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Differential Sampling of Footwear to Separate Relevant Evidentiary Particles from Background Noise

Final Technical Report 3/29/16

Award 2014-DN-BX-K011

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### Abstract

Everywhere people walk, very small particles (VSP) transfer to and from their footwear. The mere presence at a crime scene requires this contact and transfer. VSP on footwear are routinely ignored by forensic science unless there are obvious accumulations of soil or conspicuous visible particles. There is an extraordinary, untapped potential to exploit VSP found on footwear.

One predominant challenge remains to unlock this potential: VSP are invariably present as mixtures of materials that can originate before, during, or after any event of forensic interest. Their usefulness depends on our ability to separate a reliable, relevant evidentiary "signal" from the background noise.

This project tested the ability to separate particle signals on the contact surfaces of footwear soles using differential analysis of loosely held, moderately held, and strongly held particle fractions. Project objectives were to: (1) design and conduct a series of realistic environmental exposures suitable for testing of VSP transfer, retention and fractionation, (2) design, develop and test a differential sampling protocol suitable for testing the physical fractionation of VSP signals, and (3) test the ability to use differential sampling to separate specific VSP signals of interest.

Three environmental exposure sites were chosen to have different, characteristic particle types (soil minerals). Shoes of two types (work boots and tennis shoes) were tested, accumulating particles by walking 250 m in each environment. Some shoes were exposed to only one environment; others were exposed to all three, in one of six different sequences.

Sampling methods were developed to separate particles from the contact surface of the shoe based on how tightly they were held to the sole. Loosely held particles were removed by walking on paper, moderately held particles were removed by electrostatic lifting, and the most tightly held particles were removed by moist swabbing.

The resulting numbers and types of particles were determined using forensic microscopy. Particle profiles from the different fractions were compared to test the ability to objectively distinguish the order of exposure to the three environments.

Without exception, the samples resulting from differential sampling are dominated by the third site in the sequential footwear exposures. No noticeable differences are seen among the differential samplings of the loosely, moderately and strongly held particles: the same overwhelming presence of the third site is seen. It is clear from these results that the third (final) exposure results in the nearly complete removal of any particles that were transferred to the contact surfaces of the shoe from the first and second exposures. This occurs regardless of the exposure sequence and regardless of which specific site was used for the third exposure. It is also clear that under the experimental conditions loosely, moderately and strongly held particles are affected similarly, without any detectable enrichment of the earlier exposures among the more tightly held particles.

This finding is significant in that it fails to follow prior research focused on the persistence of trace evidence generally, and on footwear specifically, that strongly supports the hypothesis that, after transfer to an item, some particles are tightly held (and retained longer), while others are loosely held (and more rapidly lost). Contact surfaces of footwear, under the experimental conditions, are clearly an exception. Given that prior research has shown, in comparable studies, that a generalized sampling of footwear soles (from both contact and recessed areas) shows the retention of particles from earlier contacts, the clear implication of the present research is that, although particles *on the contact surfaces* of footwear are removed and replaced, those that are present *on the more recessed areas* of the sole are not.

The results of this project have important implications for guiding follow-on research, notably: (1) research on differential sampling of footwear should continue, focusing on the difference between particle populations found on contact surfaces and those found on recessed areas, and (2) research on related computational and statistical methods to objectively interpret mixtures of particles should continue, focusing on multivariate methods that take advantage of both qualitative and quantitative distinctions among traces and possible sources.

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## Executive Summary

Although it is well-recognized that criminals track dusts to and from every crime scene, dust particles on a suspect's shoes are very seldom used as evidence linking the accused to the crime. The major obstacle preventing the use of this type of evidence is that the shoes have mixtures of particles arising from activity before, during and after the crime itself. Methods separating the evidentiary particle "signal" from background noise would enable a powerful new and widely-applicable forensic capability. This capability would augment traditional footwear pattern evidence with objective quantitative associations, addressing one of the specific issues raised in the 2009 NAS report.

Our prior NIJ research[1,2] has shown corresponding particle sets to provide extremely strong, objective, quantitative, associative evidence. The current project tested the hypothesis that, by separately analyzing loosely held, moderately held, and strongly held particle fractions, we would be able to detect sequential footwear exposures and enable an important application of this new forensic capability.

The project was designed to test the separation of particle signals on the contact surfaces of footwear by applying a series of successively more aggressive sampling steps and contrasting the resulting types and quantities of particles. Project objectives were (1) to conduct suitable environmental exposures, (2) to develop differential sampling methods, and (3) to test the ability to separate small particle signals of interest.

Three environmental exposure sites were chosen to have different, characteristic particle types (soil minerals). Shoes of two types (work boots and tennis shoes) were tested, accumulating particles by walking 250 m in each environment. Some shoes were exposed to only one environment; others were exposed to all three, in one of six different sequences.

Sampling methods were developed to separate particles from the contact surface of the shoe based on how tightly they were held to the sole. Loosely held particles were removed by walking on paper, moderately held particles were removed by electrostatic lifting, and the most tightly held particles were removed by moist swabbing.

The resulting numbers and types of particles were determined using forensic microscopy. Particle profiles from the different fractions were compared to test the ability to objectively distinguish the order of exposure to the three environments.

Without exception, the samples resulting from differential sampling are dominated by the third site in the sequential footwear exposures. No noticeable differences are seen among the differential samplings of the loosely, moderately and strongly held particles: the same overwhelming presence of the third site is seen. It is clear from these results that the third (final) exposure results in the nearly complete removal of any particles that were transferred to the contact surfaces of the shoe from the first and second exposures. This occurs regardless of the exposure sequence and regardless of which specific site was used for the third exposure. It is also clear that under the experimental conditions loosely, moderately and strongly held particles are affected similarly, without any detectable enrichment of the earlier exposures among the more tightly held particles.

The hypothesis guiding this work was that distinguishable fractions would result from the differential sampling of the contact surface of footwear. That is, that by separating loosely held, moderately held, and strongly held particle fractions we would recover enriched particle fractions originating from different exposures, enabling the recognition and specification of sequential footwear exposures. This hypothesis is rejected. Under the experimental conditions the contact surface of footwear was found to be overwhelmingly dominated by the most recent exposure.

This finding is significant in that it fails to follow prior research focused on the persistence of trace evidence generally,[9-13] and on footwear specifically,[14,15] that strongly supports the hypothesis that, after transfer to an item, some particles are tightly held (and retained longer), while others are loosely held (and more rapidly lost). Contact surfaces of footwear, under the experimental conditions, are clearly an exception: a walk of 250 m, on a dry soil surface, results in the virtually complete removal and replacement of particles adhering to the contact surfaces from prior, similar exposures.

Given that Morgan et al.[15] have shown in comparable studies that a generalized sampling of footwear soles (from both contact and recessed areas) shows the retention of particles from earlier contacts, the clear implication of the

present research is that, although particles *on the contact surfaces* of footwear are removed and replaced, those that are present *on the more recessed areas* of the sole are not.

This research, based on realistic exposures to actual environments, and incorporating newly-developed differential sampling, has resulted in greater understanding of (1) how very small particles adhere to the soles of footwear, (2) how they can be separated, and (3) whether sequential exposures can be determined based on how tightly particles are held.

Prior work examining mixtures of particles on footwear had used test substances, simple sampling protocols, and single particle types to help determine the scope of the problem. This project used realistic field conditions, differential sampling protocols and combinations of co-occurring particle types to definitively test the adherence and transfer of very small particles adhering to the contact surfaces of footwear.

There are a number of important implications for criminal justice policy and practice in the United States.

*1. During successive exposures there is nearly complete, sequential displacement of particles from the contact surfaces of footwear.* This means that the contact surfaces of recovered footwear will have traces from the most recent areas where the footwear was worn. The implications of this finding are important. For example, in cases where a body is found and may have been transported after death from one location to another, the contact surfaces of the footwear will retain unmixed small particle traces that are directly representative of the last location where the deceased walked. Comparison with the location where the body was found will determine whether or not the body was moved and, if so, the traces will provide clues helping to locate the area from which the victim was transported. Alternatively, for footwear associated with a suspect, it is clear that the traces to be compared with crime scene locations are *not* those on the contact surfaces; rather they are those from recessed surfaces (see point 3, below).

*2. Methods for differential sampling of the contact surfaces of footwear need not be employed.* Given the absence of differences among loosely, moderately, and tightly held particle populations on the contact surfaces of footwear, there is no need for differential sampling of particles on these surfaces. Moist swabbing can be employed directly to comprehensively recover the adhering particles traces, now known to represent the most recent surfaces walked upon. This finding simplifies the collection of these particle traces and allows their separation as a distinct fraction from other accumulations of particles present on the footwear.

*3. Recessed areas of footwear are likely responsible for the observed retention of particles from prior exposures.* The mixing of particles on footwear, arising from activity before, during and after the crime itself, has been the major obstacle to the exploitation of this type of evidence as a means to test the association of an accused to a crime. Our research shows that the contact surfaces of the soles retain particles from only the more recent exposures. This indicates that the mixtures of particles seen on footwear arise from the recessed areas of footwear, and that it is these areas that should be sampled for evidence of prior exposures. Removal of the fraction from contact surfaces will reduce the complexity of the mixture and can lead to alternative approaches to differential sampling (see point 4, below).

*4. Research on differential sampling of footwear should continue, focusing on the difference between particle populations found on contact surfaces and those found on recessed areas.* Recessed areas of footwear are responsible for the mixtures of particles arising from activity before, during and after the crime itself. The results of our research allow the isolation of particles from the most recent exposures (by sampling the contact surfaces). Subtracting this background from the mixtures found within recessed areas of the footwear provides a likely means to separate the evidentiary particle “signal” from background noise and the exploration of this possibility remains an intriguing area for follow-on research.

*5. Research on related computational and statistical methods to objectively interpret mixtures of particles should continue, focusing on multivariate methods that take advantage of both qualitative and quantitative distinctions among traces and possible sources.* Given that the current differential sampling approach (based on how tightly particles are held) cannot be conveniently adapted to recessed areas of the soles, an alternative means to separate mixtures of particle signals needs to be developed. Following the approach in point 4 above, computational and statistical methods will be needed to subtract the “signals” from the most recent exposures (found on the contact surfaces) and objectively resolve the mixtures that are found in recessed areas.

