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Endotoxins in the Manufacturing Environments of Natural Rubber (NR) Latex Gloves

AZIANA, A.H.*#, IKRAM, A.*, MOK, K.L.* AND AMIR-HASHIM, M.Y.*

Measurements of endotoxin and microbial contamination during glove manufacture were made at two commercial plants and from the RRIES Gloveline. Samples were taken from solutions and air along the processing tank line, and from the stripping and packaging areas. At all three plants, high airborne endotoxin concentrations were encountered in the stripping areas and in the packing rooms, relative to the concentration range along the processing tank line. Endotoxin was also detected from surfaces at the stripping areas and in the packing rooms. The most likely cause for the final endotoxin concentrations on the formed gloves is the higher level of airborne endotoxins during stripping and packing activities. The population densities of airborne microbes were also high in the stripping area. A variety of bacteria were recorded, with a predominance of Gram-negative types. Reducing the likelihood of bacterial contamination reduces both microorganisms and pyrogens and it is vital to place greater focus on reducing microbial contaminants at these critical areas of surgical glove production.

Keywords: NR latex gloves; endotoxin; surgical gloves; examination gloves; air sampling; manufacturing environment; LAL

Endotoxin is a biological toxin that is part of the outer membrane of Gram-negative bacteria. It is known to cause powerful inflammatory responses in human and animal subjects when they find their way into the mammalian blood system, especially at high doses[1–7]. The symptoms ranged from fever and shivering to hypotension, adult respiratory distress syndrome, airway inflammation, disseminated intravascular coagulation and fatal septic shocks. During Gram-negative sepsis, endotoxin stimulates "activates" host alveolar macrophages and respiratory epithelial tissue to release proinflammatory cytokines (IL-1, IL-6, IL-10) and the tumour necrosis factor α (TNF-α), some of which travel through blood to the hypothalamus, the body's thermoregulatory centre in the brain to trigger the inflammatory response[8].

Endotoxins may adhere on implants/ medical devices even after sterilisation and can be a significant contaminant on NR latex gloves[9–14], that posed risks to patients and healthcare workers. Commercially available medical devices need to have low endotoxin levels before they can be approved for sale by the regulatory authorities. This creates the concern for endotoxin contamination control. Indeed, biocompatibility studies of biomaterials may be compromised by this hard to avoid endotoxin contamination[15].

*Rubber Research Institute of Malaysia, Malaysian Rubber Board, P.O. Box 10150, 50908 Kuala Lumpur, Malaysia
Corresponding author (e-mail: aziana@lgm.gov.my)

Endotoxins are ubiquitous in both outdoor and indoor environments, and high occupational endotoxin exposure is already known in agricultural and related industries (crop harvesting, cotton and vegetable processing, livestock barns, grain handling, slaughter houses), textiles, pulp and paper processing, sawmills, composting, sewage and domestic waste handling, wastewater treatment, fibreglass manufacturing and metal machining environments[16]. In the rubber industry, there have been no studies for endotoxin exposure assessment, to identify specific locations and tasks associated with high exposure to endotoxins or of endotoxin contamination related to product manufacture, as is common in the food, beverage and pharmaceutical industries. The present study was undertaken to monitor endotoxin concentrations during the manufacturing stages of natural rubber (NR) latex gloves, and to relate the levels to the finished product.

MATERIALS AND METHODS

Study Site

The study was conducted in two commercial plants manufacturing powdered sterile surgical gloves, and at the Rubber Research Institute Experiment Station (RRIES) Gloveline during the occasional manufacture of non-sterile NR latex examination gloves.

Liquid Material Sampling

In glove processing plants, the cleaned coagulant-dipped porcelain formers attached to a continuous chain were dipped into NR latex, leaching tanks and slurry before stripping. Liquid samples (approx. 100 mL) were taken from the latex, preleach, postleach and slurry tanks in duplicates in depyrogenated sampling bottles for the Limilus Amoebocyte Lysate (LAL) assay. The same samples (10 mL aliquots) were also serially diluted by 10 folds and plated on Trytic Soy Agar (TSA; Difco Laboratories, Detroit, MI, USA) containing 50 mg L^{-1} cycloheximide (Sigma Chemical Co., St.Louis, MO, USA) and Potato Dextrose Agar (PDA; BBL, Cockeysville, MD, USA) containing 33 mg L^{-1} rose Bengal and 30 mg L^{-1} streptomycin (Sigma Chemical Co., St.Louis, MO, USA) for total viable counts of aerobic bacteria and total fungi, respectively. The plates were incubated at 30ºC and the population densities were counted after 5 days.

