

# Characterization of a Thin Copper Film to Investigate Microbial Biofilm Formation

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Copper films, 6.7 nm nominal thickness, deposited by physical vapor deposition on germanium substrates appeared as coalescing aggregates of copper atoms when evaluated by atomic force microscopy. Evaluation of the Cu thin film by x-ray photoelectron spectroscopy indicated that the Cu at the surface of the film had the character of a 1+ oxidation state. These features were not significantly changed when the Cu thin film was exposed to ethylene oxide. No Ge signal was detectable prior to XPS depth profiling through the Cu thin film, indicating that the film was continuous. When films of this thickness were deposited on a cylindrical Ge internal reflection element and subsequently submerged in an aqueous medium, there was sufficient transmission of infra-red radiation through the Cu to obtain a significant water absorption band at  $1640\text{ cm}^{-1}$ . The intensity of the water absorption band was found to be very sensitive to changes in thickness of the copper thin film. This feature can be used to study the effect of microbial biofilms and their products on the integrity of oxidized copper surfaces in aqueous environments.

## INTRODUCTION

Although microorganisms have been implicated in many corrosion-related failures of water distribution systems, their precise role has been difficult to establish in a number of cases.<sup>1</sup> Often, corrosion measurements involve the application of unnatural potentials or currents that may influence sensitive microbial processes at the metal surface.<sup>2</sup> In addition, reactions at a metal surface/water interface where microbial biofilms exist are difficult to isolate from those in the bulk aqueous phase.<sup>3</sup> Furthermore, characterization of reaction products at surfaces following removal of the water is complicated by the introduction of artifacts during dehydration.

Attenuated total reflectance infrared (ATR/IR) spectroscopy has experienced wide use in the characterization of substances that adsorb to solid surfaces submerged in liquids.<sup>4</sup> This sampling technique has been modified to study adsorption of material to metal films deposited on internal reflection elements (IRE). Sufficiently thin metallic films have been deposited on IRE to permit adequate penetration of an evanescent wave of internally reflected IR radiation into the surrounding medium to detect the bulk liquid as well as substances adsorbed from the bulk liquid phase onto the metal film.<sup>5,6</sup> This approach has been used to describe interactions between Fe and steric acid<sup>5</sup> and between Cu and bacterial biofilms or their exopolysaccharides.<sup>6-10</sup>

Accurate interpretation of reactions that occur at solid/liquid interfaces depends on a fundamental understanding of the surface properties of the solid as well as the characteristics of the bulk liquid phase. The surface properties of a metal will likely determine the stability of the metal in aqueous environments. For this reason, it is important to characterize the thin metal films deposited on IRE in ATR/IR studies of metal corrosion.

This paper describes the properties of Cu thin films deposited by physical vapor deposition on germanium substrates. These Cu films should be useful in ATR/IR studies to identify mechanisms of biofilm-induced corrosion of copper tubing in potable and industrial water systems.

## EXPERIMENTAL

### Deposition of copper thin films by physical vapor deposition (PVD)

The surface of Ge chips (Exotic Material Limited, Costa Mesa, CA) and Ge IRE (Spectra Tech, Stamford, CT) were polished with a polishing cloth using a sequence of 5, 1, 0.3 and 0.05  $\mu\text{m}$  alumina particles (Buehler, IL). Thin films of Cu were deposited on Ge IRE (3.1 mm diameter, 38.6 mm long), Ge chips ( $2 \times 2$  mm) and glass rods (3.1 mm diameter, 38.6 mm long) in a PAC-1 vacuum evaporator (Pelco, Redding, CA). The IRE or glass rod was mounted on a Teflon holder in the vacuum chamber and rotated by the rotary feed-through at 120 rpm. Ge chips were fixed, using double-sided sticky tape, to a glass rod which was rotated at