## I. Introduction

### A. Statement of the Problem

Everywhere people walk, very small particles (VSP) transfer to and from their footwear. The mere presence at a crime scene requires this contact and transfer, and the particles are known to be persistent for long periods of time.[3,4] Even though criminals necessarily track dusts to and from every crime scene, dust particles on a suspect's shoes are very seldom used as evidence linking the accused to the crime. There is an extraordinary, untapped potential to exploit VSP found on footwear and in footwear impressions.

At the same time, there are significant challenges to unlocking this potential. Most fundamentally, VSP on footwear evidence are invariably a mixture of materials that can originate before, during, or after any event or period of forensic interest.[3,4] Their usefulness depends on our ability to separate a reliable, relevant evidentiary "signal" from background noise (or signals from other exposures). As an additional practical challenge, the VSP mixture is composed of many different particle types, which must be collected and analyzed efficiently.

We have developed methods to efficiently analyze VSP as part of our prior NIJ and other federally-funded research[5-8] and we have shown corresponding particle sets to provide extremely strong, objective, quantitative, associative evidence.[1,2] The application of these methods to VSP on (for example) shipping containers, clothing, and improvised explosive devices (IEDs) has consistently involved comparing VSP at different locations or different "layers" as one opens an item. The outer layer typically has VSP from the most recent exposures, while the innermost layer has that from earlier exposures. Intermediate layers have exposures whose relative timing depends on how and when the item was handled, assembled or opened. Invariably there is mixing among the layers, but by comparing and subtracting the particle "signals" among the levels (analogous to subtracting a background spectrum in IR spectroscopy) one is able to separate relevant evidentiary particles (for the problem at hand) from background noise.

On footwear the problem is more complex, as there are not physically separated layers (as there are in layers of packaging, or an assembled device). However, research focused on the persistence of trace evidence generally,[9-13] and on footwear specifically,[4,14] strongly supports the hypothesis that, after transfer to an item, some particles are tightly held (and retained longer), while others are loosely held (and more rapidly lost). There is a "trend of two/three stage decay..., with subsequently less rapid loss..., followed by a period of much lower decay." [15] Importantly, we observe that this explanation implies that *particles from earlier exposures will be more concentrated among the particles that are more tightly held*. From this strongly supported supposition, we hypothesized that, if we use differential sampling of footwear (which separates loosely held, moderately held, and strongly held particle fractions) we will recover *physically separated or enriched* particle fractions that originated from different exposures. This project explored the use of differential sampling of VSP from the soles of footwear as a potential method for the separation for these signals.

### B. Background and Context

*Footwear Evidence Generally.* Impressions from footwear are present and detectable at 30-60% of crime scenes.[16] The act of walking ensures close, firm contact between the soles of footwear and crime scene surfaces and the transfer of material is the fundamental basis for the production of 2-D impressions.[16,17] Footwear impressions are a mature form of physical evidence and are a major focus for proof of associations among crime scenes, victims and suspects. Comparison of crime scene impressions with test impressions from footwear are a specific focus of expertise,[16] with an associated Scientific Working Group[18] (now an OSAC sub-committee [19]) and certification.[20] Methods for the forensic analysis of footwear impressions are well developed,[16,17] including national consensus guidelines which include examiner training.[18] .

The examination of trace evidence on footwear is a separate discipline, and the need to preserve and accommodate trace evidence adhering to footwear is explicitly accommodated in guidelines for impression analysis.[21] Likewise, the need to accommodate the pattern analysis of impressions is explicitly accommodated in guidelines for trace evidence analysis.[22]

Trace evidence examiners encounter footwear as part of clothing examinations. Trace evidence commonly found on

footwear includes the major types of fragmentary material traces: glass, paint and fibers,[23,24] as well as accumulations of soil. When both footwear impressions and trace evidence are considered in a case, the trace evidence examination and recovery is performed first,[16] after which the footwear is made available to the footwear specialist for pattern examinations and test impressions. Trace evidence within shoe impressions is only rarely utilized as part of the comparison, although the potential to compare this form of trace evidence with footwear is well-recognized[17] and as methods in forensic geoscience have developed,[3,25-29] such cases are being reported.[30]

*Soil and Dust on Footwear.* Accumulations of soil on footwear or other items of evidence (such as digging tools and vehicles) have long been exploited for comparisons with reference samples of possible origin.[31] The long-standing focus has been on fairly large accumulations of soil that can reasonably be expected to be minimally mixed, or that are clearly layered, so as to allow physical separation of discrete samples. Only then can comparisons be reliably made using bulk properties of soil (such as color, particle size distributions and elemental composition). In cases where significant mixture has occurred, analysis of soil evidence is frequently stopped short as preliminary analyses indicate disparities in the bulk properties (e.g. color) that are typically used to screen for comparable specimens. This restriction severely restricts the numbers of applicable cases.

The work of Morgan, Bull and colleagues [3,4,32-35] has addressed this limitation, setting forth a conceptual framework for forensic geoscience [3] which includes specific emphasis on analytical methods that can recognize mixtures and that are applicable when mixtures are present. They describe these methods as "visual techniques" and have exploited quartz grain surface analysis for this purpose. Quartz occurs very widely in sediments and the physical appearance of quartz grains depends on fundamental geological mechanisms relating to their origin and transportation.[36] When two different sources of soil are mixed, expert quartz grain surface analysis can, with reasonable probability, detect this mixture, and comparisons of the different types of quartz grain surfaces can still be made.

This approach need not be limited to a single mineral type, or specifically to mineral particles, and it need not depend on the presence of one particle type (e.g. quartz) in each of the mixed sources.[37] What is essential is that recognizable varieties of minerals (or other particles) be exploited efficiently. Visual microscopical techniques do this: different soil or dust samples will have different suites of VSP. The presence and variety of VSP is a character that is recognizable within a mixture and meets the fundamental requirements for "visual techniques." [3,4]

*Studies of Particle Transfer and Persistence on Footwear.* Bull and Morgan's approach to recognition and analysis of mixed soil samples has continued with applied research directed at understanding mechanisms of transfer, persistence and mixing of particles deposited on footwear.[4,14] Experiments have been conducted using test substances (Plasticine) as well as using specific particle types (pollen or quartz grains). Specific case-related research has also been conducted.[34] These studies have demonstrated that (1) particles persist for a long periods of time on footwear, (2) that mixing of particles from successive exposures routinely occurs on the soles of footwear, and (3) that following exposure, some particles are loosely held (and more rapidly lost), while others are tightly held (and retained longer).

*Alternative Sampling Methods as Opposed to Differential Sampling.* Staged, alternative sampling methods are often employed in trace evidence analysis.[22,38-40] One purpose is to employ an initial method (such as picking individual fibers or paint chips) to collect loosely held traces as they are recognized, and that might otherwise be lost or redistributed as the examination proceeds. Another purpose is to preserve and document the location from which trace evidence was recovered (as in the regional taping of clothing in the recovery of fibers). Again, different methods may be used for alternative particle types (such as taping for fibers, followed by vacuuming to recover fine particles, or washing to recover pollen). However, there has not been a protocol for *differential sampling* and recovery of trace evidence with the express intention to fractionate loosely and tightly held particles, so that these populations can be compared and contrasted.

## II. Project Design and Methodology

This project was designed to test the separation of particle signals on the contact surfaces of footwear by applying a series of successively more aggressive sampling steps and contrasting the resulting types and quantities of particles. Project objectives were (1) to conduct suitable environmental exposures, (2) to develop differential sampling methods, and (3) to test the ability to separate small particle signals of interest.

Three environmental exposure sites were chosen to have different, characteristic particle types (soil minerals). Shoes of two types (work boots and tennis shoes) were tested, accumulating particles by walking 250 m in each environment. Some shoes were exposed to only one environment; others were exposed to all three, in one of six different sequences.

Sampling methods were developed to separate particles from the contact surface of the shoe based on how tightly they were held to the sole. Loosely held particles were removed by walking on paper, moderately held particles were removed by electrostatic lifting, and the most tightly held particles were removed by moist swabbing.

The resulting numbers and types of particles were determined using forensic microscopy. Particle profiles from the different fractions were compared to test the ability to objectively distinguish the order of exposure to the three environments.

### A. Site Selection

A range of candidate sites were selected in Virginia based on recorded differences in their surface geology, differences in the watershed (indicating different sources for sediments) and differences related to human activities (in the form of trail or roadside modifications). Candidate sites were also required to be conveniently assessable from roads and to have an apparently uniform path of at least 25 m (allowing the required 250 m walking exposures to be completed in 5 round trips).

A total of 37 sites were evaluated. Accessibility and suitability narrowed the candidates to 26. Soil samples from these sites were analyzed by polarized light microscopy, testing for characteristic particle profiles that would be readily distinguishable in single-specimen mounts of fine sand and silt fractions. Based on these assessments, three were selected based on their distinguishability and ease of access. These were: 1. Piney River (PR), 2. Appalachian Trail (AT) and 3. Luck Stone Quarry (LQ). The locations and appearance of these sites are shown in Figures 1 through 6.

Detailed sampling of each of these sites was conducted along a well-specified 25 m route. A combined specimen of surface particles was collected, taking pinches every two steps during two transects of each of the routes. These specimens were processed and analyzed as described below (see D. Specimen Processing and Polarized Light Microscopy), resulting in identifications and characterizations of mineral varieties, together with particle type number percentages as determined by point counting on 300+ mineral grains (see Data Set DS-1 Point Counts for Atlas Samples). An example of the data is given in Table 1. An atlas of mineral grain types was made for each of the sites (Data Sets DS-2 through DS-4). Figure 7 gives an example of one of the mineral grain type atlas entries.

Table 1. Example of Mineral Grain Data using Point Counting and Polarized Light Microscopy.