Serum Extraction of Latex

A salt coagulation method was developed to separate the rubber particles from the serum. Up to 2 g $MgSO_4.7H_20$ was slowly added into 9.9 mL Tris-HCl buffer in small tubes on a hotplate and 0.1 mL latex was added into the mixture then subsequently mixed on a Whirlmixer. The reasonably clear solution that separate from the coagulum was then centrifuged at 4,000 r.p.m. for 30 min and the extract subsampled for pH checks and adjusted to 7.0 – 8.0 using endotoxin free 0.1M NaOH or 0.1M HCl when necessary.

Air Sampling

Airborne workplace endotoxins were collected by a portable sampling system with a suction pump (KNF Neuberger VDE 0530) operating at a flow rate of 2.0 Litres/min for 5 min at each sampling location to yield a sample volume of 10 L. The filter holder was fitted with a 47 mm glass microfibre filter (Whatman GF/D; Whatman International Ltd, Maidstone, England) and 2 – 4 unexposed filters were used as experimental blanks. The exposed filters were extracted in 10 mL pyrogen-free water

as extraction solutes in depyrogenated test tubes on a platform shaker (220 r.p.m.) at 30ºC for 1 h. Where necessary, the tubes were centrifuged for 45 min at 3500 r.p.m. Endotoxin assays were performed on the filter eluates. The amount of endotoxin in the sampling fluids was converted to EU/m^3 using the airflow rate of the samples and the sampling time.

The concentrations of airborne bacteria and fungi were determined using a Microbial Air Sampler (MERCK, Germany) operated for 5 mins, using settling plates of TSA and PDA. Samples were collected in the air above the latex, preleach, postleach and slurry tanks, from the strip area and the packing rooms. All samplings were performed at 1 m height with the samplers set on a platform. Air temperatures ranged from 32ºC – 45ºC (RH, 38% – 67%) along the processing tanks.

Surface Sampling

Sterile cotton swabs moistened in 10 mL sterile Ringers solution were used to collect surface samples within the work facility. An area of 10 cm^2 was identified, and the tips of the moistened swabs were swept three times across the surfaces. The swabs were returned to the solution tubes, labelled, kept in ice and transferred to the laboratory to be used for surface plate counts and endotoxin assays. Swabs were taken from defined surfaces such as table tops, floors and walls in the packing rooms, and from the discs, formers and baskets in the stripping area.

Gloves

Gloves were assayed as previously described[17] using the procedure outlined in *BS EN 455-3: 2000*[18].

Endotoxin Assays

All glassware was depyrogenated at 180ºC for 4 h prior to use, and pyrogen-free LAL Reagent Water (LRW, <0.001 EU/mL; Associates of Cape Cod, Inc., Falmouth MA) was used as a negative control, for reconstituting the lyophilised reagents and the control standard endotoxin (CSE).

Endotoxin concentration was determined by a kinetic turbidimetric LAL assay as previously described[17]. The generated standard curves used a reference control standard endotoxin (CSE; *Eschericia coli* O113:H10). Aliquots of known amounts of endotoxin standards (positive controls), dilutions of sample extracts in 2-fold serial dilutions and LRW blanks (negative controls) in wells of pyrogen-free microtitre plates (Nunc A/S, Denmark) were reacted with the LAL reagent (Pyrotell-T®; Associates of Cape Cod, Inc, Falmouth, MA). Spiked dilutions were included to test for performance of inhibition or enhancement. The assay plate was placed in a ELx808i microplate reader (Bio-Tek Instruments, Inc., Vermont, USA) and allowed to proceed at 37 ± 1ºC for 1h. Spectrophotometric measurements at 340 nm were taken at every 20 sec interval, and data analysed using KC4™ Software (Biotek Instruments, Inc.) and the sample concentrations computed at a minimum acceptable standard curve values of r^2 at 0.998. The kinetic software calculates the "onset time" for the sample in each well to reach a specified optical density value "onset OD", generates the standard curve parameters (slope, intercept, correlation coefficient) and calculates the endotoxin concentrations in the unknown samples. All endotoxin concentrations were expressed in endotoxin units per mL solutions (EU/mL), per m^3 of air (EU/m^3), per pair of gloves (EU/glove pair) or per 10 cm^2 of swab area.