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120 rpm during evaporation. Both ends of the IRE or glass rod were masked with Teflon caps to prevent coating of the end cones during Cu deposition. The source consisted of 99.99% pure copper (EM Laboratory) contained within a tungsten wire basket positioned 16 cm from the target. The targets were cleaned by glow discharge for 3 min prior to the deposition of the copper at a pressure of  $1 \times 10^{-7}$  Torr. The copper was vaporized behind a shutter assembly for 1 min before it was deposited on the target material at a rate of  $0.4 \text{ nm Cu s}^{-1}$ . To obtain an indication of temperature changes that occur within the vacuum chamber during deposition, a fine-gauge bare wire thermocouple (J-Type, Cole-Parmer, IL) was positioned in place of the substrate. Temperature measurements were recorded as Cu was deposited directly onto the thermocouple.

#### Deposition of copper thin films in a magnetron sputtering system

The surfaces of Ge discs (6 mm diameter, 1 mm thick) were polished as outlined in the previous section. Thin films (5.3 nm) of Cu were deposited on the Ge substrates in an EnerJet Model UCV-18/6 magnetron sputtering system. Substrates were mounted on Teflon fixtures in the vacuum chamber. Target materials consisted of 99.999% pure Cu (Cerac, Inc). A pump down pressure of  $5 \times 10^{-7}$  Torr was achieved before sputtering. Neon working gas (99.99% pure from MG Industries) was introduced into the sputtering chamber at a pressure of 45 mTorr. The deposition rate was  $0.28 \text{ nm s}^{-1}$ .

#### Calculation of the thickness of deposited Cu films

A quartz crystal oscillator was used to monitor the Cu film thickness. The monitor was calibrated by determining the amount of copper deposited on glass rods. The Cu on the glass rods was dissolved in 0.05 M  $\text{HNO}_3$  (5 ml) and the Cu concentration in the solutions was determined by atomic absorption spectroscopy.

The thickness of the Cu thin film on the surface of the glass rods was calculated using the equation

$$T = \frac{V}{A}$$

where  $T$  = thickness of the Cu thin film (cm),  $A$  = area (length  $\times 2\pi r$ ),  $V$  = volume of Cu [ $V = t/\rho$ , where  $t$  = amount of Cu (g) (determined by AAS) and  $\rho$  = density of Cu ( $8.92 \text{ g cc}^{-1}$ )].

A regression equation was used to calculate the Cu thin-film thickness on the Ge IRE for a given quartz crystal oscillator response.

#### Attenuated total reflection Fourier transform infrared spectroscopy (ATR/FT-IR)

A Ge IRE coated with Cu by PVD was inserted into a micro Circle cell (Spectra Tech, Stamford, CT) positioned on an optical bench inside a Perkin-Elmer Model 1800 FT-IR spectrometer equipped with a KBr beam splitter and a liquid-nitrogen-cooled medium-

range mercury-cadmium telluride detector ( $5000\text{--}580 \text{ cm}^{-1}$ ).

The optical bench housing the Circle cell was purged with air that had been passed through a Balston air dryer (Model 75-50) to remove water vapor and  $\text{CO}_2$ . The chamber was maintained at  $25 \pm 1^\circ\text{C}$  during each experiment.

Experiments were initiated by collecting a single beam spectrum of the Cu-coated IRE in air. Double-distilled water (ddw) was added to the Circle cell and another single beam spectrum was obtained. The ddw spectrum was ratioed against the spectrum of the Cu-coated IRE in air.

Each single beam spectrum was calculated from 50 averaged interferograms. Interferograms were double-sided and apodized with a weak Beer-Norton function prior to the fast Fourier transformation using the Model 1800 software. Scans were taken over the range of  $4000\text{--}700 \text{ cm}^{-1}$  with an interval of  $2 \text{ cm}^{-1}$  and a nominal resolution of  $4 \text{ cm}^{-1}$ . The interferometer OPD mirror velocity was set at  $3 \text{ cm s}^{-1}$  and the Jacquinot stop employed was 6.

