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Effects of Added Nitrogen and Phosphorus on the Biodegradation of NR Gloves in Soil

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Biodegradation of NR latex gloves in an Oxisol in response to nitrogen (N) and phosphorus (P) application was compared with several synthetic alternatives (polychloroprene, nitrile and plasticised PVC). Pieces of glove materials placed in nylon-net mesh bags were buried for up to 40 weeks in soils within containers receiving a high – (100 mg L^{-1} N, 150 mg L^{-1} P) or a low – (10 mg L^{-1}, 15 mg L^{-1} P) nutrient treatment and an unamended control. Mass losses of NR glove pieces were rapid in the high-nutrient treatment compared to the low or the control treatment. The NR glove pieces retained 17.6% of its initial weight in the high-nutrient treatment at 40 weeks, whereas the synthetic materials remained intact except for plasticised PVC which retained 73.9% of its initial mass. The specific degradation rate of NR glove pieces, expressed as mass loss per week was three times higher under a high-nutrient regime than when left to natural attenuation processes. Degradation of the NR test pieces was consistent with the drastic reduction in tensile properties, and with the higher number of bacteria, actinomycetes and fungi colonising the material.

Key words: NR latex gloves; biodegradation; nitrogen; phosphorus; nutrient treatment; microbial colonisation; tropical soil

Hydrocarbon wastes or their derivatives added to soil and their mineralisation and formation are of significance to the terrestrial cycle of carbon[1]. The rapidity with which a given substrate is oxidised will depend on its chemical composition and the 'proper' physical/chemical conditions in the surrounding environment. The major environmental factors affecting the aerobic biodegradation of products include microbial population density, substrate bioavailability, the degree of physical contact between waste and soil microflora, availability of oxygen, mineral nutrient concentrations, temperature, soil acidity and moisture contents, and the presence of toxic/inhibitory materials[1–4]. Furthermore, the presence of a product in soil stimulates an existing microbial community to adapt to this changed environment.

Nutrients are needed to sustain optimal growth and metabolic activity of degrading organisms, with nitrogen (N) and phosphorus (P) being the major inorganic nutrients used by the microbial cell. Environments containing low concentrations of inorganic nutrients often produce excessively high C/N or C/P ratios, or both, which are unfavourable

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for microbial growth. Many tropical surface soils are nutrient-deficient in plant growth terms but these can be readily corrected through common agricultural fertilisers. Hence manipulating the treated site to favour microbial growth promotes biodegradation and alleviates potential pollution.

The efforts of many workers in studying biodeterioration of vulcanised rubber products under varying conditions have been reviewed[5–9]. In this paper, the effects of N and P supply on the biodegradation of NR gloves were compared with several synthetic alternatives in a clayey tropical soil.

MATERIALS AND METHODS

The experimental treatment was a factorial combination of (a) glove materials, (b) applied nutrients and (c) sampling stages, in a randomised-block design with three replications.

Gloves

The examination gloves used were normal NR gloves from the RRIM Glove Plant (Run 17/99), and commercial samples of polychloroprene (Neoprene®) which was a gift from Ansell Malaysia Sdn Bhd; nitrile (N-Dex®, USA) and plasticised polyvinyl chloride also known as PVC or vinyl (Tru-Touch™, USA). The gloves were cut into 15 × 9 cm^2 rectangular pieces and individually weighed before being placed in 20 × 13 cm nylon-net bags (38 μm pore size; STRENO Teknisk, Farum, Denmark). Each bag contained one glove piece, and the bags were buried vertically in soil. For a comparison, Manila-grade brown envelope paper (thickness, 143 μm) was included as a degradable control.

Soil, Applied Nutrients and Experimental Units

The soil used (Munchong series; Typic Hapludox) *(Table 1)* was collected from Field 54 (0–30 cm depth) at the RRIM Experiment Station, Sg. Buloh. The soils were air-dried, sieved (<5 mm), sub-sampled for measurements of field capacity, physical and chemical properties, and limed to pH 6.5.

The soil nutrient treatments were as follows: Control, high (100 mg L^{-1} N and 150 mg L^{-1} P) and low (10 mg L^{-1} N and 15 mg L^{-1} P) nutrient supply. N and P were applied in solution as NH_4NO_3 (35% N) and KH_2PO_4 (22.76% P), respectively, to the sieved soils, allowed to dry and shaken thoroughly before dispensing into 28.5 cm diameter (30 cm high) plastic pails (11 kg soil/pail). The pails were kept at 25°C and watered weekly to maintain a 40% field capacity throughout the experiment.