Bacterial Identification

Bacterial isolates were identified by the Biolog™ Microbial ID System[19] that uses carbon source metabolic patterns "fingerprints". The different isolates were plated on growth media and distinguished by their colonial morphology, margin characteristics, pigmentation and Gram reaction. Tests were done on samples taken from the preleach, postleach and slurry tanks, gloves as well as from the inlet water source. A 24 h pure culture of the isolate to be identified was prepared and emulsified in inoculation fluid. Cell density of the cell suspension was determined by adjusting the volume of inoculation fluid. The cell density was an indicator for the oxygen concentration which is a key control parameter for testing microorganisms in the MicroPlate™ test format. The cell suspensions were added into each well of the plates. The plates were incubated (35ºC – 37ºC) and the individual reaction was interpreted after 18 – 24 h (Gram negative bacteria) or 48 h for the Gram positive isolates. Results were read using the semi-automated MicroStation Reader™ and the purple well patterns were entered into the MicroLog Software™, which searches the database and provides the identification. Further incubation period was carried out if the MicroLog Software™ could not identify the isolates.

RESULTS AND DISCUSSION

Endotoxins in Solutions

Endotoxin concentrations in the tank solutions at the sequential processing stages involved (latex, pre-leach, post-leach, slurry) and from the inlet water ranged from 9 – 1,140 EU/mL at Factory A, with the highest values in the leaching tanks (*Table 1*). The higher concentrations in the leaching tanks were expected, since they involved washings of water-soluble proteins, excess additives and contaminant microbes from the dipped gloves, leading to accumulation of microbes. Surprisingly, the endotoxin level in the slurry solution was low. Slurry cornstarch solutions had biocides added to minimise microbial growth. At the RRIES Gloveline, high values for endotoxins were also obtained from solutions in the pre-leach (1235 EU/mL) and post-leach (622 EU/mL) tanks, while the slurry and latex tanks had lower values (17 EU/mL, respectively). In contrast, the concentrations of endotoxin in solution tanks at Factory B were far higher, ranging from 168 EU/mL in the latex tank to 12,446 EU/mL in the post-leach tank. The endotoxin concentration in the slurry tank (573 EU/mL) was also many folds higher than that encountered with surgical glove production at Factory A, or of examination gloves at the RRIES gloveline. The concentrations of inlet water were not measured at Factory B or at the RRIES Gloveline, but was only 34 EU/mL at Factory A.

While the trend in endotoxin levels from LAL assay quantification were quite similar between Factory A and the RRIES Gloveline, the results from Factory B indicate the potential for high endotoxin concentrations in processing tanks arising from extensive contamination by Gram-negative bacteria. Such high concentrations from microbial contamination were also encountered in raw waters, or from industrial and agricultural settings. For *e.g.* Jorgensen *et al*.[20] obtained endotoxin concentrations that varied from about 7.8 – 12,500 EU/mL (assuming 1 ng is equivalent to 10 EU/mL). Anderson *et al*.[21] reviewed water-associated endotoxin literature and found endotoxin concentrations in raw (untreated) waters ranging from about <10 – 10,490 EU/mL. However, the endotoxin contents of drinking water in distribution systems from treatment plants

TABLE 1. RELATIVE AMOUNTS OF ENDOTOXIN CONCENTRATION IN SPECIFIC PRODUCTION AREAS OF GLOVE MANUFACTURE BY LAL ASSAY

Location	Endotoxin concentration		
	Solution (EU/mL)	Air (EU/m^3)	Surfaces (EU/10 cm^2)
Factory A (surgical gloves)			
Latex	15	56	–
Inlet water	34	–	–
Pre-leach	1,140	48	–
Post-leach 1	250	52	–
Post-leach 2	462	52	–
Slurry	9	62	–
Outdoors	–	14	–
Strip area			
(i) disc			179
(ii) inside former		226	182
(iii) outside former			108
(iv) basket			145
Packing room (powdered)			
(i) table			138
(ii) wall		368	118
(iii) floor			n.d.
Packing room (powder-free)			
(i) table			219
(ii) wall		50	168
(iii) floor			165
Gloves (before sterilisation)	17/pair (n=10 pairs)		
Gloves (irradiated)	35/pair (n=10 pairs)		
Factory B (surgical gloves)			
Latex	168	427	–
Inlet water	n.d.	n.d.	–
Pre-leach	12,146	9,856	–
Post-leach	12,446	10,225	–
Slurry	573	1,593	–
Outdoors	–	n.d.	–
Strip area	–	1,237	
Packing room (powdered)		1,589	
Gloves (before sterilisation)	36/pair (n=10 pairs)		
Gloves (irradiated)	102/pair (n=10 pairs)		