## Point Counts for Piney River Site Reference Specimen

<b>Grain type</b>	<b>Count</b>	<b>Percent</b>
Lithic fragments - other	73	23.5
Lithic fragments - mixed feldspars	60	19.4
Alkali feldspar - fresh	32	10.3
Alkali feldspar - weathered	11	3.5
Alkali feldspar - iron oxide	0	0.0
Alkali feldspar - inclusions	0	0.0
Epidote	36	11.6
Quartz - clear	15	4.8
Quartz - with inclusions	7	2.3
Quartz - with bubbles	0	0.0
Quartz - with iron oxides	0	0.0
Plagioclase	13	4.2
Muscovite	13	4.2
Biotite - brown	0	0.0
Biotite - yellow	8	2.6
Biotite - orange	5	1.6
Biotite - green	0	0.0
Hornblende	10	3.2
High index polycrystalline	9	2.9
Iron oxides	5	1.6
Pyroxene	4	1.3
Alterite	3	1.0
Garnet	3	1.0
Opaques	2	0.6
High index high birefringence	1	0.3
	310	100

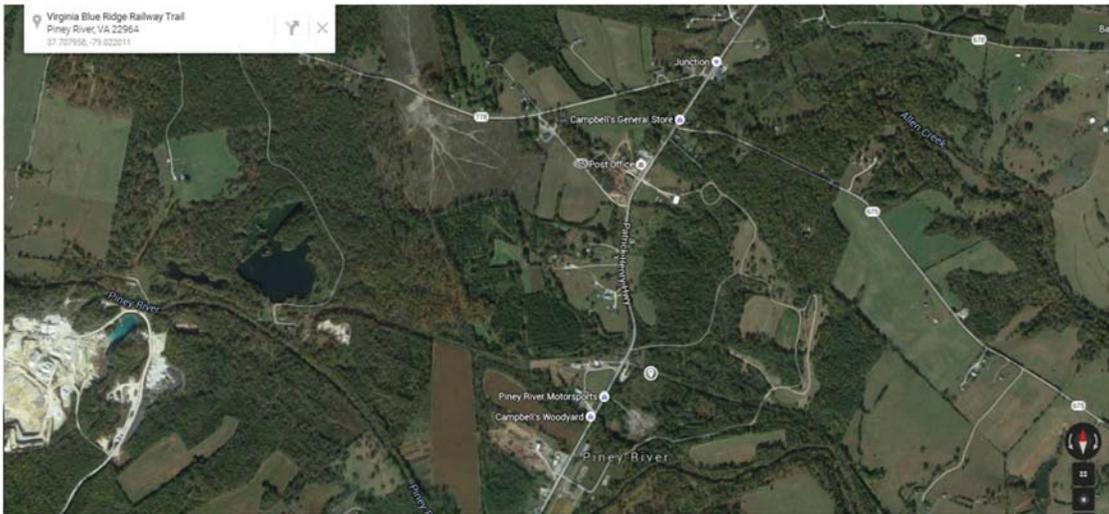


Figure 1. Overview of the location of the Piney River site, an improved hiking trail in a wooded site along the Virginia Blue Ridge Railway Trail (37.7078, -79.0220).

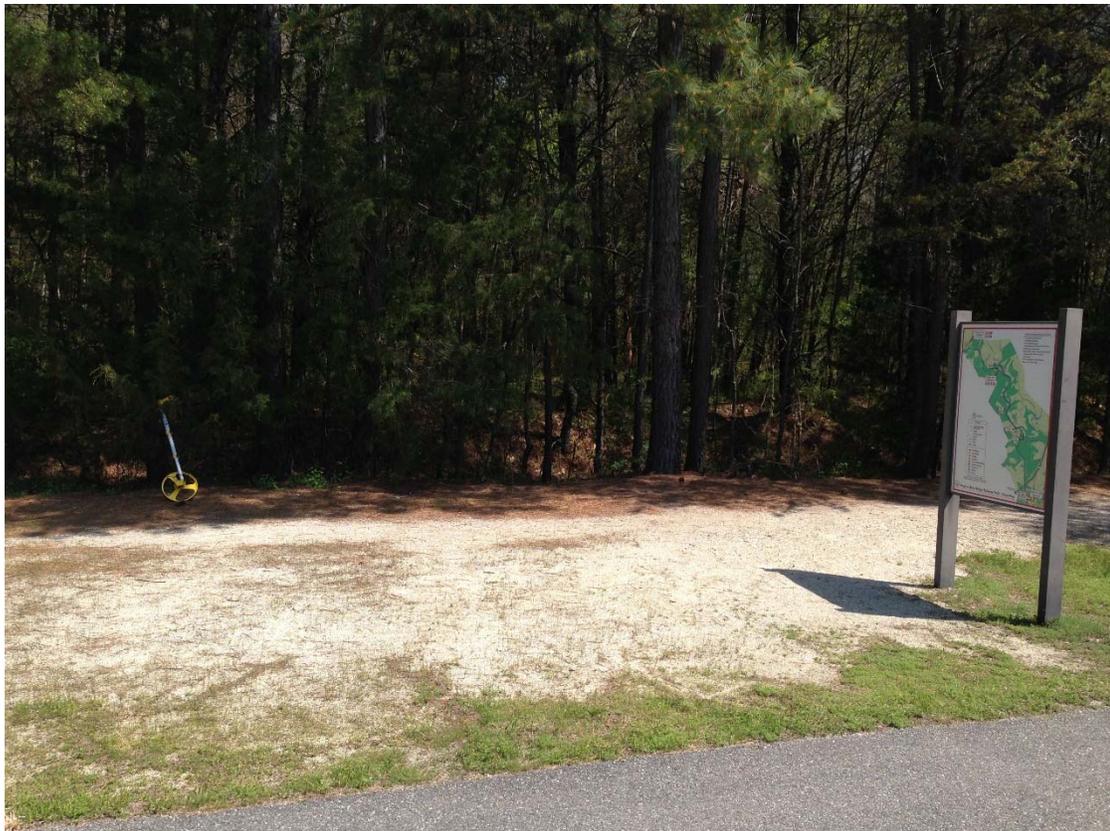


Figure 2. View of the Piney River trail site.



Figure 3. Overview of the location of the Appalachian Trail site, an unimproved minor trail just off of the main Appalachian Trail leading along the Tye River (37.8384, -79.0220).



Figure 4. View of the Appalachian Trail site.

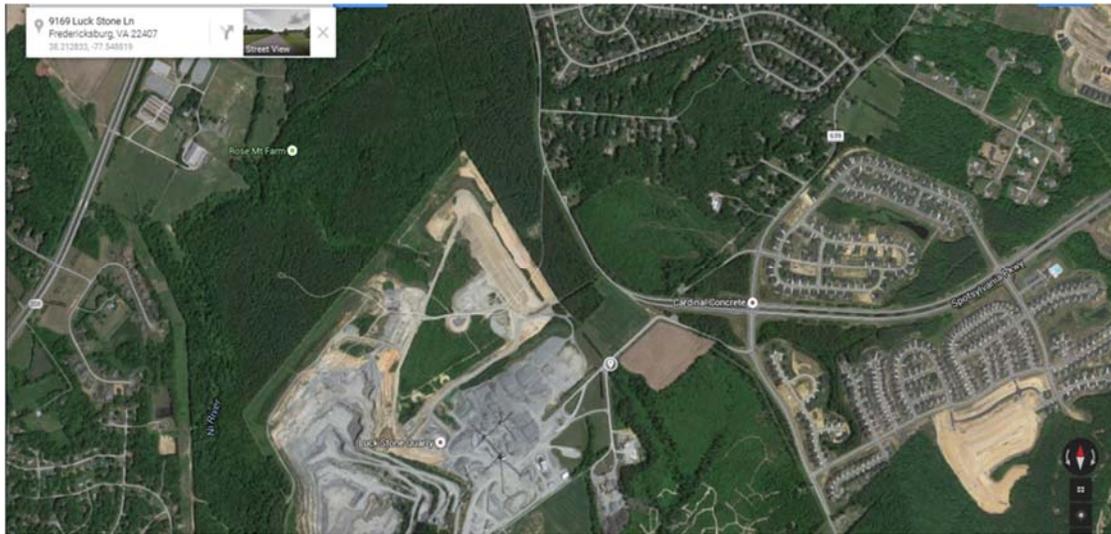


Figure 5. Overview of the location of the Luck Stone Quarry site, along the edges of a public access road outside of the Luck Stone Quarry in Fredericksburg, VA (38.2128, -77.5488).



Figure 6. View of the Luck Stone Quarry site.

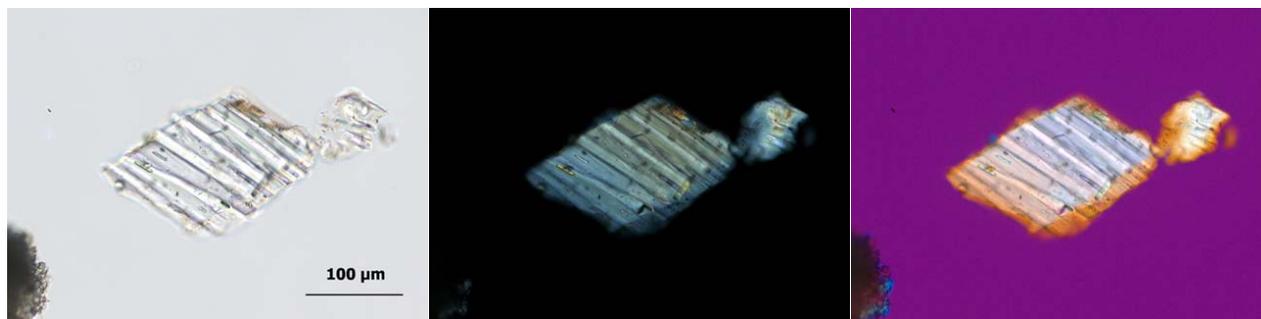


Figure 7. Example of an atlas entry documenting a mineral type found in one of the reference sites. “A lithic fragment composed of plagioclase and alkali feldspar, possibly formed due to exsolution. It is characterized by domains with higher refractive index than the mount (1.540), and domains with lower refractive index than the mount. It is typical of the mixed feldspar grains present in the fine sand of Site PR. Images are with transmitted plane polarized light (left), crossed polars (center) and crossed polars with Red I compensator (right).”

