JANNAF WORKSHOP PROCEEDINGS – FATE, TRANSPORT AND EFFECTS OF INSENSITIVE MUNITIONS: ISSUES AND RECENT DATA

Environmental Restoration Report

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## JANNAF WORKSHOP PROCEEDINGS – FATE, TRANSPORT AND EFFECTS OF INSENSITIVE MUNITIONS: ISSUES AND RECENT DATA

### Abstract

A workshop was held in Charleston, South Carolina 19 May 2014 in conjunction with the Joint Army, Navy, NASA, Air Force (JANNAF) Propulsion Committee, Safety and Environmental Protection Subcommittee to discuss recently acquired data and to assess the “state of the science” available for compounds used in insensitive munitions (IM). Representatives from the research, development, testing and evaluation (RDT&E) and acquisition provided insights regarding IM development and utility, while also providing their perspective of existing data gaps and improvements to data acquisition process. Further, principal investigators that have responded to Environmental Restoration Statement of Need (ERSON) 12-02 provided updates on current work for discussion. The workshop concluded with a discussion on future directions with a focus on existing and emerging research needs. The results of this discussion as well as contributions from all of the presenters have been captured in these proceedings.

### Subject Terms

Insensitive munitions, IMX, fate, transport, toxicity
The following proceedings are the result of a workshop held in conjunction in the 27th Safety and Environmental Protection Subcommittee Meeting of the Joint Army, Navy, NASA, Air Force (JANNAF) Propulsion Meeting held at the Charleston Convention Center on 189 May 2014. Many of these data and discussions were the result of research supported by the Strategic Environmental Research and Development Program (SERDP)/Environmental Restoration Program, ER-2223.

The views expressed in this paper are those of the authors and do not necessarily reflect the official policy of the Department of Defense, Department of the Army, U.S. Army Medical Department or the U.S.

Respectfully submitted,

Mark S. Johnson, Editor
Chair, Safety and Environmental Protection Subcommittee
JANNAF
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OVERVIEW

A workshop was held in Charleston, South Carolina 19 May 2014 in conjunction with the Joint Army, Navy, NASA, Air Force (JANNAF) Propulsion Committee, Safety and Environmental Protection Subcommittee to discuss recently acquired data and to assess the “state of the science” available for compounds used in insensitive munitions (IM). Largely, this workshop was a follow-on to two other workshops intended to scope out research needs and data gaps needed to better understand the toxicity, fate and transport for these compounds.

Representatives from the research, development, testing and evaluation (RDT&E) and acquisition provided insights regarding IM development and utility, while also providing their perspective of existing data gaps and improvements to data acquisition process. Further, principal investigators that have responded to Environmental Restoration Statement of Need (ERSON) 12-02 provided updates on current work for discussion.

The workshop concluded with a discussion on future directions with a focus on existing and emerging research needs. The results of this discussion as well as contributions from all of the presenters have been captured in these proceedings.

INTRODUCTION

The U.S. Armed Forces have experienced needless loss of life and equipment due to inadvertent detonation of munitions via sympathetic detonation, non-explosive impact, or the action of fire upon munitions. As a result of the Department of Defense (DOD)-wide initiative to improve the safety of munitions, the U.S. Army is developing insensitive munitions for use in future weapon systems (Duncan 2002). Currently, there are initiatives in the RDT&E and the acquisition stages to develop formulations to replace high explosive fills in warheads where possible. The objectives are to develop an explosive formulation that will only detonate when desired, and will be insensitive to other environmental effectors. IMX-101 is an example of a formulation that is intended to provide explosive performance comparable to current explosive formulations, but not be sensitive to other environmental and/or operational insults, whether by accident or enemy action, which can directly save lives of military personnel and reduce potential for loss of materiel.

Insensitive munitions are "munitions which reliably fulfill their performance, readiness and operational requirements on demand, and which minimize the probability of inadvertent initiation and severity of subsequent collateral damage to weapon platforms, logistic systems, and personnel when subjected to unplanned stimuli” (Beyard 2007). In addition to minimizing collateral damage from weapon or ordnance accidents, insensitive munitions offer logistical advantages on the battlefield; more munitions can be stored in a given area if quantity-distance requirements are reduced, resulting in more efficient use of available land and smaller targets for potential enemy action. As modern battlefields increasingly shift into populated urban centers, insensitive munition inventories represent a less desirable target for terrorists and minimize the threat to surrounding communities. Less-sensitive munitions could potentially be more cost effective and efficient to transport if granted reduced DOD/Department of Transportation hazard
classification rankings (3). IMX-101 is planned for use in several weapons systems including the M795 155mm artillery projectile.