Sampling and Measurements

The glove pieces were retrieved at 12, 24 and 40 weeks after burial from the nylon-net bags and transferred to bottles of sterile 50 mL diluent[10] containing a non-ionic detergent (0.05%) and 1.0 mm glass beads (4 g) for estimating counts of microorganisms. The bottles were subjected to mechanical shaking (250 r.p.m.) for 20 min – 30 min. Population densities of bacteria and actinomycetes from 10-fold dilution series of the glove washings were made on tryptic soy agar (TSA, Difco Laboratories, Detroit, MI) containing 50 mg L^{-1} cycloheximide (Sigma Chemical Co., St. Louis, MO)[11] and total fungi on peptone-dextrose agar (BBL, Cockeysville, MD) containing 33 mg L^{-1} rose bengal and 30 mg L^{-1} streptomycin (Sigma Chemical Co., St. Louis, MO)[12]. Samples for initial microbial

TABLE 1. PROPERTIES OF THE SOIL USED

Property	
Coarse sand (%)	20.3
Fine sand (%)	9.8
Silt (%)	5.8
Clay (%)	64.1
pH	4.5
Org. C (%)	0.96
Total N (%)	0.09
Total P (p.p.m.)	446
Avail. P (p.p.m.)	6
Exch. K (cmol/kg)	0.04
Exch. Ca (cmol/kg)	0.12
Exch. Mg (cmol/kg)	0.03
Total Mn (p.p.m)	25

colonisation (designated as O-time) were obtained after one day of soil burial. The samples were later washed in running water and dried (60°C, 24 h) prior to weighing. Mechanical properties were measured according to *ISO 37* (Type 2) standards using an Instron model 5500 tensile testing machine. Small pieces from the dried NR glove samples were also prepared for microscopy.

Microscopy

Cut degraded glove pieces (1 mm^2) for light and transmission electron microscopy (TEM) were fixed with Sorensen's buffered OsO_4 overnight at room temperature, washed twice in phosphate buffer (10 min) and distilled water (10 min) and dehydrated in a graded alcohol series (10%, 20%, 50%, 70%, 90%) prior to infiltration with absolute ethanol:LR White resin (2:1) overnight. This was followed by pure LR White for 6 h and another change in pure LR White in airtight capsules for polymerisation at 60°C.

Embedded samples were cut off from the hardened resin, re-embedded in technovit with a new orientation, and sectioned with an ultramicrotome (Reichert Ultracut E) to 80 nm thickness. Subsequently, the sectioned samples were deposited on a glass slide for light microscopy and on formvar coated grids for electron microscopy. Sections were multiple-stained (light microscopy) or stained with uranyl acetate and lead citrate (TEM) for examination under a Phillips Transmission Electron Microscope CM12 at 40 kV.

Degraded NR glove pieces (25 mm^2) for scanning electron microscopy (SEM) were mounted on specimen stubs using double-sided tape, sputter-coated (JEOL model JFC-1100) with gold to a thickness of 10 nm and examined in a scanning electron microscope (JEOL JSM-5300) with secondary electrons at 20 kV acceleration voltage. The untreated control sample was taken from the centre palm area, washed with water and air-dried prior to coating and SEM observation.

Statistics

Analysis of variance was carried out on all the measured variables and the treatment means compared at the $P<0.05$ probability level.

RESULTS

Mass Loss

The rates of breakdown of the glove materials were assessed by changes in mass loss. After 12 weeks of burial, breakdown of

NR and the paper control were higher in the high nutrient treatments compared to the low nutrient treatment or the unamended controls (*Table 2*). Neoprene, nitrile and vinyl showed no significant weight losses between all treatments.

After 24 weeks, NR also degraded faster in the high nutrient treatment than in the low or control treatments. Irrespective of treatment levels, the synthetic glove materials showed no significant loss from the controls.

By 40 weeks, the NR test pieces had a mean of 17.6% of its initial weight remaining in the high nutrient treatment. With one exception, the synthetics neoprene and nitrile showed no significant losses from the controls. In contrast, vinyl mass losses in the high nutrient treatment was significantly higher than in the controls. For the same period, the Manila-grade paper degraded the fastest with only 4.7% of its initial weight remaining in the high nutrient treatment.