### B. Footwear Exposures

Two types of footwear were used: athletic shoes with flexible rubber soles and work boots with hard rubber soles. The chosen athletic shoes were Kirkland Signature™ Men’s Athletic Shoes (Figure 8, left) and the chosen work boots were Grabbers Black Steel Toe EH Non-Slip Work Boots (Figure 8, right). Exposures to test sites were made by walking a distance of 250 m along a route, achieved by ten transects of 25 m (five round trips). Eighteen pairs of each footwear type were exposed: 6 pairs for single-environment exposures (two duplicate pairs for each of the three test sites) and 12 pairs for sequential exposures to all three environments (2 duplicate pairs for each of the six alternative sequences).

Surfaces were raked to remove large plant matter prior to exposures. The surfaces were dry and dusty. Exposures were conducted over a five day rain-free period during which the surfaces remained dry. Following each exposure the footwear was gently re-packaged in its original box, between folds of butcher paper.



Figure 8. The Kirkland Signature™ Men’s Athletic Shoes (left) and Grabbers Steel Toe EH Non-Slip Work Boots (right) used in this study.

**C. Differential Sampling**

The more loosely adhering particles were removed by walking: 18 firm, smooth walking steps on butcher paper (92 kg individual, shoe size 10.5). Twelve steps were found to be sufficient to remove the most loosely held particles, as further steps recovered no perceptible additional particles. Particles were collected from the paper by moist swabbing with pre-filtered 3% ethanol (Figure 9).

Moderately adhering particles were removed using an electrostatic lifter (Sirche Electrostatic Dust Print Lifter Kit ESP900). Electrostatic lifting was conducted using a reverse procedure (foil side of lifting film down) and employing a piece of foil taped to the floor as a conductor.[41] With the full voltage setting, 6 smooth steps (92 kg individual, shoe size 10.5) were found to be sufficient to remove the moderately held particles, as further steps recovered no perceptible additional particles. Particles were collected from the electrostatic film by moist swabbing (Figure 10).

Direct moist swabbing of the contact surfaces of the footwear soles was conducted to remove and collect the most tightly held particles (Figure 11). Each of the three sampling methods resulted in particle suspensions in 3% ethanol within 1.5 mL microcentrifuge tubes. These were filled with an equal amount of pre-filtered ethanol to prevent the formation of mold. Table 2 (page 16) gives a summary of the program specimens resulting from the differential sampling. There are a total of 108 specimens, each with a six-character alphanumeric designation comprised as follows:

	1 <sup>st</sup> Position	2 <sup>nd</sup> Position	3 <sup>rd</sup> Position	4 <sup>th</sup> Position	5 <sup>th</sup> Position	6 <sup>th</sup> Position	
	Footwear Type	First Site Exposure	Second Site Exposure	Third Site Exposure	Replicate Number	Sampling Method	
	T (Trainers) B (Boot)	A (Site AT); L (Site LQ); P (Site PR)			1 or 2	W, E or S	Overall Designation
Single Site Exposure Example	B	P	P	P	1	W	BPPP1W
Sequential Site Exposure Example	T	A	L	P	2	E	TALP2E



Figure 9. Removal of the most loosely held particles by walking on paper.



Figure 10. Removal of moderately held particles using an electrostatic lifter.



Figure 11. Removal of the most tightly held particles by direct swabbing of contact surfaces of the sole.

Table 2. Summary of Program Specimens

	Single Site Exposures					
	Athletic Shoes			Work Boots		
	Site AT	Site LQ	Site PR	Site AT	Site LQ	Site PR
Walking	TAAA1W TAAA2W	TLLL1W TLLL2W	TPPP1W TPPP2W	BAAA1W BAAA2W	BLLL1W BLLL2W	BPPP1W BPPP2W
Electrostatic Lifter	TAAA1E TAAA2E	TLLL1E TLLL2E	TPPP1E TPPP2E	BAAA1E BAAA2E	BLLL1E BLLL2E	BPPP1E BPPP2E
Swabbing	TAAA1S TAAA2S	TLLL1S TLLL2S	TPPP1S TPPP2S	BAAA1S BAAA2S	BLLL1S BLLL2S	BPPP1S BPPP2S
	Sequential Site Exposures					
	Athletic Shoes					
	AT, LQ, PR	AT, PR, LQ	LQ, AT, PR	LQ, PR, AT	PR, AT, LQ	PR, LQ, AT
Walking	TALP1W TALP2W	TAPL1W TAPL2W	TLAP1W TLAP2W	TLPA1W TLPA2W	TPAL1W TPAL2W	TPLA1W TPLA2W
Electrostatic Lifter	TALP1E TALP2E	TAPL1E TAPL2E	TLAP1E TLAP2E	TLPA1E TLPA2E	TPAL1E TPAL2E	TPLA1E TPLA2E
Swabbing	TALP1S TALP2S	TAPL1S TAPL2S	TLAP1S TLAP2S	TLPA1S TLPA2S	TPAL1S TPAL2S	TPLA1S TPLA2S
	Sequential Site Exposures					
	Work Boots					
	AT, LQ, PR	AT, PR, LQ	LQ, AT, PR	LQ, PR, AT	PR, AT, LQ	PR, LQ, AT
Walking	BALP1W BALP2W	BAPL1W BAPL2W	BLAP1W BLAP2W	BLPA1W BLPA2W	BPAL1W BPAL2W	BPLA1W BPLA2W
Electrostatic Lifter	BALP1E BALP2E	BAPL1E BAPL2E	BLAP1E BLAP2E	BLPA1E BLPA2E	BPAL1E BPAL2E	BPLA1E BPLA2E
Swabbing	BALP1S BALP2S	BAPL1S BAPL2S	BLAP1S BLAP2S	BLPA1S BLPA2S	BPAL1S BPAL2S	BPLA1S BPLA2S

#### D. Specimen Processing

Distilled water was added to the particle suspensions to a height of three cm in the 1.5 mL microcentrifuge tubes, followed by mixing using a vortex mixer (Thermolyne Type 16700 Mixer) with occasional brief sonication (Fisher Scientific FS6 Ultrasonic Cleaner). After mixing, samples were allowed to settle in the water for approximately 9 seconds (3 seconds per cm of water height) allowing all particles with a density greater than 2.61 and diameter greater than 62.5  $\mu\text{m}$  to settle.[42] After settling, the liquid and portion of sample still suspended were removed using a pipette and not examined further. The size cut-off of 62.5  $\mu\text{m}$  was chosen because that is the lower limit of the sand-sized fraction, as defined by the Wentworth scale commonly used by sedimentary geologists.[43] The density value of 2.61 was selected because it is close to the density of quartz. Due to its abundance in igneous, sedimentary and metamorphic rocks, quartz is the most common detrital mineral in sediments on the Earth's surface.[44] Quartz is also abundant in the three sites selected for this study.

The cleaned sand-sized fraction was wet-sieved, dividing the sample into two fractions, that greater 180  $\mu\text{m}$  (the coarse sand fraction) and that less than 180  $\mu\text{m}$  (the fine sand fraction). Wet sieving was performed using an Endecotts, Ltd. brand stainless steel sieve with 180  $\mu\text{m}$  openings. The fraction passing the sieve was washed into a glass petri dish and recovered by pipette (Figure 12). The coarse sand fraction was not examined further. This size selection was performed for convenience, as particles larger than about 180  $\mu\text{m}$  are more difficult to mount under a coverslip and to examine at high magnification using light microscopy.

Where practical, the entire fine sand fraction was transferred onto a microscope slide by direct pipetting and allowing it to air dry (Figure 13). Where the large sample size made it impractical to mount the entire fraction, a representative subsample of the fine sand fraction was mounted for analysis by polarized light microscopy using the method of McVicar and Graves.[45] The entire fraction was transferred to a crucible, moistened with distilled water, and allowed to partially dry. When the excess water had evaporated, but the bulk powder still adhered together and moved coherently, a small portion of the moist sand was removed with a wooden toothpick and transferred to a clean glass microscope slide, where it was allowed to dry completely (Figure 14). After drying the specimen was mounted under a coverslip using Cargille mounting medium (Series A) 1.540<sub>nD</sub>, 25°C.



Figure 12. The sieve and petri dish used for isolating the fine sand fraction.

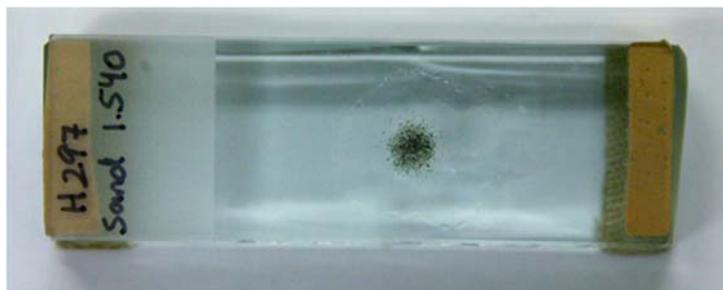


Figure 13. Example of an entire fine sand fraction drying on a microscope slide.