TABLE 1 (CONT.). RELATIVE AMOUNTS OF ENDOTOXIN CONCENTRATION IN SPECIFIC PRODUCTION AREAS OF GLOVE MANUFACTURE BY LAL ASSAY

Location	Endotoxin concentration		
	Solution (EU/mL)	Air (EU/m^3)	Surfaces (EU/10 cm^2)
RRIES Gloveline (examination gloves)			
Latex	17	62	–
Inlet water	n.d.	–	–
Pre-leach	1,235	437	–
Post-leach 1	622	234	–
Slurry	17	76	–
Outdoors	–	253	–
Strip area			
(i) disc			47
(ii) inside former		140	–
(iii) outside former			102
(iv) basket			70
Packing room (powdered)			
(i) table			n.d.
(ii) wall		n.d.	n.d.
(iii) floor			n.d.
Gloves (non-sterile)	28 EU/pair (n=4 pairs)		

n.d.: not determined

surveyed were relatively low, ranging from 2-32 EU/mL[21–23].

Endotoxins in Air

At Factory A, the endotoxin concentrations in the atmospheres above the processing tanks ranged from 48 – 62 EU/m^3, with relatively high values in the stripping area (226 EU/m^3) and in the powdered packing room (368 EU/m^3), although the level in the powder-free clean room was only 50 EU/m^3 (*Table 1*). As a comparison, the level of endotoxin obtained from the outside air was only 14 EU/m^3. It was observed that such high levels could be related to the higher amounts of dust and powder floating about in the air at these sites. Such particles are usually associated with carrying spores of airborne bacteria and fungi. Endotoxins in air also increase in situations where there is significant water damage or dampness. At the RRIES Gloveline, the airborne endotoxin concentrations followed the solution endotoxin trend in the sequential stages involved, being high around the pre-leach (437 EU/m^3) and post-leach (234 EU/EU/m^3) tank areas, followed by the slurry (76 EU/m^3) and latex tank (62 EU/m^3) areas. The atmosphere around the stripping area was 140 EU/m^3. As was the case with solution endotoxins, the levels of airborne endotoxins in Factory B were the highest, measuring around 10,000 EU/m^3 above the leaching tanks followed by a decrease in the slurry tank (1593 EU/m^3) and in the stripping (1237 EU/m^3) and packing areas (1589 EU/m^3).

Representative figures for airborne endotoxin concentrations in association with dust were known to be high in sewage

treatment plants (up to 500 EU/m^3)[24], fibreglass wool manufacturing (4 – 7590 EU/m^3)[25], paper pulping (6 – 19,400 EU/m^3)[26], swine enclosures (438 – 41,307 EU/m^3)[27], sewages and buildings with "sick building syndrome" (2,540 EU/m^3)[28]. In studies on water-related endotoxin exposure, Rylander and Haglind[29] measured airborne endotoxins levels at 1300 – 3900 EU/m^3 in a printing factory caused by a Pseudomonas-contaminated humidifier, and Joung *et al*.[30] obtained a range 6 – 1247 EU/m^3 at wastewater treatment plants. The concentration range of endotoxins in sawmills were 207 – 17,063 EU/m^3 and 2,026 – 11,297 EU/m^3 in swinebarns[31]. Spaan *et al*.[32] listed an overall range of airborne endotoxins in agricultural industries that varied from 2 – 149,060 EU/m^3 in grain production and processing, 2 – 191,430 in horticulture and 2 – 8,120 in the animal production sector. All of these workplace levels were much higher than that reported for domestic endotoxins or of outdoor air. For instance, inhalable endotoxins in Southern California ranged from 0.03 – 5.44 EU/m^3 [33], from <3 – 27.8 EU/m^3 in a German residential neighbourhood[34] or a mean of 1.5 EU/m^3 in aircraft cabins[35]. Our figures for the recovery and measurement of airborne endotoxins could not be broadly compared against data for other settings due to differences in efficiencies of the air sampling device, *e.g*. sampling approaches, use of impingers, choice of filter media, extraction protocols and assay buffers.