#### Atomic force microscopy

Atomic force microscopy (AFM) images were taken of the surface of four polished Ge chips. One chip remained uncoated while three were coated by PVD with films of Cu; 3.35, 6.7 and 13.4 nm nominal thickness. The surfaces were imaged in air using a Nanoscope II AFM (Digital Instruments, Santa Barbara, CA). A  $100 \mu\text{m}$  silicon nitride cantilever (Digital Instruments with a spring constant ( $k$ ) of  $0.58 \text{ N m}^{-1}$  was used on all of the samples. The applied force while imaging was  $\sim 10^{-8} \text{ N}$ .

#### Preparation of Cu thin films for examination by transmission electron microscopy

Electron microscope (em) grids (100 mesh) coated with formvar (0.4% in 1,2-dichloroethane) were carbon coated (4.8 nm C) in a PAC-1 vacuum evaporator. The carbon-coated em grids were mounted on a glass rod using double-sided sticky tape and Cu-coated by PVD as outlined previously for Ge chips. The Cu-coated grids were examined using a Jeol (JEM-1200 EX II) transmission electron microscope, operated at 80 kV.

#### Ethylene oxide sterilization of Cu thin films

Duplicate germanium discs were coated with a thin film of Cu using a magnetron sputtering system. One Cu-coated Ge chip was immediately inserted into the XPS for analysis while the other coated Ge chip was sterilized by exposure to ethylene oxide, for 12 h, using Anprolene sterilization gas ampules (Andersen Products, Inc., Oyster Bay, NY). Immediately after sterilization, the Cu-coated Ge disc was inserted into the XPS for analysis.

## X-ray photoelectron spectroscopy (XPS)

Qualitative and quantitative analyses of the Cu-coated Ge substrates were carried out in a Perkin-Elmer Physical Electronics Model 5400 ESCA system equipped with a hemispherical analyzer. The energy scale was calibrated by analyzing Ar<sup>+</sup> sputter-cleaned Cu (2p<sub>3/2</sub> line at 932.6 ± 0.1 eV), Ag (3d<sub>5/2</sub> line at 368.3 ± 0.1 eV) and Au (4f<sub>7/2</sub> line at 84.0 ± 0.1 eV) foil standards. The deposited thin film peak positions were referenced to adventitious C (1s line at 284.6 eV) and corrected for sample charging. All spectra were acquired using Mg K $\alpha$  radiation, an energy resolution of 0.10 eV per step, a pass energy of 71.55 eV and a take-off angle of 45°. Spectra were not smoothed. Sample exposure to x-radiation was < 2 h.

To evaluate the possibility of x-ray or thermal (from x-ray source)-induced oxidation changes of the Cu, anhydrous CuSO<sub>4</sub> was exposed to Mg K $\alpha$  x-radiation from the XPS analyzer for 15 h.<sup>11,12</sup> No noticeable changes in the Cu 2p and Auger lines were noted.

## RESULTS AND DISCUSSION

Thin films of metallic Cu were evaporated on the surface of germanium internal reflection elements for the purpose of studying the stability of Cu in aqueous environments. The effect of the Cu film on the depth of penetration of the evanescent wave of infrared radiation into the surrounding aqueous medium was evaluated by monitoring the absorbance of water at 1640 cm<sup>-1</sup> (Fig. 1). Absorbance decreased with increasing film thickness over the range of 3–7.5 nm. The decrease in absorbance is due to both the high refractive index and absorptivity of Cu.<sup>6</sup> A film thickness of > 7.2 nm reflected too much energy to obtain an adequate signal-to-noise ratio. Thin films up to 7.2 nm nominal thickness transmitted sufficient energy to give a good signal-to-noise ratio. The data suggest that the intensity of the water absorption band at 1640 cm<sup>-1</sup> is very sensitive to changes in thickness or stability of the copper film.