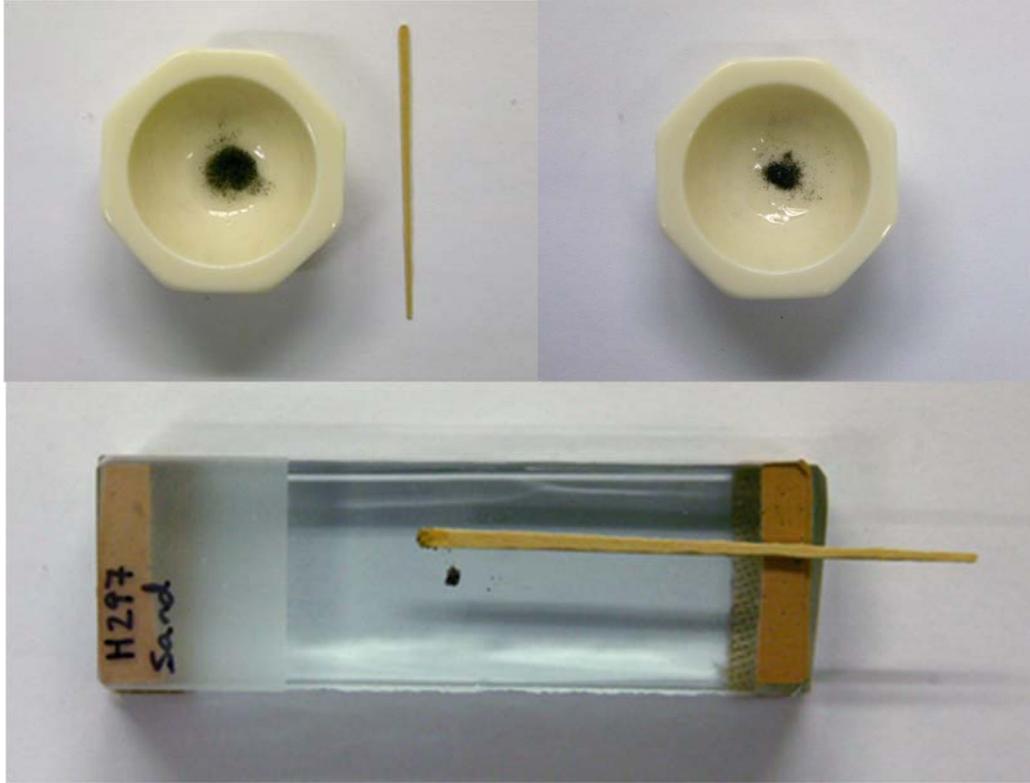


Figure 14. The crucible method used for subsampling large sand fractions.[45] A large sand fraction after being transferred to the crucible (upper left), and again after the liquid evaporated sufficiently to make the grains adhere to each other (upper right). A subsample is then recovered from the crucible using a wooden toothpick and transferred to a glass slide to dry (below).

#### E. Polarized Light Microscopy

Polarized light microscopy was performed using a Leitz DMRP polarized light microscope and a Leica DMLP polarized light microscope, at 200 times magnification. Photomicrographs were taken with a Zeiss AxioCam MRcS camera mounted on a Zeiss AxioImager A1m polarized light microscope. Optical identification and characterization of minerals was based on comparisons to known samples and reference data [46-48] and as detailed later in this section. Variations in morphological and optical features were determined as described by Bowen.[49] Categorization of some mineral grains, as viewed in single mounts, required (as a practical matter under examination conditions) a subjective judgement with respect to placing certain grains into a category. The number of grains that requiring subjective placement into a category was small (less than a few percent) for the samples in this study.

Quantitative determinations were made by point counting of particles using the ribbon method.[50] The microscope slide is moved horizontally across the stage and every grain encountered in a horizontal strip, or “ribbon”, is counted. The upper and lower limits of the field of view were used to define the ribbon. Each grain counted was characterized optically and morphologically and placed into a category. Classifications of grain types were made based on the mineral grain characterization criteria detailed below.

## F. Mineral Grain Classification Criteria

*Quartz.* Grains that exhibited low birefringence, the absence of twinning, the absence of extensive weathering, having one refractive index very close to 1.540 and another somewhat higher were classified as quartz. The index close to 1.540 ( $\omega = 1.544$ ) exhibits characteristic blue and yellow-orange dispersion colors for quartz; these dispersion colors must be observed in order to classify a grain as quartz, although sometimes they are subtle on thin edges.

*Subclasses of Quartz.* Quartz grains were classified as being “clear” when they lacked conspicuous solid inclusions, fluid inclusions, or iron oxide coatings. Quartz grains were classified as having “inclusions” when they contained conspicuous solid inclusions. Quartz grains were classified as having “bubbles” when they contained very dark, rounded inclusions with light centers, consistent with fluid inclusions. Quartz grains were classified as having “iron oxide coatings” when they had conspicuous reddish, orange, or brown surface staining.

*Alkali Feldspar.* Grains that exhibited low birefringence and have all of their refractive indices significantly lower than 1.540 were classified as alkali feldspar. Alkali feldspar grains commonly exhibit twinning. Refractive indices are considered to be significantly lower than 1.540 when there are no dispersion colors apparent at the edges of the grain in any orientation. This is because colored Becke lines become visible in a 1.540 liquid for grains with refractive index values higher than about 1.536,[51] and the vast majority of alkali feldspar compositions have refractive index  $\gamma$  values lower than 1.536 (only those with compositions very close to the albite end member have  $\gamma$  values equal to 1.536.[46])

*Subclasses of Alkali Feldspar.* Alkali feldspar grains were classified as being “fresh” when they lacked conspicuous surface weathering, inclusions, iron oxide coatings, or cross-hatched twinning patterns. Alkali feldspar grains were classified as being “weathered” when they had significant surface discoloration that was not orange, reddish, or brown in color, or when they exhibited light scattering due to rough surface textures. Alkali feldspar grains were classified as having “iron oxide coatings” when they had conspicuous reddish, orange, or brown surface staining. Alkali feldspar grains were classified as having “inclusions” when they contained conspicuous solid inclusions. Alkali feldspar grains were classified as “microcline” when they exhibited cross-hatched twinning patterns.

*Plagioclase Feldspar.* Grains that exhibited low birefringence and have refractive index values slightly lower than and/or greater than 1.540 were classified as plagioclase feldspar if they met additional criteria. Grains that have both refractive index values lower than 1.540 with at least one index close enough to produce dispersion colors were classified as plagioclase. Grains with one index lower than 1.540 and the other index greater than 1.540 were classified as plagioclase. Grains with both refractive index values greater than 1.540 that did not exhibit the characteristic dispersion colors of quartz were classified as plagioclase feldspar. Finally, grains that resemble quartz but exhibit twinning and/or extensive weathering were classified as plagioclase feldspar.

*Hornblende.* Grains that exhibited moderate birefringence, high refractive indices, pleochroism in shades of greens and browns with deepest color parallel to their length, and positive elongation were classified as hornblende.

*Biotite.* Thin flat flakes that exhibited high refractive indices and yellow, orange, brown, or green colors were classified as biotite. Biotite flakes on edge may appear as elongated crystals exhibiting strong pleochroism.

*Subclasses of Biotite.* Biotite flakes were further classified as being one of the following four types based on their absorption color in plane polarized light: “brown”, “green”, “yellow”, or “orange”. For flakes on edge that exhibited pleochroism, the darkest color visible was used for classification.

*Unidentified Highly Altered Grains.* Grains that were too highly altered to clearly observe Becke line behavior and/or birefringence, to the point that no classification of the grain into another category was possible, were classified as unidentified highly altered grains (alterite).

*Epidote.* Grains that exhibited high refractive indices, moderate birefringence, and anomalous retardation colors (first order gray is noticeably blue, and with a full wave compensator and the grain oriented to produce subtraction, bands that should be black due to complete compensation retain a purplish hue), sometimes with a pale yellow or green absorption color, were classified as epidote.

*Titanite.* Grains that exhibited very high refractive indices and very high birefringence, were classified as titanite. Titanite grains often exhibit high dispersion (observable in an interference figure or as dispersed extinction).

*Lithic Fragments.* Individual grains that had more than one mineral type making up significant portions of the grain were classified as lithic fragments.

*Subclasses of Lithic Fragments.* Lithic fragments that were composed primarily of two or more feldspars with different compositions, exhibiting different contrast and/or Becke line behavior, were classified as “mixed feldspars.” Lithic fragments that were composed of either quartz or feldspar along with numerous small high index crystals were classified as “same as mixed feldspars” due to their occurrence at the PR site together with the mixed feldspar lithic fragments. Lithic fragments that did not fall into one of the other two categories were classified as “other” lithic fragments.

*Black Opaques.* Grains that were opaque and had a black hue in oblique top lighting were classified as black opaque grains.

*Iron Oxides.* Grains exhibiting characteristic red, orange, or reddish-brown hues, isotropic or with low-moderate birefringence, exhibiting high refractive indices, were classified as iron oxides.

*Pyroxene.* Grains were classified as pyroxene when they exhibited high refractive indices, moderate birefringence, positive elongation, and none of the following: characteristic zircon morphology, anomalous retardation colors typical of epidote, or high dispersion. Pyroxene grains can exhibit a variety of pale colors, subtle pleochroism, and weathering.

*Amphibole.* Grains were classified as amphibole when they were elongated and exhibited high refractive indices, cleavage parallel to their length, moderate birefringence, positive elongation, and none of the following: characteristic zircon morphology, anomalous retardation colors of epidote, or high dispersion.

*Muscovite.* Thin, colorless flakes that exhibited moderately high refractive indices along with biaxial negative interference figures with a small optic axial angle were classified as muscovite. Muscovite flakes on edge may appear as thin, elongated crystals with high birefringence.

*Apatite.* Grains that exhibited moderately high refractive indices, subhedral and/or rounded hexagonal prisms, smf s negative optic sign, were classified as apatite.

*Zircon.* Grains that exhibited very high refractive indices, moderate birefringence, subhedral and/or rounded tetragonal prisms terminated by pyramids, a positive optic sign, and the absence of either anomalous retardation colors or high dispersion, were classified as zircon.

*Rutile.* Golden, yellow, brown or reddish grains that exhibited very high refractive indices, very high birefringence, subhedral and/or rounded tetragonal prisms terminated by pyramids, and subtle pleochroism, were classified as rutile.

*Garnet.* Isotropic grains that exhibited high refractive indices were classified as garnet.

*Carbonates.* Grains that exhibited very high birefringence with one or both refractive indices greater than 1.540, and that were uniaxial with a negative optic sign were classified as carbonates. Rhombohedral morphology is also common among carbonates.