TABLE 2. MEAN PERCENT OF INITIAL WEIGHT REMAINING OF DECOMPOSING GLOVE MATERIALS BURIED IN SOIL AS AFFECTED BY NUTRIENT TREATMENTS[a]

Gloves material	Nutrient treatment		
	High	Low	Control
(a) 12 weeks			
NR	63.0 g	82.1 ef	86.4 de
Neoprene	102.3 ab	101.2 a–c	104.9 a
Nitrile	97.5 bc	98.7 bc	98.3 bc
Vinyl	86.8 de	92.4 cd	94.1 b–d
Manila-paper	15.6 h	60.6 g	74.4 f
(b) 24 weeks			
NR	38.5 g	76.4 e	82.7 c–e
Neoprene	112.2 a	106.6 ab	119.1 a
Nitrile	97.2 bc	97.6 bc	97.1 bc
Vinyl	82.9 de	88.6 c–e	91.3 cd
Manila-paper	*5.5 h*	*45.5 fg*	*56.3 f*
(c) 40 weeks			
NR	17.6 h	61.5 f	70.3 ef
Neoprene	100.3 a	87.0 cd	98.9 ab
Nitrile	95.7 a–d	96.8 a–c	96.5 a–d
Vinyl	73.9 e	86.6 d	88.9 b–d
Manila-paper	4.7 i	16.9 h	30.8 g

[a] Means of 3 replicates, 15 × 9 cm^2 glove pieces. For each sampling stage, values across rows and columns not followed by common letters are significantly different ($P<0.05$)

At all these sampling stages, differences in mass loss of the NR glove pieces between the low and the unamended control treatment were not significant. The regression equations describing the mass loss trajectories for the degrading NR glove pieces were as follows:

- High nutrient: $Y = -1.605X + 80.375$ ($r^2 = 0.983$)
- Low nutrient: $Y = -0.749X + 92.316$ ($r^2 = 0.971$)
- Unamended control: $Y = -0.605X + 96.805$ ($r^2 = 0.763$)

where Y is the percent of initial mass remaining and X, the time in weeks. A comparison of the linear regression slopes shows the different rates of mass loss, with the more rapid loss rate from the high nutrient treatment.

The specific degradation rate, expressed as mass loss per week, were highest in the initial stages of burial for all treatments (*Table 3*). The average rate of NR biodegradation in the high nutrient treatment (36.9 mg/week) was three times higher than that in the unamended controls (12.0 mg/week).

Microbial Colonisation of Glove Pieces

The populations of bacteria colonising NR glove pieces at 12, 24 and 40 weeks were significantly higher in the high nutrient treatment compared to the low or unamended control treatment (*Table 4*). The populations of bacteria were also significantly higher in either the high or low nutrient treatments relative to the controls at 12 and 24 weeks for neoprene and vinyl, or at 24 weeks for nitrile. However, the number of bacteria for all the synthetic materials except vinyl at 40 weeks were not different between all treatments.

The fungal populations on NR glove pieces in the high nutrient treatment were also significantly greater than populations in the low and control treatments during the later stages (24, 40 weeks) of degradation (*Table 5*). Within the high nutrient treatment for NR test pieces, there appeared to be a dramatic increase in numbers from 12 weeks onwards. The effect of high nutrient application alone in significantly increasing fungal populations throughout the 12, 24 and 40 week samplings occurred in vinyl. The effect of nutrient levels, both high and low, in increasing fungal populations of the remaining synthetic

TABLE 3. AVERAGE SPECIFIC DEGRADATION RATE OF NR GLOVE PIECES DURING SOIL BURIAL

Treatment	Specific degradation rate (mg/week)[a]			Average
	0 – 12 weeks	0 – 24 weeks	0 – 40 weeks	
High	50.3	25.8	34.7	36.9
Low	25.1	9 7	15.8	16.9
Control	18.8	5.1	12.0	12.0

[a] Calculated as (Final weight – initial weight)/Time

TABLE 4. BACTERIAL COLONISATION OF GLOVE PIECES DUE TO NUTRIENT APPLICATION DURING SOIL BURIAL[a]

Glove material	Sampling stage (weeks)	Nos/mg weight glove piece		
		High	Low	Control
NR	0	5.9115 s–v	6.6153 p–t	5.2543 t–v
	12	13.5266 a–c	11.8187 d–f	11.7158 d–g
	24	14.3134 a	12.3733 b–d	10.5719 e–k
	40	13.8888 ab	11.5066 d–g	10.2139 g–l
Neoprene	0	6.1946 q–u	5.7004 l–v	6.2093 q–u
	12	10.6966 e–i	10.6733 e–j	9.0172 k–o
	24	10.6991 e–i	9.1099 j–n	7.6697 o–r
	40	9.9098 h–m	8.6960 l–o	8.2382 no
Nitrile	0	6.1535 r–v	7.5065 o–s	5.8372 t–v
	12	11.8813 d–f	10.3376 f–k	10.8398 d–i
	24	11.8484 d–f	10.9757 d–h	8.5954 m–o
	40	11.2770 d–h	10.7782 e–i	11.1272 d–h
Vinyl	0	4.6037 v	4.8669 uv	8.1742 n–q
	12	11.6939 d–g	11.0862 d–h	9.3349 i–n
	24	12.0693 c–e	10.1402 g–m	7.7405 o–q
	40	10.1932 g–l	8.6936 l–o	8.7211 l–o