The growth of a thin metal film on a substratum is influenced by a number of factors, including temperat-

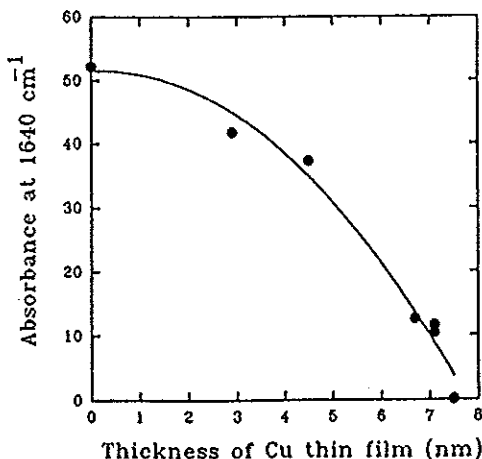


Figure 1. Absorbance of water at 1640 cm<sup>-1</sup> as a function of thickness of a Cu thin film on a Ge IRE.

ure.<sup>13,14</sup> The temperature of the substratum during Cu deposition by vacuum deposition was determined over a range of film thicknesses using a thermocouple as the substratum. From Fig. 2 it can be seen that during deposition of Cu films with nominal thicknesses ranging from 3 to 7 nm, the temperature measured at the thermocouple surface only increased from 22.7°C (ambient) to 32°C. Deposition at near-ambient temperatures has been shown to produce Cu thin films with a smaller crystal size and of a more homogeneous nature than films deposited at much higher temperatures.<sup>14,16</sup>

The growth of the Cu thin film on a flat Ge chip was evaluated by atomic force microscopy (Fig. 3). The surface of the bare, polished, Ge substratum was determined to be relatively smooth, with variations of only a few nanometers (Fig. 3(A)). A 3.7 nm thick Cu film (thickness based on a mass-calibrated quartz crystal) appeared as discrete aggregates of Cu atoms distributed as islands over the Ge substrate (Fig. 3(B)). A Cu film of 6.7 nm nominal thickness appeared as aggregates of Cu atoms which had converged upon each other (Fig. 3(c)). Films with a nominal thickness of 13.5 nm were composed of aggregates of Cu atoms which coalesced to form a smoother surface than those obtained with thinner film thicknesses (Fig. 3(D)).

The morphology of a 6.7 nm thick copper film deposited by vacuum evaporation was also evaluated by transmission electron microscopy in order to permit a comparison with thin films produced in previous studies.<sup>6,16</sup> Films of this nominal thickness, when deposited on carbon-coated, formvar-coated, copper electron microscope grids (Fig. 4), appeared to be uniform and similar in appearance to images presented by Iwaoka *et al.*<sup>6</sup> and Ishida and Griffiths.<sup>16</sup> Clusters of copper adatoms, which averaged 50 nm in diameter, were separated by 5–10 nm gaps. It was not possible, however, to determine by these microscopic techniques whether these gaps were true discontinuities or simply low spots in the film.

The surface of copper thin films deposited simultaneously on Ge discs in a magnetron sputtering system

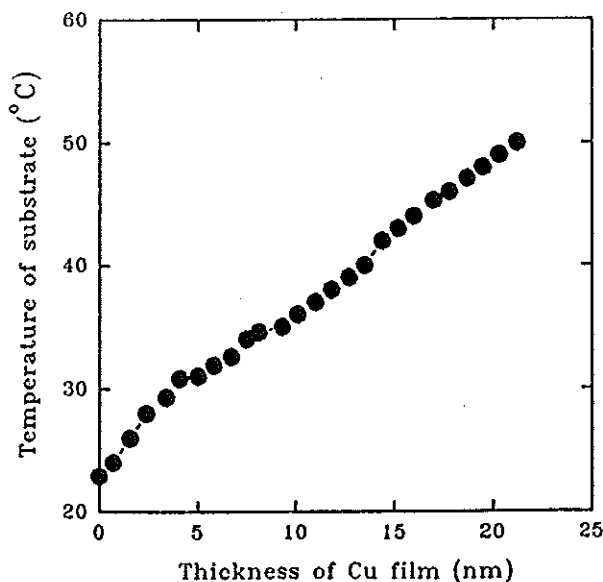


Figure 2. Thermocouple surface temperature as a function of the thickness of a Cu thin film deposited on the thermocouple.