There is a well-established link between occupational exposure to airborne endotoxins and airway respiratory diseases[36]. Thus, measurement of exposure to endotoxins in the workplace is a powerful tool to assess potential risk in health. A study carried out by Schwartz *et al*.[36] demonstrated that endotoxin is a significant risk factor for both occupational and non-occupational asthma, especially among those with organic dust exposure. A few researches on endotoxin in relation with asthma, concluded that the endotoxins derived were due to the high level of dust exposure in the working environment[37]. Besides asthma, endotoxins could also cause symptoms on the upper respiratory tract, gastrointestinal tract as well as eye and skin irritations.

Endotoxins on Surfaces

At Factory A, the endotoxin concentrations on the surfaces in the stripping area and powdered packing rooms ranged from 108 – 182 EU/10 cm^2, and the higher values for table surfaces in the powder-free cleanroom (219 EU/10 cm^2) was expected since disinfecting the tables lysed bacterial cells that released endotoxins (*Table 1*). The surfaces in the strip area and packing room at Factory B or of the packing room at the RRIES gloveline were however not determined.

Endotoxins on Gloves

The surgical glove samples taken from the tumbler at Factory A showed a mean concentration of 17 EU/glove pair (mean of 10 pairs), while their irradiated counterpart from the same lot had a more than two-fold higher mean concentration of 35.4 EU/pair glove (*Table 1*). These would not pass the US Food and Drug Administration (FDA) test specified for medical devices. Medical devices have to be made sufficiently clean to meet the 20 EU/device endotoxin levels as specified by USP23 <161> (Supplement 1)[38]. Similarly, glove samples from Factory B produced concentrations of 36 and 102 EU/pair before and after irradiation, respectively. Samples of the non-sterile examination gloves produced at the RRIES Gloveline had concentrations of 28 EU/glove pair.

In our previous study of glove samples from commercially available brands, the endotoxin

concentrations ranged from 3 – 114 for sterilised surgical gloves from 10 brands, and from <8 – 9,632 EU/glove pair from 8 brands[17]. Thorne *et al.* (2005)[14] analysed 9 types of examination gloves and found the endotoxin content ranging from below detection <1.5 EU to 5810 EU. Asplund Peiro *et al.*[11] did test limited samples of sterile surgical gloves and categorised endotoxin contamination as minor (0.4 – 18 EU/pair glove), moderate (30 – 62/pair glove) or heavy (276 – 2142/pair glove).

Culturable Airborne Aerobic Bacteria and Fungi

The populations of total airborne aerobic bacteria were highest in the stripping area, followed by the latex tank, and in the powdered

TABLE 2. CONCENTRATIONS OF AIRBORNE BACTERIA AND FUNGI IN SPECIFIC PRODUCTION AREAS OF GLOVE MANUFACTURE

Location	Microbial population (cfu/m^3)	
	Bacteria	Fungi
Factory A (surgical gloves)		
Latex tank	782	586
Pre-leach	107	202
Post-leach	168	396
Slurry	231	97
Strip area	2,096	173
Packing room (powdered)	589	196
Packing room (powder-free)	308	18
Outdoor	217	120
Factory B (surgical gloves)		
Latex tank	n.d.	n.d.
Pre-leach	3,489	n.d.
Post-leach	3,641	n.d.
Slurry	968	n.d.
Strip area	3,525	n.d.
Packing room (powdered)	589	n.d.
Packing room (powder-free)	n.d.	n.d.
Outdoor		
RRIES Gloveline (examination gloves)		
Latex tank	125	104
Pre-leach	13	340
Post-leach	120	126
Slurry	99	140
Strip area	100	150
Packing room (powdered)	n.d.	
Outdoor	101	231

*Mean of 2 replicate samples
n.d.: not determined

and powder-free packing rooms at Factory A (*Table 2*). In contrast, the populations of airborne fungi was relatively less. At the RRIES Gloveline, the numbers of organisms from the air measured was far less in the strip areas, latex tank and in the packing room. At both these sites, there was no correlation between solution endotoxins and the numbers of bacteria in the air above the processing tanks. At factory B, high populations of aerobic bacteria were found in both leaching tank and stripping areas. For factory B, the higher numbers of airborne bacteria reflect the higher levels of endotoxins in these areas.