Current regulations require assessment of human health and environmental effects arising from exposure to substances in soil, surface water, and ground water. In the past, explosives and other energetics manufactured and used have caused unintended adverse consequences through environmental releases. Given the need to develop and produce an insensitive product for our forces in theater, efforts to implement these formulations have been hastened.

Two multi-agency workshops were previously held to scope out data needs, requirements, and methods to help address environmental, safety, and occupational health consequences from production and use. Following the outcome of those workshops, the Strategic Environmental Research and Development Program (SERDP) published a request for proposals to address a statement of need (SON) ER-1202, Environmental Fate and Impacts of Insensitive Munitions Compounds. Several investigators were subsequently funded and tasked to provide data to address some of these concerns. These investigators, along with representatives from the RDT&E and acquisition phases of IM development and use provided a synopsis of their issues, data, and direction for future work, where appropriate. This synopsis took the form of a workshop conducted at the Charleston Convention Center, Charleston, SC in conjunction with the Joint Army, Navy, NASA, Air Force (JANNAF) Propulsion Committee, Safety and Environmental Protection Subcommittee meeting, 19 May 2014. Presenters were asked to provide reports of their work which formed the basis of these proceedings.
1.0 Balancing Insensitive Munitions, Environmental, Safety, and Occupational Health (ESOH) and Performance in Next Generation Weapon Systems: The JIMTP Perspective

William Ruppert, IV – U.S. Army Research Laboratory, Program Manager, Office of the Under Secretary of Defense (Acquisition, Technology & Logistics) / Tactical Warfare Systems / Land Warfare and Munitions, Aberdeen Proving Ground, MD
Noah J. Lieb, P.E., CSP – Hughes Associates, Inc., Baltimore, MD

The Joint Insensitive Munitions Technology Program

The Joint Insensitive Munitions Technology Program (JIMTP) addresses applied research and advanced technology development associated with improving the lethality, reliability, safety, and survivability of munitions and weapon systems of the Department of Defense (DOD), with an emphasis on Insensitive Munitions (IM) technology. The goal is to develop joint enabling technologies that can be used by the Services as they develop their specific weapon programs to ensure they meet the requirements identified in USC, Title 10, Chapter 141, Section 2389 December 2001 “Ensuring safety regarding insensitive munitions. The Secretary of Defense shall ensure, to the extent practicable, that insensitive munitions under development or procurement are safe throughout development and fielding when subject to unplanned stimuli.” These unplanned stimuli take the form of rapid or slow heating events, such as a fuel fire on a vehicle or aircraft or an adjacent fire in a vehicle or storage magazine; impact by fragments or bullets, due to shrapnel from nearby explosions or small arms fire from combat or terrorist events; sympathetic reaction due to the detonation of adjacent munitions; and shaped charge jet attack from rocket propelled grenade or similar weapons used by friendly and enemy forces.

The JIMTP strives to ensure the development of technology with the broadest applicability while avoiding duplication of efforts. The program is directed by the Office of the Undersecretary of Defense, Acquisition, Technology and Logistics, Tactical Weapons Systems, Land, Warfare & Munitions.

The fundamental premises of the program are that: (1) IM is one important munition attribute; (2) sensitivity is one important energetic attribute; (3) incremental improvement is important; (4) technology options enable acquisition decisions; and (5) technology solutions which provide positive impact for the warfighter and mission are the primary drivers on whether or not a technology is pursued. The program measures its success by transitioning solutions to munitions program managers who incorporate the technology in weapons which are deployed to the field, water, or air environments.

To reduce the hazard level of munitions, researchers are working technology solutions to (a) increase the stimulus level that is required to cause a reaction, (b) decrease the probability that a reaction will occur, (c) decrease the severity of the reaction to an unplanned stimuli, and (d) increase the time between the introduction of the unplanned stimuli and the reaction to the event. Munitions reactions are categorized as Type I through Type VI, where Type I reactions are a detonation, Type II a partial detonation, Type III an explosion, Type IV a deflagration or
propulsive event, Type V a burning reaction, and Type VI a no reaction. (A complete description of each reaction Type can be found in MIL-STD 2105D.)