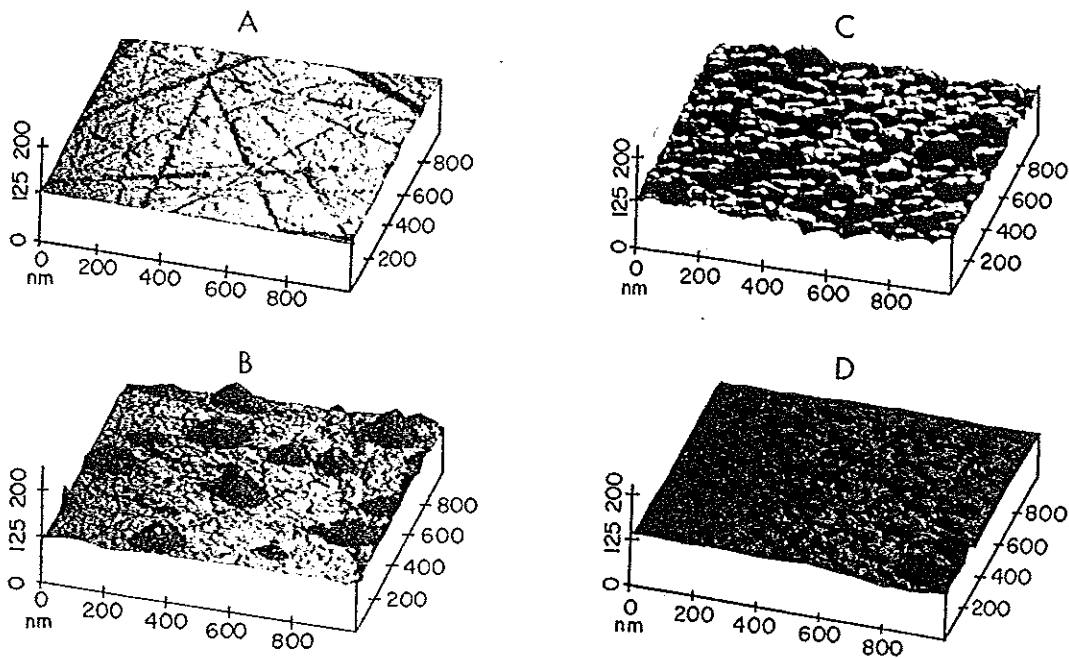


Figure 3. AFM images of polished Ge chips coated with vacuum-deposited Cu thin films of various thicknesses: (A) uncoated; (B) 3.4 nm Cu; (C) 6.7 nm Cu; (D) 13.5 nm Cu.

were characterized by XPS. Unsterilized copper films and copper films sterilized by exposure to ethylene oxide for 12 h exhibited Cu  $2p_{1/2}$  binding energy lines at 952.3 and 952.4 eV, respectively, while sputter-cleaned Cu foil yielded a peak at 952.3 (Fig. 5, Table 1). The unsterilized and the ethylene oxide-sterilized Cu films also displayed Cu  $2p_{3/2}$  binding energy lines at 932.4

and 932.5 eV, while the copper foil yielded a line at 932.4 eV (Fig. 5, Table 1). The Cu Auger kinetic energy lines occurred at 916.4 and 916.3 eV for the unsterilized and ethylene oxide-sterilized Cu films, respectively, reflecting an Auger shift to lower energy compared to the 918.8 eV line produced by the sputter-cleaned foil (Fig. 5, Table 1). Based on comparisons with the positions of the Cu  $2p$  and Cu LVV lines of copper oxide reference compounds, the copper on the surface of the unsterilized Cu film appeared to be in a  $1+$  valence state, possibly as cuprous oxide. Ethylene oxide treatment caused no significant changes in the character of the copper on the surface of the film. All spectral intensities were referenced to the sputter-cleaned Cu foil for general comparison only.