[a] Means of 3 replications, as ln (X) transformed data. Values across rows and columns not followed by common letters are significantly different ($P<0.05$)

materials occurred in some cases, for *e.g.* on neoprene at 24 weeks, and on nitrile at 12 weeks.

The numbers of actinomycetes colonising NR glove pieces at 12, 24 and 40 weeks were also significantly greater in the high nutrient treatments *(Table 6)*. The effect of nutrients on actinomycete populations for the remaining glove materials occurred in some instances, for *e.g.* at 24 and 40 weeks for neoprene and vinyl, and at 12 and 24 weeks for nitrile.

Tensile Properties

There was a drastic reduction in tensile strength, elongation at break and modulus of the NR glove test pieces after soil burial (*Table 7*). After 12 weeks, the samples could not be tested due to severe degradation. The tensile properties of neoprene and nitrile gloves were slightly lowered after 40 weeks burial. In contrast, buried vinyl gloves gave an increased tensile strength and moduli values by 40 weeks.

TABLE 5. FUNGAL COLONISATION OF GLOVE PIECES DUE TO NUTRIENT APPLICATION DURING SOIL BURIAL[a]

Glove material	Sampling stage (weeks)	Nos/mg weight glove piece		
		High	Low	Control
NR	0	3.0693 q–u	4.6171 i–p	6.6714 b–d
	12	4.5815 i–q	4.1766 j–t	4.6831 h–o
	24	6.7386 bc	4 9865 e–m	3.6726 l–u
	40	10.2297 a	4.8624 f–n	6.1727 b–h
Neoprene	0	2.8141 s–u	3.3723 n–u	2.8277 r–u
	12	5.9599 b–i	3.1109 p–u	2.6439 tu
	24	5.5943 c–k	4.1787 j–t	2.5828 u
	40	5.6569 c–j	4.9288 f–m	4.0894 k–u
Nitrile	0	3.8859 l–u	5.1547 d–l	4 3667 j–r
	12	7.4818 b	5.7048 c–j	3.9354 l–u
	24	6.3189 b–g	4.8701 f–n	3.5004 m–u
	40	6.7415 bc	6.4998 b–e	4.7411 h–n
Vinyl	0	3.1709 o–u	4.7963 g–n	4.3477 j–s
	12	6.4029 b–f	4.2486 j–s	3.1194 p–u
	24	6.0453 b–i	3.9238 l–u	2.8951 r–u
	40	6.1990 b–h	3.0289 r–u	4.0429 l–u

[a]Means of 3 replications, as ln (X+1) transformed data. Values across rows and columns not followed by common letters are significantly different ($P<0.05$)

Microscopy

Light microscopy of the NR glove pieces (X600) showed microorganisms growing embedded in the rubber matrix (*Figures 1* and *2*), beginning with weak colonisation at 12 weeks of soil incubation and progressing to dense mycelial masses as colonisation increased during the time course of the experiment. The rubber matrix appeared completely covered with hyphae by 40 weeks. The late samples showed material disintegration and under TEM, spores were seen formed within the rubber matrix.

The morphology of the degraded NR glove surfaces as examined in the SEM revealed obvious traces of severe deterioration in the form of craters and cavities (*Figure 3*).

DISCUSSION

Biodegradation is a removal mechanism that results in a net decrease in contaminant mass. The paucity of data on the rate of disappearance of NR gloves in soils from laboratory and field experiments limits judgement on their degradation potential in

TABLE 6. ACTINOMYCETE COLONISATION OF GLOVE PIECES DUE TO NUTRIENT APPLICATION DURING SOIL BURIAL[a]