**Environmental Concerns Associated with IM Compounds**

The JIMTP is developing and testing new chemicals and munitions concepts in order to meet performance and sensitivity goals. This introduces a potential additional risk because new chemicals are usually not well characterized for potential environmental, safety, and occupational health (ESOH) impacts and often these materials will have no available data at all. The JIMTP recognizes this issue and has established a working relationship with the ESOH community to begin to develop data. Select projects in the JIMTP have used the voluntary ASTM® E-2552-08, Standard Guide for Assessing the Environmental and Human Health Impacts of New Energetic Compounds, phased approach to begin to evaluate ESOH data for new materials. However, without a standardized, consistent process or oversight, some materials will not have adequate ESOH data when they transition to the acquisition community. Limited ESOH data for regulators or uncertainty for new materials has led to delays in production, loading and eventually, implementation of new, more insensitive munitions.

The IM community also faces an occasional relative risk decision between IM and environmental concerns. In some cases, a potential environmental contaminant has been shown to provide significantly reduced sensitivity. For example, ammonium perchlorate has been used in highly insensitive explosive formulations like PAX-21 even though it is a known groundwater contaminant. In the end, it is up to the acquisition community to make the trade-off between much less sensitive, safer munitions and potential long-term environmental contamination.

The Explosive Ordnance Disposal (EOD) of IM materials has been shown to have the potential for additional environmental concerns. Unexploded ordnance with very insensitive energetic fills are more difficult for EOD to destroy efficiently using traditional blast in place (BIP) methods. BIP can result in cracked casing which can release uncombusted energetic in the environment. With the very insensitive material, there is a higher risk of environmental release. This was confirmed by a study conducted by the Cold Regions Research and Engineering Laboratory and is further documented in their paper in this workshop entitled “On the Importance of Environmental Testing of Munitions.” All of these issues in combination show the need to understand the potential ESOH risks of new insensitive explosives.

**The Need for Data**

The prevailing paradigm in U.S. regulatory agencies is that the regulator must first prove that a chemical poses a risk - defined as both hazard and exposure - before regulation can occur. Current regulation has very limited data requirements and does not provide a clear path to identifying key ESOH data points. As a result, when there is no available data, regulators must rely on models, data from analog materials, or other methods with high levels of uncertainty in order to determine if a risk exists. This current paradigm makes it difficult for these agencies to evaluate and potentially restrict materials.
There is a movement in the international community to move to a precautionary approach to regulation. The European Union (EU) was one of the first regulatory communities to make this change through their Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH) regulation. REACH shifts the burden of proof of hazard (or conversely, safety) from the regulator to the manufacturer or supplier of a chemical, which in turn requires collection of a large, defined quantity of environmental / toxicity data for both new and existing chemicals in commerce. Both China and Korea have a similar regulation in place. While regulatory schemes are developing, environmental non-governmental organizations (eNGOs) are placing pressure on retailers as well. Large retailers such as Walmart and Target have put programs in place to restrict hazardous materials used in their supply chain. In part due to this action, the U.S. Environmental Protection Agency (EPA) is shifting focus where possible to this precautionary approach and they may be forced to take action with proposed new regulation. Toxic Substance Control Act (TSCA) Reform is gaining bipartisan support in Congress in both the Senate, through the proposed Chemical Safety Improvement Act, and the House of Representatives through the Discussion Draft of the Chemicals in Commerce Act. Both of these TSCA reform bills propose that new chemicals will only enter into commerce after EPA finds that material will NOT present an unreasonable risk of injury to health/environment. This could also be applied to existing chemicals. This will give EPA an increased ability to restrict materials in commerce and require sufficient ESOH data for evaluation. As such, DOD needs to be prepared for a change in regulatory philosophy.