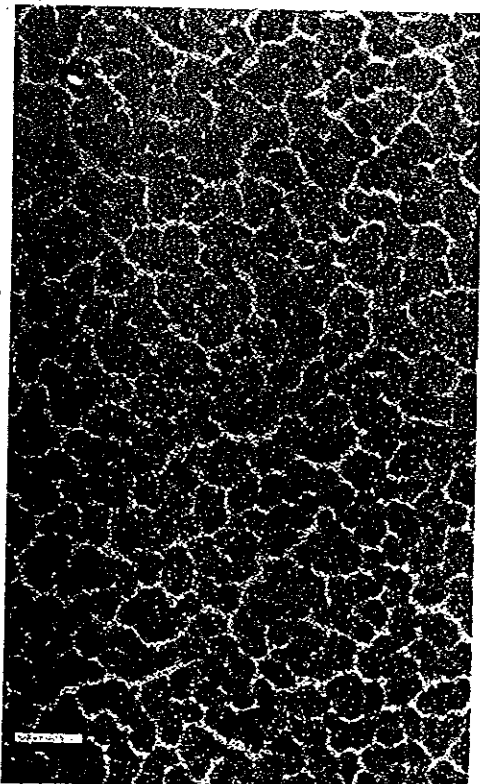


Figure 4. Transmission electron micrograph of a carbon-coated em grid, coated with a vacuum-deposited Cu thin film (6.7 nm). Bar = 50 nm.

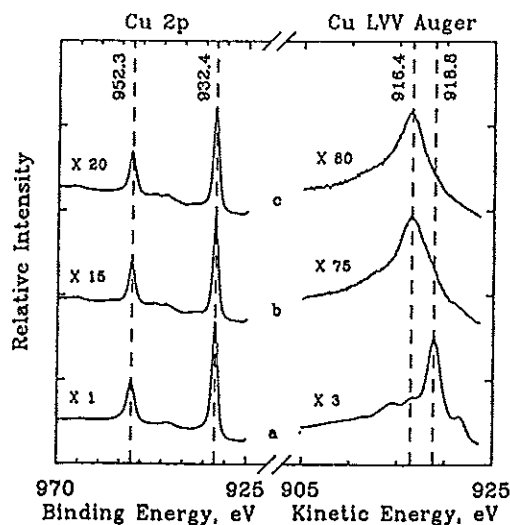


Figure 5. Cu  $2p$  and Cu  $L_{2,3}M_{4,5}M_{4,5}$  spectra from (a) sputter-cleaned Cu foil, (b) sputter-deposited Cu prior to ethylene oxide treatment and (c) after treatment. Scaling factors are included (e.g.  $\times 1$ ).

Table 1. Energy values observed for the Cu 2p<sub>1/2</sub>, Cu Auger LVV and Ge 3d lines for spectra of Figs 5-7

Sample	Cu 2p <sub>1/2</sub> <sup>a</sup>	Cu 2p <sub>3/2</sub> <sup>a</sup>	Cu LVV <sup>b</sup> Auger	Ge 3d <sup>a</sup>
Cu foil	952.3	932.4	918.8	NA <sup>c</sup>
Sputtered Cu				
Pre-treat	952.3	932.4	916.4	NA <sup>c</sup>
Post-treat	952.4	932.5	916.3	NA <sup>c</sup>
PVD Cu with treatment				
0 s	952.4	932.5	916.9	ND <sup>d</sup>
30 s	952.7	932.7	916.9	ND <sup>d</sup>
75 s	952.6	932.6	917.0	31.3
105 s	952.4	932.5	918.7	31.3
135 s	952.7	932.9	918.4	29.5

<sup>a</sup> Binding energy (eV).

<sup>b</sup> Kinetic energy (eV). LVV refers to L<sub>3</sub>M<sub>4,5</sub>M<sub>4,5</sub>.

<sup>c</sup> NA = not applicable.

<sup>d</sup> ND = not detectable.