Culturable Aerobic Bacteria and Fungi in Solutions

Apart from the slurry, the numbers of culturable aerobic bacteria in almost all tanks were low at Factory A and the RRIES Gloveline (*Table 3*). On the other hand, fungi

TABLE 3. MEAN TOTAL VIABLE COUNTS OF AEROBIC BACTERIA AND FUNGI RECOVERED IN SOLUTIONS DURING GLOVE MANUFACTURE

Location	Microbial population (No.s/mL)	
	Bacteria	Fungi
Factory A (surgical gloves)		
Latex	0	0
Inlet water	5 × 101	0
Pre-leach	3.5 × 101	0
Post-leach 1	5	0
Post-leach 2	5	0
Slurry	1.07 × 103	0
Pre-irradiated gloves (n=4 pairs)	1.91 × 101	0
Factory B (surgical gloves)		
Latex	n.d.	n.d.
Inlet water	n.d.	n.d.
Pre-leach	n.d.	n.d.
Post-leach 1	n.d.	n.d.
Post-leach 2	n.d.	n.d.
Slurry	n.d.	n.d.
RRIES Gloveline (examination gloves)		
Latex	1.91 × 101	0
Inlet water	n.d.	n.d.
Pre-leach	5.62 × 104	0
Post-leach 1	2.35 × 102	0
Post-leach 2	–	–
Slurry	1.54 × 102	1.83 × 102

*Means of 2 replicates
n.d.:not determined

were not observed in these solutions, except for the slurry tank at the RRIES Gloveline. The poor correlation between microbial counts in the solution tanks as against their endotoxin concentrations could be due to the unsuitable culture media used. This would mean that a variety of different culture media would have to be used to obtain a larger proportion of bacterial populations for bacterial counts to be used as a good predictor of endotoxin levels. The higher numbers of bacteria in the slurry at both these sites also did not correlate with the lower concentrations of endotoxins, indicating that much of these bacteria were not Gram-negative types.

Bacterial Identification

A variety of contaminant bacteria was detected at these glove manufacturing factories, with a predominance of Gram-negative types and therefore responsible for the high endotoxin contents measured (*Table 4*). *Burkholderia glumae* was the most frequent bacteria isolated and identified from liquid samples of inlet water, slurry tank, and powder gloves from Factory A. These bacteria were also found in latex dip tank from Factory B. From the species composition studied, the numbers of bacteria isolated from Factory B appeared fewer and not proportional to the high level of

TABLE 4. CULTURABLE AEROBIC BACTERIA IDENTIFIED

Source	Gram reaction	BIOLOGTM Identification
Factory A		
Inlet water	GN	Burkholderia glumae
Pre-leach tanks	GN	Actinobacillus capsulatus
	GN	Pantoea stewartii subsp. stewartii
	GN	Vibrio campbelli
Post-leach tanks	GN	Pseudomonas stutzeri
Slurry tank 1	GP	Unidentified
	GP	Clavibacter agropyri (Corynebacterium)
Slurry tank 2	GN	Burkholderia glumae
	GN	Sphingomonas sanguinis
Gloves	GP	Clavibacter agropyri (Corynebacterium)
	GN	Burkholderia glumae
	GN	Pseudomonas syringae pv. persicae
Powder	GN	Pantoea stewartii subsp. stewartii
	GN	Burkholderia glumae
Factory B		
Latex dip	GN	Burkholderia glumae
Pre-leach tanks	GN	Rahnella aquatilis
Physical testing room	GN	Acinetobacter baumanii genospecies 2
	GP	Brevibacterium casei
Packing room (bench)	GN	Mersocella esiciensis
	GN	Acinetobacter calcoaceticus genospecies 1

endotoxin detected because the Biolog system could not identify some of the isolates.

CONCLUSION

This study gives an insight into endotoxin exposure and microbial contamination during commercial glove manufacture. At all three plants, high airborne endotoxin concentrations were encountered in the stripping areas and the packing rooms, relative to the concentration range along the processing tank lines. Endotoxin was also detected from the surfaces at the stripping areas and in the packing rooms. The most likely cause for the final endotoxin concentrations on the formed gloves is the higher level of airborne endotoxins during stripping and packing activities that impact. The densities of airborne microbes were also high in the stripping area. The relatively lower endotoxin concentration in solutions at the slurry stage relative to the leaching tanks indicated that it could not have been a contributory factor to final endotoxin levels after drying out of gloves. In such contaminated environments, a variety of bacteria were recorded, with a predominance of Gram-negative types responsible for the high endotoxin levels recorded. Reducing the likelihood of bacterial contamination reduces both microorganisms and pyrogens and it is vital to place greater focus on reducing microbial contaminants at these critical areas of production.

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