*Tourmaline.* Grains that exhibited moderate birefringence, high refractive indices, strong pleochroism with the deepest color parallel to their width, and negative elongation were classified as tourmaline.

*High Index Clear/Prism.* Grains consisting of colorless single crystals that exhibited low to moderate birefringence, high refractive indices, and that lacked characteristic features that would place them in another category, were classified as high index clear/prism grains. A number of the grains in this group had a prismatic habit.

*High Index Polycrystalline.* Grains having high refractive indices that were composed of multiple smaller crystals that all appeared to be a single mineral type, and that lacked characteristic features that would place them in another category, were classified as high index polycrystalline grains.

*Yellow/Orange Rounded.* Rounded grains exhibiting a yellow to orange absorption color and low birefringence with refractive indices relatively close to 1.540, but lacking the characteristic features of biotite, were classified as yellow/orange rounded grains.

*Yellow Striated.* Yellow polycrystalline grains exhibiting cleavage striations were classified as yellow striated grains.

**III. Results**

**A. Raw Point Count Data and Data Reduction**

Point count data for the 108 program specimens (see Table 2) can be found in Data Sets DS-5 (single site exposures), DS-6 (athletic shoe sequential exposures) and DS-7 (boots sequential exposures). In these Excel files there is an individual worksheet for each specimen, with worksheet names corresponding to the specimen designations in Table 2. The raw data, with point counts of all mineral varieties (e.g., Table 1) appears in columns B through D. Counts after consolidation of mineral varieties are given in columns G through I. Of these consolidated groups, thirteen occurred above 2% in one or more of the single-site exposures. Point count data for all specimens were grouped into these 13 categories with a 14<sup>th</sup> category of “other.” These groupings appear in columns K and L of the spreadsheets. An example of the raw data, the consolidation of mineral varieties and the grouping into 14 categories is given in Table 3.

Table 3. Example of Raw Data, Consolidation of Mineral Varieties and Grouping to the 14 Most Abundant Types. These data are for Athletic Shoe with Sequential Exposure AT, PR, LQ, replicate 1. Data for each specimen in this study are given in the submitted Data Sets.

Point Counts with Varieties			Point Counts without Varieties			Grouped into 14 Categories		
Grain type	Count	%	Grain type	Count	%	Grain type	Count	%
Quartz - clear	125	34.0	Quartz	147	39.9	Alkali feldspar	30	8.2
Quartz - with inclusions	8	2.2	Hornblende	71	19.3	Alterite	12	3.3
Quartz - with bubbles	10	2.7	Alkali feldspar	30	8.2	Biotite	10	2.7
Quartz - with iron coatings	4	1.1	Plagioclase	66	17.9	Epidote	14	3.8
Hornblende	71	19.3	Biotite	10	2.7	High index	0	0.0
Alkali feldspar - fresh	19	5.2	Alterite	12	3.3	Hornblende	71	19.3
Alkali feldspar - weathered	6	1.6	Epidote	14	3.8	Iron oxides	0	0.0
Alkali feldspar - inclusions	1	0.3	Titanite	4	1.1	Lithic Fragments	2	0.5
Alkali feldspar - microcline	2	0.5	Lithic fragments	2	0.5	Muscovite	1	0.3
Alkali feldspar - with iron coatings	2	0.5	Opauques	6	1.6	Opauques	6	1.6
Plagioclase	66	17.9	Iron oxides	0	0.0	Plagioclase	66	17.9
Biotite - brown	7	1.9	Pyroxene	0	0.0	Quartz	147	39.9
Biotite - green	3	0.8	Amphibole - colorless	0	0.0	Titanite	4	1.1
Biotite - yellow	0	0.0	Muscovite	1	0.3	Other	5	1.4
Biotite - orange	0	0.0	Apatite	1	0.3		368	100.0
Alterite	12	3.3	Carbonates	4	1.1			
Epidote	14	3.8	Zircon	0	0.0			
Titanite	4	1.1	High index clear/prism	0	0.0			
Lithic fragments	0	0.0	High index polycrystalline	0	0.0			
Lithic fragments - mixed feldspars	0	0.0	Garnet	0	0.0			
Lithic fragments - other	2	0.5	Yellow striated	0	0.0			
Opauques	6	1.6	Rutile	0	0.0			
Iron oxides	0	0.0						
Pyroxene	0	0.0			368	100		
Amphibole - colorless	0	0.0						
Muscovite	1	0.3						
Apatite	1	0.3						
Carbonates	4	1.1						
Zircon	0	0.0						
High index clear/prism	0	0.0						
High index polycrystalline	0	0.0						
Garnet	0	0.0						
Yellow striated	0	0.0						
Rutile	0	0.0						
	368	100						

B. Retention of Particles on Athletic Shoe vs. Work Boot Soles

Data relating to the effect of the alternative footwear types is illustrated in Table 4 which gives the overall mean fractions of mineral categories for single site exposures. In this table (and several others that follow) color coding of cell values, as indicated on the key below the table, has been used to facilitate comparisons among columns of data. When comparing the results for athletic shoes and work boots exposed to the same sites, the recovered mineral fractions are very close and there is no evidence that the alternative sole types result in different accumulations or the different mineral types. Pearson correlations of 0.995, 0.989 and 0.998 were observed for the three sites, and paired two-sample t-tests (hypothesized difference of means = 0) resulted in P(t<=t) of nearly 1. Table 4 also shows that the three sites are clearly distinguished by their mineral classification fractions.

Table 4. Overall Fractions Mineral Categories for Single-Site Exposures Contrasting Results for Work Boots and Athletic Shoes.

	SITE AT		SITE LQ		SITE PR	
	BOOTS	SHOES	BOOTS	SHOES	BOOTS	SHOES
Alkali feldspar	0.556	0.561	0.085	0.076	0.115	0.115
Alterite	0.065	0.046	0.025	0.015	0.027	0.017
Biotite	0.003	0.006	0.056	0.040	0.050	0.049
Epidote	0.019	0.019	0.031	0.026	0.139	0.138
High index	0.007	0.009	0.002	0.004	0.052	0.053
Hornblende	0.005	0.008	0.219	0.200	0.019	0.015
Iron oxides	0.020	0.015	0.001	0.000	0.004	0.010
Lithic Fragments	0.008	0.013	0.016	0.015	0.364	0.340
Muscovite	0.002	0.002	0.006	0.004	0.069	0.083
Opagues	0.064	0.047	0.015	0.014	0.002	0.002
Plagioclase	0.013	0.009	0.068	0.129	0.083	0.081
Quartz	0.217	0.228	0.452	0.453	0.046	0.069
Titanite	0.001	0.000	0.010	0.011	0.015	0.010
Other	0.021	0.037	0.012	0.013	0.015	0.018
Total	1.000	1.000	1.000	1.000	1.000	1.000

>0.25	0.10-0.25	0.05-0.10	0.02-0.05	<0.02
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### C. Differential Sampling of Sequential Site Exposures

An example of results from differential sampling following sequential site exposures is given in Table 5 for work boots exposed to the test sites in the sequence AT, PR, LQ. Data are given for raw particle counts and mineral classification fractions for each of the three differential samplings (walking, electrostatic lifting, and swabbing) for each of the two duplicate sequential exposures (walking 1, walking 2, etc.). The proportions of mineral classifications for the three differential samplings are highly similar to one another with each corresponding to the proportions seen in site LQ: the last of the sites to which the boots were exposed. This observation was consistent across each of the six exposure sequences, for both footwear types.

For each of the 72 mineral classification profiles (2 footwear types X 6 exposure sequences X 3 sample differential samplings X 2 replicates) a measure of distance from each of the test sites was calculated using the chi-square statistic. Table 6 shows the calculation for the electrostatic lifter sampling from one of the boots exposed to the site sequence AT, PR, LQ. Corresponding tables for each of the specimens can be found in Data Sets DS-5 (single site exposures), DS-6 (athletic shoe sequential exposures) and DS-7 (boots sequential exposures) in columns N through AA of each spreadsheet.

Percentages of the total distances for each of the shoe samples are shown on a ternary diagram in Figure 15 following the method of Graham and Midgely.[52] Figure 16 shows the corresponding diagram for the boot samples. These diagrams illustrate the distance of each of the shoe samples from each of the test sites. The three sides of the triangle correspond to the three test sites (LQ, PR and AT) and the black circles on the diagrams are the mean observed values for the single site exposures. The colored triangular points represent the calculations of the chi square distance for each of the differential samples recovered from the footwear soles, from each of the test sites. Points close to only one of the sites align along the edge corresponding to that site. Points close to more than one site would appear in the central portion of the chart. The legend shows the colors corresponding to each of the six sequences of test site exposures. One point is plotted for each of the samples recovered from footwear, with six points for each exposure sequence (from each of two replicates and the three differential samplings). In Figure 15 (for athletic shoes) all of the samples, whether representing loosely, moderately or tightly held particles, align on the axis edge corresponding to the last of the sites to which the shoes were exposed. This means that the samples are uniformly dominated by the third site in the sequential footwear exposures. Likewise, in Figure 16 (for work boots) nearly all samples align on the axis edge corresponding to the last of the sites to which the boots were exposed. The one LQ, PR, AT sample that departs noticeably from the AT boarder is from a swabbing specimen, recovering the most tightly held particles, and is still primarily composed of particles attributable to the last exposure site.

Table 5. Raw Counts, Fractions and Comparisons of Mineral Classifications for Boots Exposed to Test Sites in the Sequence AT, PR, LQ.