**Implementing ESOH Evaluation**

In 2010, the Army Research, Development and Engineering Command created the Developmental Environment, Safety and Occupational Health Evaluation (DESHE) process to collect ESOH data for use in all stages of research, development, test and evaluation (RDT&E) for all new materials, processes or technologies (not just energetics). The DESHE built on the framework of ASTM E-2552-08. The process is intended as a lead-in to the Programmatic Environment, Safety and Occupational Health Evaluation (PESHE) required for weapon system acquisition programs. The DESHE outlines the overall phased process to gather, develop and document ESOH data for materials, processes and technologies. The DESHE uses a question-driven approach that is tied to existing Army metrics of RDT&E budget activity and technology readiness level. In the early stages of the DESHE, qualitative data are developed, often through computational models or comparative or analog materials. As the new material, process, or technology matures through the stages of RDT&E, more robust, quantitative data are developed. The intent is twofold: 1) to use the data for decision making in the next stage of RDT&E and 2) to hand off the final data to the customer for their own use in decision making, as documented through the PESHE, Toxicity Clearances, Health Hazard Assessments, etc.

In 2014, the JIMTP initiated a project to implement the recommendations in the 2010 “Guide to Performing a DESHE” to gather ESOH data points for materials proposed for use in all projects. This pilot scale process will evaluate data availability for each material based on maturity level (as defined by RDT&E Budget Activity) to determine if adequate data are available at each stage of development. The JIMTP is not performing a risk assessment on these materials and is not generating additional ESOH data at this time. As an example, it is
recommended that BA2 projects will be able to gather supporting data (either computer models or experimental, in vitro testing) to determine if the material has the potential to transport in the environment, or if it has the potential to be toxic in aquatic environments. At BA3, the DESHE recommends a more thorough robust understanding of transport in the environment, to include potential for degradation in different media or the impacts of acute exposures in aquatic environments or mammals. The findings will be reported to the project leads for the JIMTP so that the data can transition with these materials throughout RDT&E into acquisition.

**Recommendations**

The JIMTP will continue to implement this pilot process of the DESHE; however, for further widespread implementation, additional steps must be taken. There are several offices in place that do collect and evaluate ESOH data when available. This includes the Army Public Health Command and the Office of the Secretary of Defense Chemical and Material Risk Management Directorate. These offices do not have oversight over RDT&E or the responsibility to require data collection across all of the Services at this time. In order to ensure that ESOH data is available for all RDT&E, it is recommended that DOD establish responsibility for ESOH data collection in a single office. Until there is a requirement or a single responsible party, each RDT&E program will act independently. Once this responsibility is established, it will be possible to implement a joint, cross-functional DESHE-like process to collect ESOH data and evaluate all materials in a phased approach. At the very least, this should identify and fulfill minimum data requirements for different technology maturity levels. An ESOH evaluation program will also require directed funding specific to ESOH data generation and evaluation. Funding from individual programs is more likely to be difficult to manage and execute. Finally, DoD will need to formalize a process to incorporate ESOH data into RDT&E decision making and risk evaluation prior to implementation. With this process in place, DOD will be a leader in ESOH evaluation and serve as a model for more environmentally sustainable RDT&E.
2.0 IMX 101/104 Development into New and Legacy Weapon Systems: Environmental, Safety, and Occupational Health (ESOH) Concerns and Project Manager (PM) Perspective

J. Chang, Program Manager, Combat Ammunition Systems, Picatinny Arsenal, NJ.

ABSTRACT

In compliance with Insensitive Munitions (IM) requirements provided in Title 10, U.S. Code section 2389 and DoD Directive 5000.1, the U.S. Army, Project Manager – Combat Ammunition Systems (PM-CAS) developed and continues to transition insensitive high explosive formulations IMX-101 and IMX-104 into artillery and mortar cartridge production. Both IMX-101 and IMX-104 were developed to reduce the reaction of the High Explosive (HE) when exposed to unintended stimuli, such as fire, bullet and/or fragment impacts. These new melt cast explosives are replacements for trinitrotoluene (TNT), Composition B and 1st generation IM PAX-21 legacy explosives. The IMX-101 formulation consists of 3-Nitro-1, 2, 4-triazol-5-one (NTO), Dinitroanisole (DNAN), and Nitroguanidine (NQ), while the IMX-104 formulation includes NTO, DNAN and RDX. The IMX-101 has been evaluated and qualified for production in the following artillery rounds: 105mm HE M1, the 155mm HE M1122 trainer, and the 155mm HE M795 artillery projectiles. The IMX-104 has completed qualification testing in both the 81mm and 60mm HE mortar cartridges and is being evaluated in the 120mm HE Mortars.