Ethylene oxide-sterilized copper films (6.7 nm nominal thickness), deposited by PVD on Ge discs using the same approach as that used to deposit copper films on Ge IRE and Ge chips examined by atomic force microscopy, were evaluated by XPS sputter depth profiling. Like the films deposited by the magnetron sputtering system, the Cu 2p and Cu LVV Auger lines indicated that the Cu on the surface of the vacuum-evaporated film had a 1+ oxidation state character (Fig. 6(a), Table 1). The Cu LVV Auger kinetic energy peak shifted from 916.9 to 918.4 eV after sputtering with

Ar<sup>+</sup> for 75 s or longer (Fig. 6(c)-(e), Table 1). No significant change in the Cu 2p binding energy lines was observed during this time. These results suggest that beneath the layer of Cu<sup>1+</sup> at the surface, the film exhibited a ground-state Cu<sup>0</sup> character. The overall intensity of the Cu lines decreased during sputtering.

The Ge 3d binding energy line was not detectable until after the ethylene oxide-treated PVD Cu film had been sputtered with Ar<sup>+</sup> for 75 s (Fig. 7). The appearance of a broad line centered at 31.1 eV after sputtering for this period of time suggests that the Ge had an oxidized character (Fig. 7(c), Table 1). After sputtering for 105 s, a line at 29.5 eV, characteristic of ground-state Ge<sup>0</sup>, appeared; this energy line predominated after 135 s of sputtering (Fig. 7(d), (e), Table 1).

The XPS sputter depth profile (Fig. 8) indicated that the maximum concentrations (at.%) of Cu, C and Ge were 62%, 93% and 31%, respectively. It appears as if the surface of the ethylene oxide-treated PVD Cu film was contaminated with carbon, either in the deposition process or by residual ethylene oxide, its degradation products or other carbonaceous material adsorbed from the atmosphere (Fig. 8). The copper intensity increased as the surface layer of carbon was eroded, achieving a 62% atomic concentration within 0.5 nm of the surface (Fig. 8). The percentage atomic Cu concentration then decreased over the 2 nm depth interval underlying the 0.5 nm thick surface layer. After 30 s of sputtering, the carbon concentration appears to follow the Cu concentration; this indicated that the C may have been incorporated in the Cu thin film.

The detection of Ge close to the surface (Fig. 8) suggests that only a very thin film of copper coated the Ge substrate in some areas. Since Cu<sup>0</sup> was not detectable during the first 30 s of sputtering, it appears that all the copper present in the thinly coated areas was oxidized

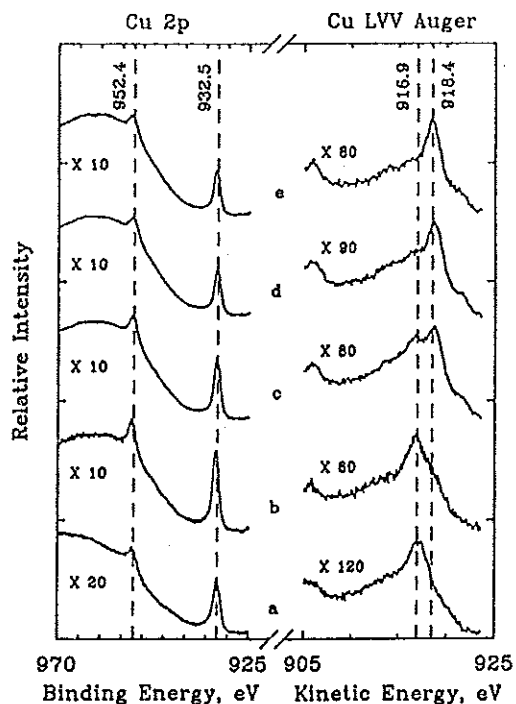


Figure 6. Cu 2p and Cu L<sub>3</sub>M<sub>4,5</sub>M<sub>4,5</sub> spectra from (a) an ethylene oxide-treated PVD Cu thin film (6.7 nm), (b) 30 s of Ar<sup>+</sup> sputtering, (c) 75 s, (d) 105 s and (e) 135 s. Scaling factors are included (e.g. × 10).