	RAW COUNTS						FRACTIONS						MEAN	COMPARISON		
	WALKING 1	WALKING 2	LIFTER 1	LIFTER 2	SWABBING 1	SWABBING 2	WALKING 1	WALKING 2	LIFTER 1	LIFTER 2	SWABBING 1	SWABBING 2	BOOTS AT-PR-LQ	LQ - BOOTS	AT - BOOTS	PR - BOOTS
Alkali feldspar	39	24	21	20	16	25	0.118	0.074	0.059	0.059	0.046	0.070	0.071	0.085	0.556	0.115
Alterite	8	8	8	1	11	2	0.024	0.025	0.022	0.003	0.031	0.006	0.019	0.025	0.065	0.027
Biotite	10	21	40	11	17	14	0.030	0.065	0.112	0.033	0.048	0.039	0.055	0.056	0.003	0.050
Epidote	13	4	7	11	13	20	0.039	0.012	0.020	0.033	0.037	0.056	0.033	0.031	0.019	0.139
High index	0	0	0	3	1	1	0.000	0.000	0.000	0.009	0.003	0.003	0.002	0.002	0.007	0.052
Hornblende	71	55	85	67	83	87	0.215	0.170	0.237	0.199	0.236	0.242	0.217	0.219	0.005	0.019
Iron oxides	0	0	0	0	0	1	0.000	0.000	0.000	0.000	0.000	0.003	0.000	0.001	0.020	0.004
Lithic Fragments	4	12	3	12	3	6	0.012	0.037	0.008	0.036	0.009	0.017	0.020	0.016	0.008	0.364
Muscovite	0	3	7	3	0	4	0.000	0.009	0.020	0.009	0.000	0.011	0.008	0.006	0.002	0.069
Opagues	4	1	9	4	4	7	0.012	0.003	0.025	0.012	0.011	0.019	0.014	0.015	0.064	0.002
Plagioclase	28	26	30	35	28	35	0.085	0.080	0.084	0.104	0.080	0.097	0.088	0.068	0.013	0.083
Quartz	147	165	140	162	171	147	0.445	0.511	0.391	0.481	0.487	0.409	0.454	0.452	0.217	0.046
Titanite	2	2	4	2	0	3	0.006	0.006	0.011	0.006	0.000	0.008	0.006	0.010	0.001	0.015
Other	4	2	4	6	4	7	0.012	0.006	0.011	0.018	0.011	0.019	0.013	0.012	0.021	0.015
Total	330	323	358	337	351	359	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000

>0.25
0.10-0.25
0.05-0.10
0.02-0.05
<0.02

Table 6. Example of Chi-Square Distance Calculation Using Electrostatic Lift Sampling from Boot #1 Exposed to the Site Sequence AT, PR, LQ. Tables for each specimen in this study are given in the submitted Data Sets.

	Raw Count	Distance from Site AT				Distance from Site LQ				Distance from Site PR			
		Mean AT	Observed	Expected	(O-E)**2/E	Mean LQ	Observed	Expected	(O-E)**2/E	Mean PR	Observed	Expected	(O-E)**2/E
Alkali feldspar	21	0.556	21	199.1	159.36	0.085	21	30.4	2.91	0.115	21	41.2	9.91
Alterite	8	0.065	8	23.4	10.11	0.025	8	8.9	0.09	0.027	8	9.8	0.32
Biotite	40	*				0.056	40	20.1	19.74	0.050	40	17.8	27.66
Epidote	7	0.019	7	6.8	0.01	0.031	7	11.0	1.45	0.139	7	49.9	36.88
High index	0	*				*				0.052	0	18.6	18.61
Hornblende	85	*				0.219	85	78.5	0.54	0.019	85	6.8	900.96
Iron oxides	0	0.020	0	7.1	7.15	*				*			
Lithic Fragments	3	*				0.016	3	5.8	1.36	0.364	3	130.3	124.37
Muscovite	7	*				*				0.069	7	24.5	12.53
Opagues	9	0.064	9	23.0	8.50	0.015	9	5.3	2.50	*			
Plagioclase	30	*				0.068	30	24.4	1.30	0.083	30	29.8	0.00
Quartz	140	0.217	140	77.8	49.78	0.452	140	161.9	2.97	0.046	140	16.4	930.31
Titanite	4	*				*				0.015	4	5.5	0.39
Other	4	0.058	173	20.8	1111.92	0.033	15	11.7	0.92	0.021	13	7.4	4.21
Total	358	1.000	358	358.0	1346.83	1.000	358	358.0	33.78	1.000	358	358.0	2066.16

\* Mineral classifications where expected counts fell below 5.0 are included in the "Other" classification.

Figure 15. Ternary diagram illustrating the distance of each of the shoe samples from each of the test sites. The three sides of the triangle correspond to the three test sites (LQ, PR and AT) and the black circles are the mean observed values for the single site exposures. The colored triangular points represent the calculation of the chi square distance of each of the shoe samples from each of the test sites. Points close to only one of the sites align along the edge corresponding to that site. Points close to more than one site would appear in the central portion of the chart. The legend shows the colors corresponding to each of the six sequences of test site exposures. One point is plotted for each of the samples recovered from shoes, with six points for each sequence (from each of two shoes samples recovered by walking, electrostatic lifting and swabbing). All of the samples, whether representing loosely, moderately or tightly held particles, align on the axis edge corresponding to the last of the sites to which the shoes were exposed. This means that the samples are uniformly dominated by the third site in the sequential footwear exposures.

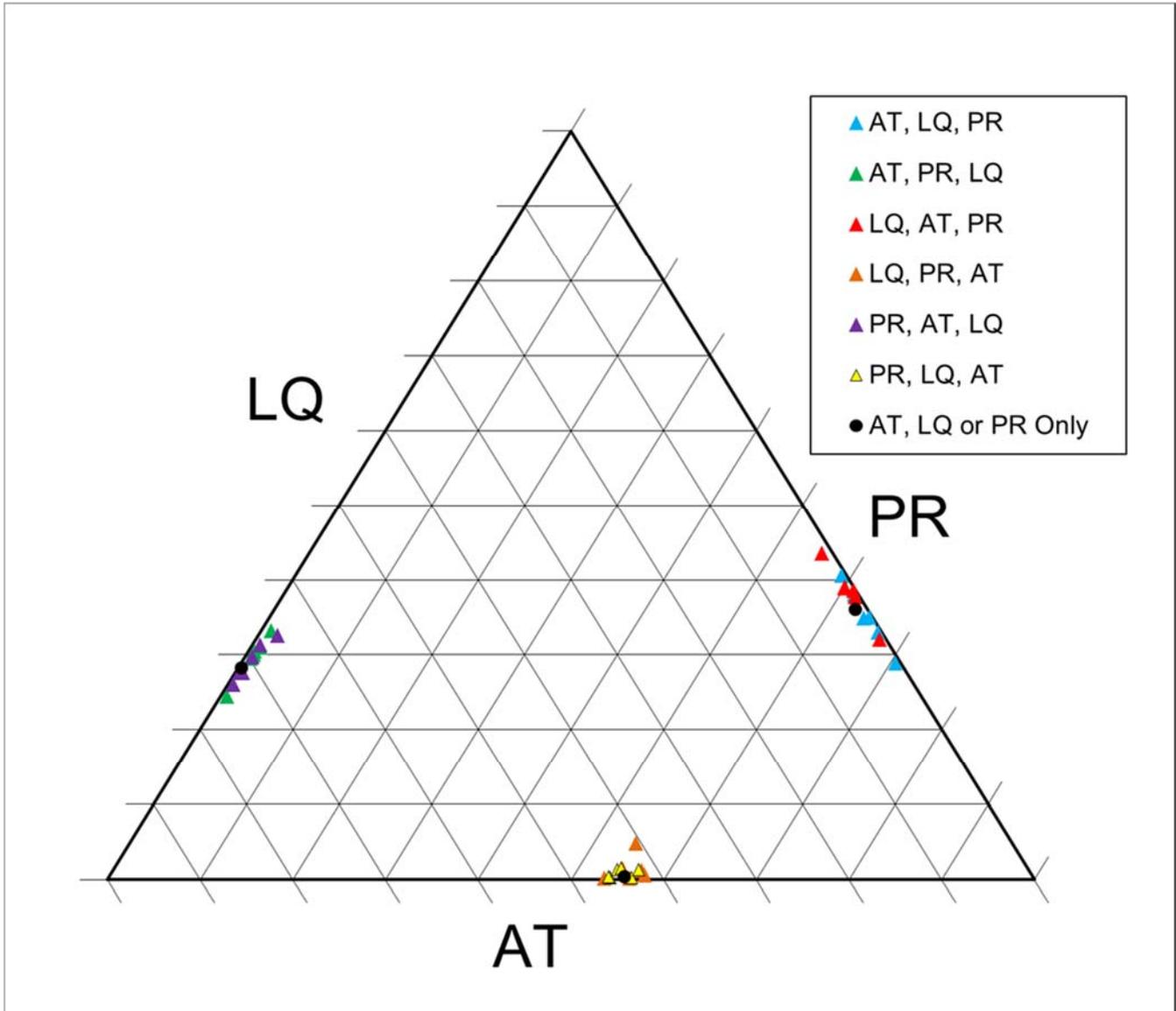
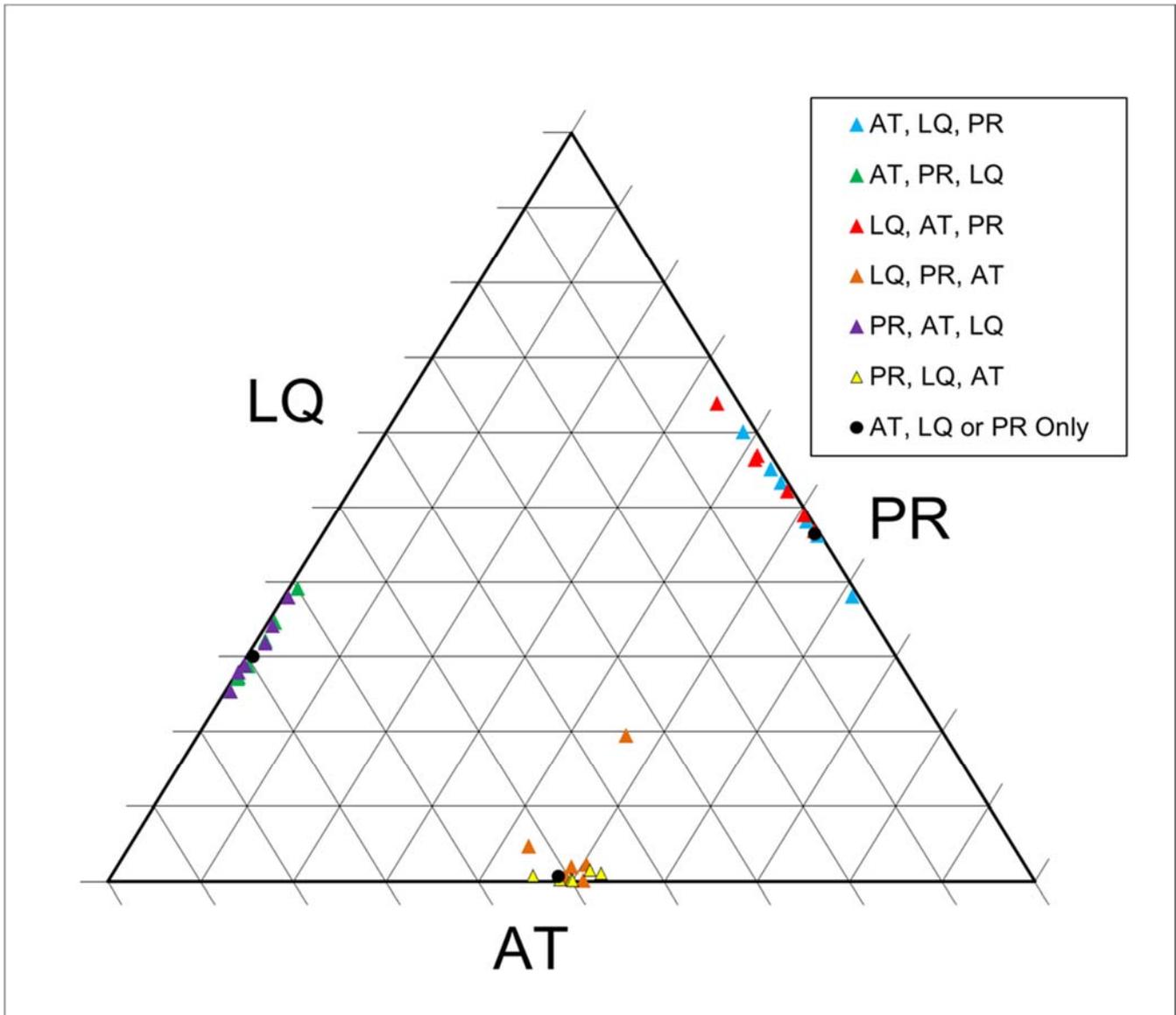