Glove material	Sampling stage (weeks)	Nos/mg weight glove piece		
		High	Low	Control
NR	0	2.4840 l–q	3.7992 i–n	2.3084 l–q
	12	11.0652 a	8.3458 b–e	7.2809 c–f
	24	11.3611 a	3.8000 i–n	1.8691 n–q
	40	11 3718 a	4.2993 h–l	4 1348 h–m
Neoprene	0	1 4706 pq	1.7368 n–q	1.4773 pq
	12	6.0076 f–h	1.5467 o–q	4 9129 g–j
	24	9.6479 ab	6.6283 d–g	2.7871 k–q
	40	7.3470 *c–f*	3.1809 *j–p*	4.7610 g–k
Nitrile	0	1.3947 pq	1.5525 o–q	1.0872 pq
	12	6.2494 e–h	6.8373 d–g	3.7716 i–n
	24	9.4548 a–c	5.6145 f–i	3.7252 i–o
	40	5.993 f–h	5.6534 f–i	3.7977 i–n
Vinyl	0	0.9162 q	1.3683 pq	1.2726 pq
	12	5.2175 f–j	5.1605 f–j	4.2656 h–l
	24	8.6503 b–*d*	4.7416 g–k	1.9040 n–q
	40	5.3652 f–j	6.1316 f–h	1.9717 m–q

[a] Means of 3 replicates, as ln (X+1) transformed data. Values across rows and columns not followed by similar letters are significantly different ($P<0.05$)

landfills. As not all environmental conditions are within optimal ranges in the field, natural attenuation processes, both biological and physical, occur and nature therefore caters to the nutritional and physiological needs of indigenous degrading microorganisms. Fertiliser application of N and P to an aerobic environment potentially increases the rate of biodegradation by stimulating growth of indigenous heterotrophs. For instance, in the bioremediation of petroleum hydrocarbons, soil N levels of 20 p.p.m. and P at 10 p.p.m. were needed to supplement growth of microorganisms[13]. It was clear from this experiment that mass losses of NR glove materials under elevated levels of N and P were more rapid compared to natural biodegradation in unamended soils. In undisturbed soils where a delicate equilibrium between N gain and losses exist, little readily-available N is present and any that is added, especially when accompanied by liming or additions of phosphate, is rapidly utilised (immobilised) by the soil microflora[14]. The fit of the linear regression models describing the mass loss trajectories of NR test pieces as indicated by the r^2 values was good. On the other hand, the synthetic glove materials with the exception of vinyl remained intact after 40 weeks. Vinyl glove pieces did not biodegrade in soils and the small mass losses were confined to plasticiser and other additive losses[15].

