasonable because the flouros endosperm consists of densely packed polygonal starch granules (Hoseney et al. 1983). Most but not all kernels tested showed this pathway into the endosperm via the germ. The SO$_2$ appears to enter the endosperm by two methods. One is to enter the tip cap, diffuse around the kernel in the area between the pericarp and seed coat, and then diffuse into the endosperm. The second is to diffuse through the germ and enter through the scutellar epithelium between the germ and endosperm. The left section was taken near the top of the kernel. SO$_2$ has diffused into this section in a manner similar to that shown in the center section, although diffusion through the seed coat is more observable because this section is close to the very top of the kernel.

**CONCLUSION**

The results indicate that sulfur dioxide enters the intact corn kernel primarily through the tip cap, diffuses around the kernel in the area between the pericarp and the seed coat, and then diffuses through the seed coat into the endosperm. The grain is fully equilibrated with pure SO$_2$ within 2 min. The seed coat offers resistance to SO$_2$ diffusion into the endosperm. The results of the test also show a second diffusion path into the endosperm. SO$_2$ apparently diffused up through the germ and then entered the endosperm and diffused outward toward the seed coat. Not all kernels tested showed this pathway, which may result from a poorly developed or broken scutellar epithelium between the germ and endosperm. Kernels with apparent surface cracks or other breaks in the pericarp showed increased sorption of SO$_2$. The cracks acted as extra entrance points for SO$_2$. SO$_2$ sorption appears to be most rapid and complete in the germ. This is beneficial, because many fungal infections occur primarily in the germ. There was no area within the kernel that the SO$_2$ could not penetrate.

**LITERATURE CITED**

BEESKOW, H. C. 1924. The Selective Semipermeability of the Seed Coats of Corn. M.S. dissertation. The University of Chicago, Chicago, IL.


ECKHOFF, S. R., TUITT, J. F., FOSTER, G. H., ANDERSON, R. A.,
and OKOS, M. R. 1985. Inhibition of microbial growth during ambient
air corn drying using sulfur dioxide. Trans. ASAE 27:907.
FAN, L. T., CHEN, H. C., SHELLENBERGER, J. A., and CHUNG, D.
S. 1965. Comparison of the rates of absorption of water by corn kernels
FREYBERG, W. 1931. Beitrage zur Kenntnis der physiologie der
HOSENEY, R. C., ZELEZNAK, K., and ABDELRAHMAN, A. 1983.
Chem. 61:466.
PARK, S. W., and KYLE, R. G. 1975. Sorption kinetics and equilibrium of
carbon tetrachloride on wheat. Cereal Chem. 52:611.
SCHROETER, L. C. 1966. Sulfur Dioxide Applications in Foods,
Grains and Their Products. J. A. Anderson and A. W. Alcock, eds.
American Association of Cereal Chemists: St. Paul, MN.
VanCAWENBERGE, J. E., ECKHOFF, S. R., BOTHAST, R. J., and
with the trickle sulfur-dioxide process for drying high-moisture corn.
Trans. ASAE 25:1431.
WOLF, M. J., BUZAN, C. L., MacMASTERS, M. M., and RIST, C. E.
1952. Structure of the mature corn kernel III. Microscopic structure of
the endosperm of dent corn. Cereal Chem. 29:349.

[Received February 10, 1988. Revision received August 16, 1988. Accepted August 19, 1988.]