Figure 16. Ternary diagram illustrating the distance of each of the boots samples from each of the test sites. The three sides of the triangle correspond to the three test sites (LQ, PR and AT) and the black circles are the mean observed values for the single site exposures. The colored triangular points represent the calculation of the chi square distance of each of the boot samples from each of the test sites. Points close to only one of the sites align along the edge corresponding to that site. Points close to more than one site would appear in the central portion of the chart. The legend shows the colors corresponding to each of the six sequences of test site exposures. One point is plotted for each of the samples recovered from boots, with six points for each sequence (from each of two boots, samples recovered by walking, electrostatic lifting and swabbing). Nearly all samples, whether representing loosely, moderately or tightly held particles, align on the axis edge corresponding to the last of the sites to which the boots were exposed. This means that the samples are uniformly dominated by the third site in the sequential footwear exposures. The one LQ, PR, AT sample that is noticeably in the central area is from a swabbing specimen (recovering the most tightly held particles).



#### IV. Conclusions

Without exception, the samples resulting from differential sampling are dominated by the third site in the sequential footwear exposures. No noticeable differences are seen among the differential samplings of the loosely, moderately and strongly held particles: the same overwhelming presence of the third site is seen.

It is clear from these results that the third (final) exposure results in the nearly complete removal of any particles that were transferred to the contact surfaces of the shoe from the first and second exposures. This occurs regardless of the exposure sequence and regardless of which specific site was used for the third exposure.

It is also clear that under the experimental conditions loosely, moderately and strongly held particles are affected similarly, without any detectable enrichment of the earlier exposures among the more tightly held particles.

The hypothesis guiding this work was that distinguishable fractions would result from the differential sampling of the contact surface of footwear. That is, that by separating loosely held, moderately held, and strongly held particle fractions we would recover enriched particle fractions originating from different exposures, enabling the recognition and specification of sequential footwear exposures. This hypothesis is rejected. Under the experimental conditions the contact surface of footwear was found to be overwhelmingly dominated by the most recent exposure.

This finding is significant in that it fails to follow prior research focused on the persistence of trace evidence generally,[9-13] and on footwear specifically,[4,14] that strongly supports the hypothesis that, after transfer to an item, some particles are tightly held (and retained longer), while others are loosely held (and more rapidly lost). Contact surfaces of footwear, under the experimental conditions, are clearly an exception: a walk of 250 m, on a dry soil surface, results in the virtually complete removal and replacement of particles adhering to the contact surfaces from prior, similar exposures.

Given that Morgan et al.[14] have shown in comparable studies that a generalized sampling of footwear soles (from both contact and recessed areas) shows the retention of particles from earlier contacts, the clear implication of the present research is that, although particles *on the contact surfaces* of footwear are removed and replaced, those that are present *on the more recessed areas* of the sole are not.

This research, based on realistic exposures to actual environments, and incorporating newly-developed differential sampling, has resulted in greater understanding of (1) how very small particles adhere to the soles of footwear, (2) how they can be separated, and (3) whether sequential exposures can be determined based on how tightly particles are held.

Prior work examining mixtures of particles on footwear had used test substances, simple sampling protocols, and single particle types to help determine the scope of the problem. This project used realistic field conditions, differential sampling protocols and combinations of co-occurring particle types to definitively test the adherence and transfer of very small particles adhering to the contact surfaces of footwear.

There are a number of important implications for criminal justice policy and practice in the United States.

*1. During successive exposures there is nearly complete, sequential displacement of particles from the contact surfaces of footwear.* This means that the contact surfaces of recovered footwear will have traces from the most recent areas where the footwear was worn. The implications of this finding are important. For example, in cases where a body is found and may have been transported after death from one location to another, the contact surfaces of the footwear will retain unmixed small particle traces that are directly representative of the last location where the deceased walked. Comparison with the location where the body was found will determine whether or not the body was moved and, if so, the traces will provide clues helping to locate the area from which the victim was transported. Alternatively, for footwear associated with a suspect, it is clear that the traces to be compared with crime scene locations are *not* those on the contact surfaces; rather they are those from recessed surfaces (see point 3, below).

*2. Methods for differential sampling of the contact surfaces of footwear need not be employed.* Given the absence of differences among loosely, moderately, and tightly held particle populations on the contact surfaces of footwear, there is no need for differential sampling of particles on these surfaces. Moist swabbing can be employed directly to

comprehensively recover the adhering particles traces, now known to represent the most recent surfaces walked upon. This finding simplifies the collection of these particle traces and allows their separation as a distinct fraction from other accumulations of particles present on the footwear.

*3. Recessed areas of footwear are likely responsible for the observed retention of particles from prior exposures.* The mixing of particles on footwear, arising from activity before, during and after the crime itself, has been the major obstacle to the exploitation of this type of evidence as a means to test the association of an accused to a crime. Our research shows that the contact surfaces of the soles retain particles from only the more recent exposures. This indicates that the mixtures of particles seen on footwear arise from the recessed areas of footwear, and that it is these areas that should be sampled for evidence of prior exposures. Removal of the fraction from contact surfaces will reduce the complexity of the mixture and can lead to alternative approaches to differential sampling (see point 4, below).

*4. Research on differential sampling of footwear should continue, focusing on the difference between particle populations found on contact surfaces and those found on recessed areas.* Recessed areas of footwear are responsible for the mixtures of particles arising from activity before, during and after the crime itself. The results of our research allow the isolation of particles from the most recent exposures (by sampling the contact surfaces). Subtracting this background from the mixtures found within recessed areas of the footwear provides a likely means to separate the evidentiary particle “signal” from background noise and the exploration of this possibility remains an intriguing area for follow-on research.

*5. Research on related computational and statistical methods to objectively interpret mixtures of particles should continue, focusing on multivariate methods that take advantage of both qualitative and quantitative distinctions among traces and possible sources.* Given that the current differential sampling approach (based on how tightly particles are held) cannot be conveniently adapted to recessed areas of the soles, an alternative means to separate mixtures of particle signals needs to be developed. Following the approach in point 4 above, computational and statistical methods will be needed to subtract the “signals” from the most recent exposures (found on the contact surfaces) and objectively resolve the mixtures that are found in recessed areas.

## V. References

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**VI. Dissemination of Research Findings (As of 3/29/16)****Presentations**

3	Stoney, D.A. and Stoney, P.L., Differential Sampling of Footwear to Separate Alternative Particle Signals, American Academy of Forensic Sciences 68th Annual Meeting, Las Vegas, NV, February 11, 2016.
2	Stoney, D.A. and Stoney, P.L., The Need and Characteristics of a New Approach to Trace Evidence Analysis, NIJ Impression, Pattern and Trace Evidence Symposium, San Antonio, TX, August 27, 2015.
1	Stoney, D.A. and Stoney, P.L., Particle Combination Analysis: Developing a New Approach to Forensic Trace Evidence, National Institute of Justice 2015 Research Grantees Meeting, Orlando, FL, February 17, 2015.

**Publications**

2	Stoney, D.A. and Stoney, P.L. Differential Sampling of Footwear to Separate Alternative Particle Signals, Proceedings of the American Academy of Forensic Sciences, Vol. 22, p. 285, February, 2016.
1	Stoney, D.A. and Stoney, P.L., The Need and Characteristics of a New Approach to Trace Evidence Analysis, in Conference Proceedings: 2015 Impression, Pattern, and Trace Evidence Symposium; Roper-Miller, J.D., Daye, C.M., and Eldridge, H.H., Eds, RTI Press Publication No. CP-0001-1512, 2015