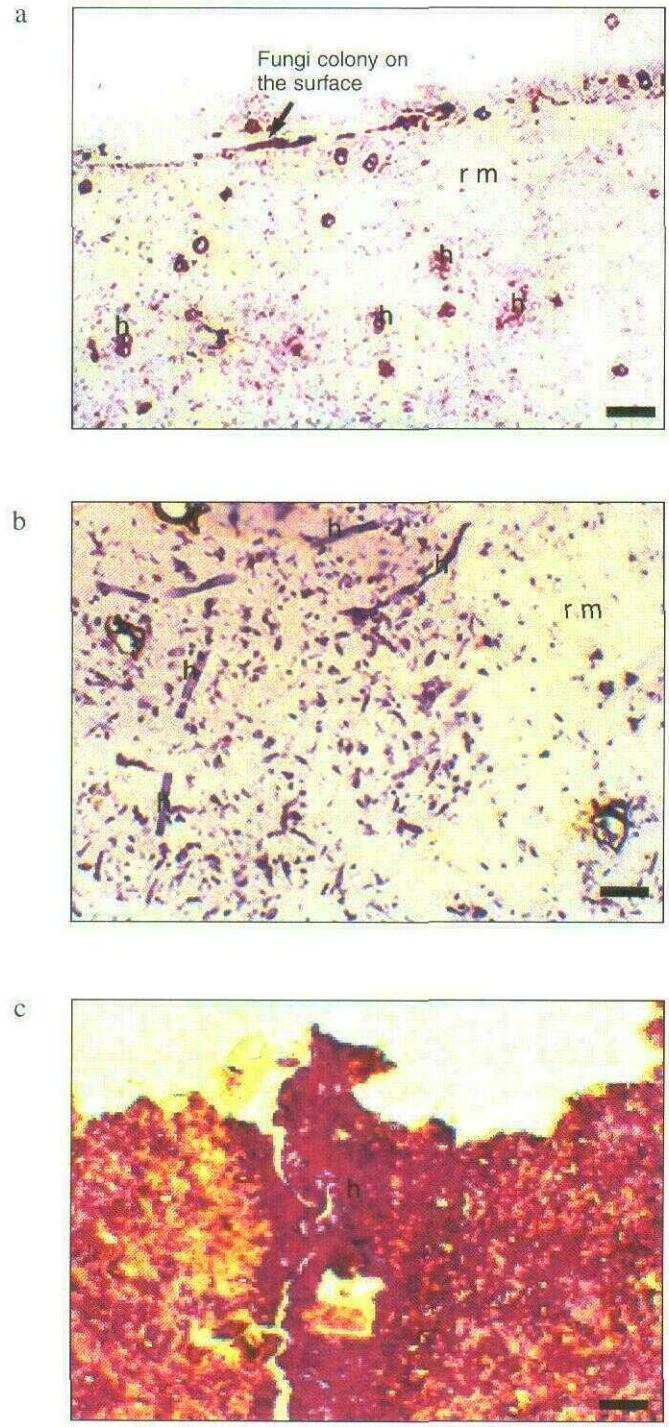


Figure 1. Micrographs of degraded NR latex glove pieces showing hyphae on surfaces and within the rubber matrix at: (a) 12 weeks; (b) 24 weeks; and (c) dense colonisation at 40 weeks. Bars = 10 μm; h = hyphae and rm = rubber matrix.

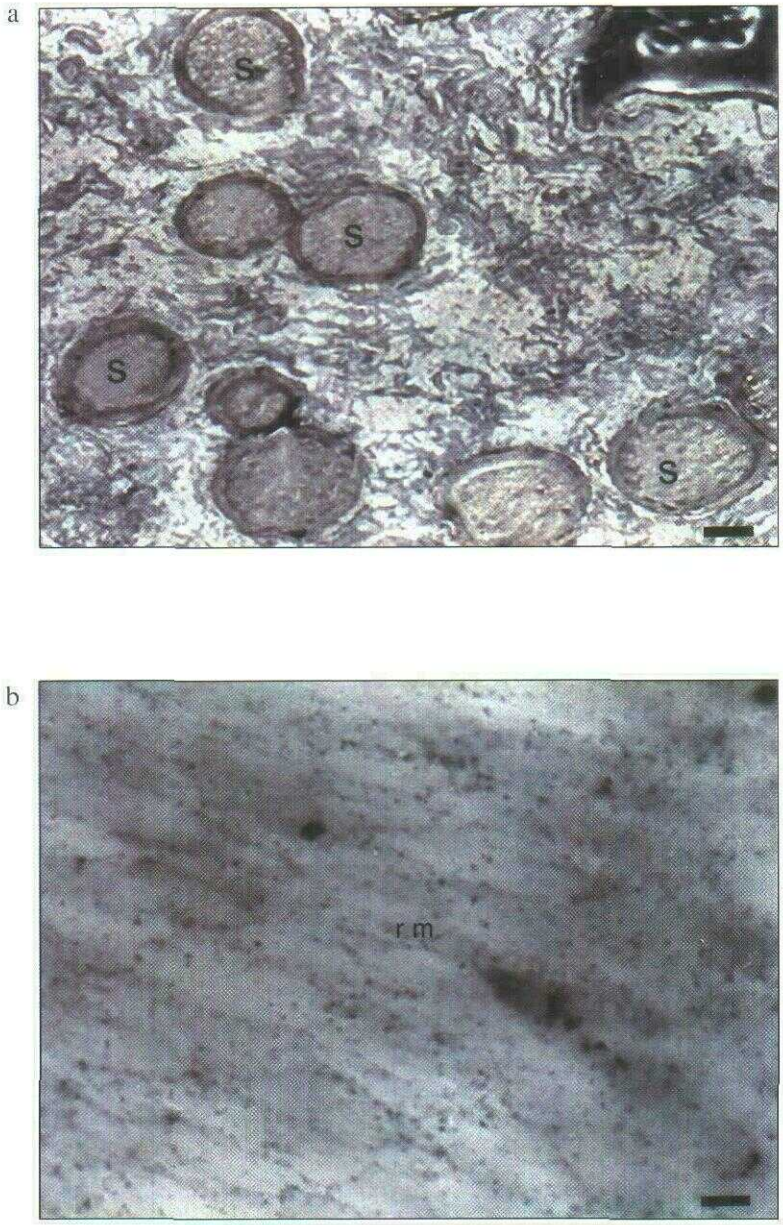
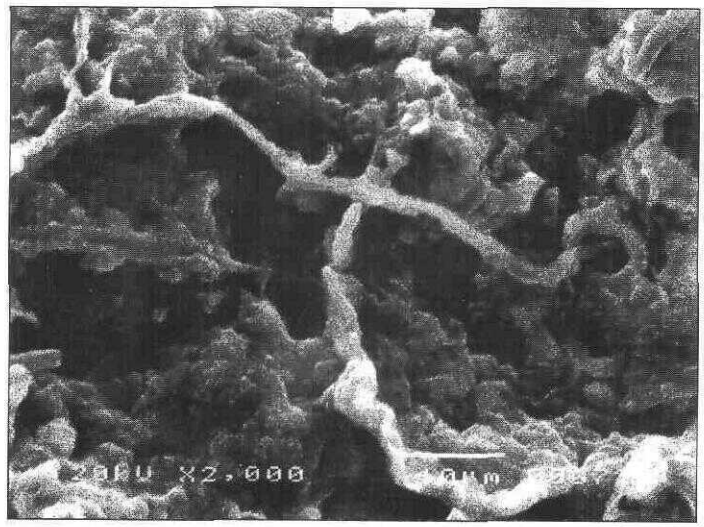