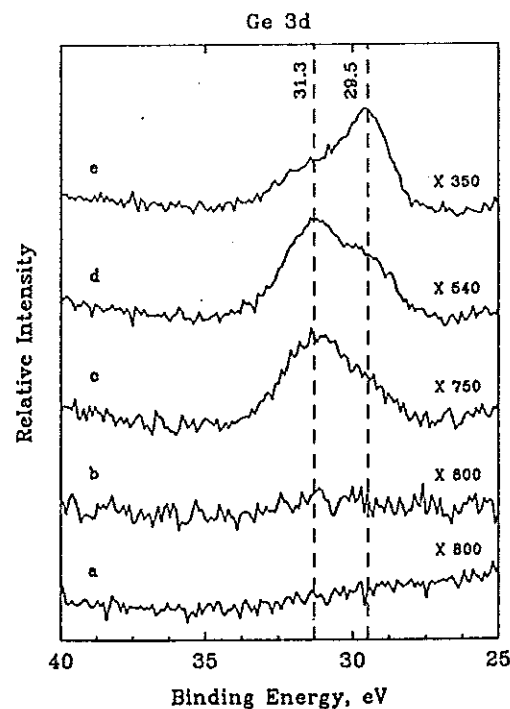


Figure 7. Ge 3d spectra from (a) an ethylene oxide-treated PVD Cu thin film (6.7 nm), (b) 30 s of Ar<sup>+</sup> sputtering, (c) 75 s, (d) 105 s and (e) 135 s. Scaling factors are included (e.g. × 1).

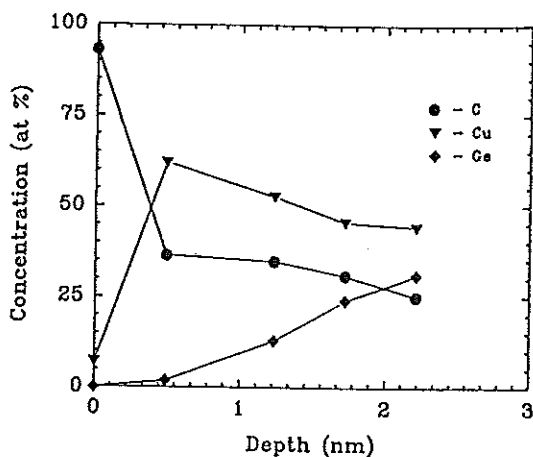


Figure 8. XPS sputter depth profile of an ethylene oxide-treated PVD Cu thin film (6.7 nm). Atomic per cent concentrations are plotted as a function of depth.

copper (possibly as  $\text{Cu}_2\text{O}$ ). The copper concentration decreased as the Ge concentration increased; this is what we would expect if the thin film over the surface of the Ge was continuous. However, to verify that the thinly coated areas, which likely correspond to the gaps seen in images obtained by transmission electron microscopy and the low areas seen by atomic force microscopy, contain a continuous layer of oxidized Cu, additional data over narrower sputter depth intervals using a micro-probe that could look into the 5–10 nm

valleys in a segregated mode are required. Unfortunately, such micro-probes have yet to be developed.

## CONCLUSIONS

The results indicate that Cu thin films of 6.7 nm nominal thickness deposited on Ge substrates by vacuum evaporation exist as converging aggregates of copper adatoms. The thickness of the Cu film varies over the Ge surface. Both unsterilized and ethylene oxide-sterilized Cu thin films contain an oxidized surface layer of  $\text{Cu}^{1+}$ , possibly as cuprous oxide. The surface of ethylene oxide-treated Cu films is heavily contaminated with carbon. The film is sufficiently thin to permit penetration of IR radiation into the surrounding environment, which in the case of water provides a sensitive means of evaluating film integrity. Copper thin films deposited in this manner have been shown to be sensitive to certain microorganisms and their products and provide a novel way to study microbial-influenced corrosion.<sup>6,9</sup>

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