Introduction:
It is important for veterinary hospitals to maintain high standards of environmental cleanliness to prevent the transmission and spread of infectious diseases to both animals and humans. The purpose of this study was to examine the cleanliness of the VTH environment in an objective manner through the use of quantitative bacterial cultures. Two bacterial culture methods (contact plating and Spiral plating) were compared. The sampling methods used in this study have a potential to be a useful tool in longitudinal monitoring of the cleanliness of the hospital environment and for directed investigations of various hygiene protocols. Documenting baseline bacterial loads for multiple areas and surfaces throughout the hospital will provide valuable data for comparison in future investigations.

Materials and Methods:
Sampling and culture methods: Two hundred environmental sites throughout the James L. Voss Veterinary Teaching Hospital were sampled and cultured. For spiral plating, cotton-tipped swabs moistened with a commercial broth containing neutralizers for common disinfectants (Difco) were used to sample 10cm × 10cm areas at each site. Swabs were then placed in neutralizing broth and held at room temperature until processed at the laboratory. Samples were also obtained using contact plates (TSA and MacConkey agar) at sites adjacent to those sampled for spiral plating. Contact plates were gently pressed against the surface for 5 seconds, covered, and held at room temperature until processed at the laboratory. Samples were also obtained using contact plates (TSA and MacConkey agar) at sites adjacent to those sampled for spiral plating. Contact plates were gently pressed against the surface for 5 seconds, covered, and held at room temperature until processed at the laboratory. All environmental samples were processed at the laboratory within 2 hrs of sampling. Samples collected for spiral plating were vortexed for 5 sec and then plated to tryptic soy agar (TSA) and McConkey agar using a spiral plater (Spiral Systems Inc). All plates (spiral plates and contact plates) were incubated at 35°C for 48 hours, and colony forming units were quantified according to the manufacturer's instructions at 24 and 48 hrs.

Statistics:
All data were expressed as CFU/100cm2. Data were distinctly non-normal and were therefore transformed to facilitate statistical analyses. When estimation of colony counts on contact plates was not possible, samples were arbitrarily assigned a higher colony count (999 CFU/100cm2 on TSA agar, and 99 CFU/100cm2 on MacConkey agar). Log10 transformation of data was used for colony counts estimated using TSA agar, while colony counts estimated from MacConkey agar were transformed by ranking because of the irregular distributions. Linear regression (Proc GENMOD, SAS v8.2) was used to evaluate data, controlling for repeated nature of data using generalized estimating equations. Associations were evaluated between the estimated bacterial counts
and the following sampling variables: area of the hospital, surface composition, surface use, and the cleanliness of the surface when sampled. Simple associations were evaluated for each outcome variable, and those found to be significantly associated with outcome variables (P<0.20) were included in multivariable modeling. Backward selection was used to determine final models (P<0.10). Least-square mean bacterial counts were then determined for variables retained in the final models.

Results:
Evaluation of crude data showed that the heaviest bacterial contamination was detected in equine areas, while small animal surgery area was the cleanest. Among the surfaces tested, linoleum and concrete had the highest bacterial contamination, while stainless steel and plastic or rubber had the lowest contamination. However, after controlling for hospital area and surface characteristics, formica, stainless steel and vinyl were the surfaces associated with the highest bacterial counts, while concrete and plastic or rubber had the lowest. Floor surfaces had higher bacterial burden than hand-contact surfaces. Surfaces designated as "dirty" at the time of sampling showed higher bacterial contamination than "clean" surfaces. However, the highest bacterial burden was detected on surfaces classified as "not determined".

Discussion:
The colony counts obtained from spiral plates were on average 1 log10 greater than those estimated from contact plates. This could be due to the use of disinfectant neutralizers when collecting samples for spiral plating, which were not incorporated in contact plates. Also, the swabbing technique used with the spiral plating method resulted in more "aggressive" sampling compared to simply touching surfaces with contact plates. Gram negative bacteria accounted for only approximately 10% of the total colony counts. This may reflect a low level of contamination with enteric organisms throughout the hospital. Overall, floor sites had the highest Gram negative bacterial load, which likely reflects the carriage of fecal contamination on feet and wheeled equipment.
Samples of surfaces designated as "clean" or "dirty" were specifically obtained in areas where there is great concern about environmental hygiene, including the small animal critical care unit (CCU), equine surgery, and small animal surgery. Both "clean" and "dirty" samples obtained from these areas had less bacterial contamination when compared with other sites in the hospital which were sampled only once (cleanliness status was recorded as "not determined" in these locations). This likely reflects more strict cleaning and hygiene protocols that are used in the CCU and surgery suites in comparison with other hospital areas.