Figure 2. Transmission electron micrographs of degraded NR latex glove pieces at 40 weeks showing: (a) spores; (b) unburied controls. Bars = 2 μm (a); and 1 μm (b). s = spore; rm = rubber matrix.

a



b

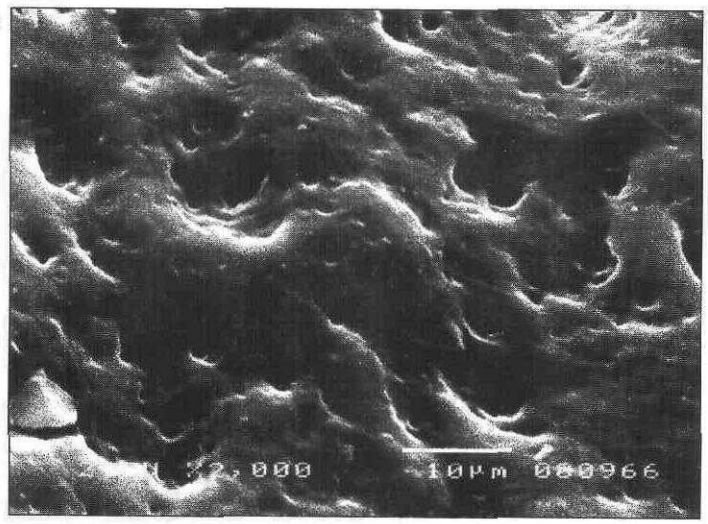


Figure 3. Scanning electron micrographs showing: (a) the effect of soil burial (40 weeks) on the surface appearance of NR latex glove specimens; and (b) unburied control. Bar = 10 μm.

TABLE 7 TENSILE PROPERTIES OF NR, NEOPRENE, NITRILE AND VINYL GLOVE PIECES AS AFFECTED BY LEVEL OF NUTRIENTS APPLIED TO SOIL DURING BURIAL TESTS[a]

Material	Treatment	Sampling stage (weeks)			
		0–time	12	24	40
(a) Tensile strength, MPa					
NR	High	19 3	–	–	–
	Low	8 4	1 5	–	–
	Control	16 0	3 1	–	–
Neoprene	High	19 5	15 1	15 5	15 1
	Low	19 1	16 8	17 0	15 7
	Control	18 4	17 0	17 1	15 5
Nitrile	High	24 4	17 9	16 9	14 7
	Low	27 4	20 1	19 0	12 7
	Control	25 6	20 1	23 9	17 2
Vinyl	High	12 9	15 9	17 4	19 2
	Low	12 9	14 4	16 4	16 9
	Control	12 7	13.3	15 4	16 4
(b) Elongation at break (%)					
NR	High	925	–	–	–
	Low	750	400	–	–
	Control	880	650	–	–
Neoprene	High	800	750	750	750
	Low	800	750	750	750
	Control	800	750	750	750
Nitrile	High	600	650	650	650
	Low	600	600	650	650
	Control	600	600	600	650
Vinyl	High	400	400	400	350
	Low	400	400	400	400
	Control	400	400	450	400
(c) Modulus at 100%, MPa					
NR	High	0 5	–	–	–
	Low	0 4	0 4	–	–
	Control	0 4	0 7	–	–
Neoprene	High	1 1	1 3	1 2	1 3
	Low	1 1	1 2	1 1	1 3
	Control	1 1	1 3	1 3	1 3

TABLE 7 (CONT.). TENSILE PROPERTIES OF NR, NEOPRENE, NITRILE AND VINYL GLOVE PIECES AS AFFECTED BY LEVEL OF NUTRIENTS APPLIED TO SOIL DURING BURIAL TESTS[a]

Material	Treatment	Sampling stage (weeks)			
		0-time	12	24	40
Nitrile	High	1.5	1.2	1.0	1.0
	Low	1.5	1.2	1.0	1.0
	Control	1.5	1.2	1.2	1.1
Vinyl	High	6.0	7.5	9.2	10.0
	Low	5.6	6.5	7.0	7.7
	Control	5.5	6.0	6.1	7.5
(d) Modulus at 300%, MPa					
NR	High	0.9	–	–	–
	Low	1.0	0.9	–	–
	Control	1.0	1.2	–	–
Neoprene	High	1.8	2.0	2.0	2.2
	Low	1.8	2.0	2.0	2.2
	Control	1.8	2.2	2.0	2.0
Nitrile	High	3.0	2.2	2.0	1.7
	Low	3.2	2.5	2.0	2.0
	Control	3.0	2.5	2.5	2.0
Vinyl	High	10.9	14.1	15.9	17.0
	Low	10.6	11.9	12.4	14.7
	Control	10.7	11.2	11.1	13.5
(e) Modulus at 500%, MPa					
NR	High	1.7	–	–	–
	Low	1.7	1.2	–	–
	Control	1.7	2.2	–	–
Neoprene	High	3.0	3.5	3.7	4.0
	Low	3.0	3.7	3.6	4.0
	Control	2.9	3.8	3.8	3.7
Nitrile	High	8.2	5.2	4.0	3.5
	Low	10.0	7.5	4.5	4.0
	Control	9.0	6.2	7.2	4.5
Vinyl	High	–	–	–	–
	Low	–	–	–	–
	Control	–	–	–	–

[a] Median values of 3 replicate glove pieces

The biodegradation of the NR test pieces were also consistent with the drastic reduction in tensile properties, and with the higher numbers of microorganisms colonising the material under a high nutrient regime. The mean microbial populations on the synthetic glove materials, averaged over nutrient treatment and sampling stage, were significantly lower than on NR. An interesting observation was the dramatic increase in fungal populations on the degrading NR test pieces in the high nutrient application treatment towards the later stages of burial.

Microscopic examination of the deteriorated NR pieces showed dense colonisation by microbial hyphae on the surface and within the rubber matrix. This appeared to be the pattern of microbial growth and spread prior to final disintegration of the substrate. In this study, degraded areas initially appear as dark spots over the test pieces that eventually darkened the whole material. Potent degraders of NR glove materials have been reported to be actinomycetes[16-22] and Gram-negative bacteria[23,24]. Heisey and Papadatos[18] grew actinomycetes (*Streptomyces* sp. S1G, S4D and *Nocardia* sp. S3F) on NR latex gloves in aseptic cultures and showed penetration of the organisms into the rubber as well as growing embedded in the rubber matrix during 45 days incubation. Similarly, Linos and Steinbuchel[21] reported nocardioform actinomycetes (*Gordonia* sp.) able to disintegrate and solubilise solid rubbers in liquid cultures. Linos *et al.*[24] had also shown biofilm coverage of NR latex gloves by the Gram-negative bacterium *Pseudomonas aeruginosa* AL98 with cells merging into the rubber material prior to disintegration, in an analagous manner to some actinomycetes[20,21].

The rate and extent of product degradation is affected by the rubber formulations, the microorganisms present and their interactions with the environment. Resistance to biodegradation of an industrial product can be due to the presence of curing agents and highly effective antioxidants (phenol, quinone or aromatic amine derivatives) added during manufacture. Although the normal range of inclusion levels of the accelerators, zinc oxide and antioxidants had little effect in reducing biodeterioration of vulcanised rubber samples[25,26], extraction of antioxidants from NR latex gloves by organic solvents was shown to enhance growth of rubber-degrading bacteria belonging to species of *Gordonia*, *Mycobacterium*, *Micromonospora* and *Pseudomonas*[22]. The NR gloves used in this study had 0.5% alkylated monophenol as the antioxidant and the inclusion level did not indicate a reduction of microbial activity. The presence of antioxidants in the formulation of the synthetic glove materials were however proprietary information. The mechanism of NR biodegradation is initially peroxidation, biotic or abiotic, followed by biodegradation of the low molecular weight oxidation products and their bioassimilation into the environment[27]. Products generated during oxidation of the NR poly(isoprene) chain include CO_2, formic acid, formaldehyde, acetic acid, levulinaldehyde and laevulinic acid[28,29]. The role of a monooxygenase enzyme in the initial oxidative attack of the double bonds in the poly(isoprene) chain has been implicated although the enzymes were never isolated, characterised nor genetically assigned[16,23,24].

We have shown in this study that the results were related to the nutritional limitations of the different microbial populations in the soil community, and that the rate of NR glove biodegradation in acid infertile soils can be enhanced by extraneous supplies of inorganic nutrients.

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