Throughout the development and qualification of these insensitive explosive formulations, PM-CAS engaged the ESOH community to characterize these IM formulations for the purpose of program execution/assessment and documentation. Based on the resulting ESOH guidelines PM-CAS provided resources to fund and augment production transition efforts at multiple facilities.

As the PM-CAS team increased interactions with the ESOH community, as more stakeholders showed interests, the team witnessed an extended “storming” phase of interaction dynamics. Examples of these obstacles are: different objectives/cultures leading to trust issues; unbounded data desires; differing horizon/magnitude utilized for risk assessments; many highly technical/esoteric reports but few translational discussions/documents for lay team members’ use; inadequate appreciation of the unique HE production environment/industry; a need for documented requirements shifts; a need for coordinated decision process; and a need for a well-rounded, over-arching leadership group to drive disciplined periodic reviews/updates.

Having expanded significant resources over these now recognized challenges, PM-CAS is working towards influencing a community structure that can function more effectively under harmonized Insensitive Munitions and ESOH DoD objectives to support the path to fielding for IMX 101/104- to move off of the “storming” into the “norming” phase. Simultaneously PM-CAS would advocate for quick establishment of a cross-community, requirements driven, risk balanced ESOH protocol, that keeps the DoD facility/personnel capabilities in view, to be used for acquisition planning for future materiel. PM-CAS’s goal is to leverage the IMX lessons to
enhance synergy for better efficiencies in the pursuit of improved IM explosives and energetic materials for the Warfighter.

**IMX-101 and IMX-104 Background**

The mission of the PM-CAS includes performing life-cycle management of tube-launched indirect fire munitions. Contained within this area are HE projectiles and cartridges for artillery and mortar applications. There are a total of 15 HE indirect fire munitions: four for 105-mm artillery, three for 155-mm artillery, three for 60-mm mortar, three for 81-mm mortar, and two for 120-mm mortar. Most of these HE munitions use either TNT or Comp B, with the exception of Excalibur and two of the 60-mm mortars, which use PBXN-9 and PAX-21, respectively.

In 2005, a holistic material change program was developed by PM-CAS to address the IM requirements provided in Title 10, U.S. Code section 2389 and DoD Directive 5000.1. These IM requirements were implemented to reduce the reaction violence of munitions when subjected to accidental or unplanned events such as fires, bullet, fragment and shaped-charge jet impacts, as well as mass propagation when a single munition experienced a detonation. When PM-CAS items filled with TNT and Comp B explosives underwent IM testing, the resulting reactions were violent detonations of the item as well as adjacent munition items, prompting a need for major design changes necessary to meet the stringent IM requirements.

The PM-CAS Common Low-Cost Insensitive Munitions Explosives (CLIMEx) program was developed with the primary goal of developing a single common explosive fill for all artillery and mortar products. However, the munitions’ performance requirements and the state of technology rendered two explosive fills, one that was common for replacement of TNT and another that was common for replacement of Comp B. Through an extensive down-selection process that included IM and performance testing in representative munitions and cost (including both unit cost of the explosive and cost impact to the loading process at the final munition manufacturing site), IMX-101 was selected to replace TNT and IMX-104 was chosen to replace Comp B. The IMX-101 formulation consists of NTO, DNAN, and NQ, while the IMX-104 formulation includes NTO, DNAN and RDX.

Support for the CLIMEx program, both technical and financial, was received from multiple Army and DoD organizations, including the Joint Insensitive Munitions Technology Program (JIMTP), the Program Executive Office for Ammunition’s (PEO Ammo’s) IM Technical Thrust Area, the Armaments Research, Development and Engineering Center (ARDEC), as well as from contractors such as BAE Holston. In addition, panels such as the Army Insensitive Munitions Board (AIMB) the Army Energetics Material Qualification Board (EMQB), the Naval Ordinance Safety and Security Agency (NOSSA), and the Joint Services Insensitive Munitions Technical Panel (JSIMTP) provided guidance with regard to testing standards and programmatic efforts.
Qualification and Transition to the Field

Subsequent to the development and selection of IMX-101 and IMX-104, these new explosives entered the standard U.S. Army qualification process that includes the following:

- IMX-101 Qualification as a melt-pour explosive for large caliber munition applications
- IMX-104 Qualification as a melt-pour explosive for medium caliber munition applications
- PBXW-14 Qualification as a booster explosive (Insensitive booster explosive required to ensure IM and reliability requirements were both met)
- Qualification of the 155mm IM HE M795 artillery projectile (with IMX-101 HE fill)
- Qualification of the 155mm IM HE M1122 artillery projectile (with IMX-101 HE fill)
- Qualification of the 105mm IM HE M1 artillery projectile (with IMX-104 fill)
- Qualification of the 81mm IM HE M821A3, M889A3, and M889A4 mortar cartridges (with IMX-104 fill)
- Qualification of the 60mm IM HE M720A2, M768A1, and M888A1 mortar cartridges (with IMX-104 fill)

Each of these qualification test series lasted at a minimum of 1 year, with some going as long as 2 years, and cost upwards of $1-2M each.

For explosive qualification, the test series evaluate compatibility and thermal stability; ignition temperatures; stimuli sensitivities; chemical, physical and mechanical properties; variations in property due to aging, detonation products, and performance properties. Once testing is complete the EMQB evaluates the results and issues a qualification memo if all criteria are met.

For system qualification, extensive performance, reliability and safety testing is completed and coordinated with the Army Test & Evaluation Center (ATEC) to ensure all testing is completed according to applicable requirements. These tests include adverse environmental conditioning at extreme operating temperatures as well as sequential safety and rough handling testing, which includes dropping, vibrating, and abusing the munitions to ensure the safety and suitability for warfighter use. The efforts included lethality tests to characterize the performance of the munitions, Explosive Ordnance Disposal (EOD) tests to develop procedures for disposal, and IM tests on the final configuration to evaluate the munitions final IM reactions.

Once testing is completed, a series of evaluations and documents are developed to describe the life-cycle of the munitions. These include environmental reviews to describe any environmental concerns during production or in the field, and health evaluations that would dictate occupation safety precautions to be taken during the manufacturing and life-cycle of the munitions; technical data packages (TDPs) with munition and packaging drawings, nomenclature, National Stock Numbers, Department of Defense Identification Codes, etc.; production quality standards and specifications; system safety and hazard analyses; hazard classification for storage and transportation; performance evaluations that review safety, lethality, IM performance, and ballistics; logistics documents that describe how the munitions are used, handled, stored, transported, and disposed of at the end of their life cycle; EOD procedures for the disposal of unexploded ordnance.
ESOH for the M795

The first munition that went through qualification and transition to use an IMX explosive was the 155mm M795 artillery projectile. When the material change program (MCP) for this projectile was developed (after the completion of the CLIMEx program), the plan was to modify the current M795’s TDP through an Engineering Change Proposal (ECP) rather than through Type Classification (TC) and Full Materiel Release (FMR). This approach allowed PM-CAS to tailor documentation requirements and ultimately save money and time to field the new IM munition. While the shortened ECP approach was used, the guidelines required for the TC and FMR in accordance with (IAW) Department of Defense Instruction (DoDI) 5000.02, “Operation of the Defense Acquisition System”, dated 8 December 08, implementing Department of Defense Directive DoDD 5000.01, “The Defense Acquisition System”, dated 12 May 2003 were followed with regard to ESOH.

In compliance with the DoD’s acquisition process requirement to assess the environmental impacts, a Life Cycle Environmental Assessment (LCEA) was developed by the Armament Research Development and Engineering Center (ARDEC) and approved by PM-CAS in 2010 to satisfy the National Environmental Policy Act (NEPA). This document outlined the evaluation of affected environments during the production and deployment phase, the operations and support phase, and the demilitarization and disposal phase of the M795’s life cycle. Environments evaluated included air quality, surface/ground water quality, noise generation, electromagnetic emissions, and energy consumption. The overall finding of the LCEA was a Finding of No Significant Impact (FNSI).

To address occupational health, PM-CAS funded the United States Army Public Health Command (USAPHC) to develop Occupation Exposure Limits (OELs) for DNAN, NTO, and NQ as well as a toxicity clearance for IMX-101. Once the toxicity clearances and OELs were completed, PM-CAS began working with ARDEC loading facilities, as well as explosive and artillery production contractors, to develop appropriate personal protective equipment (PPE) requirements. This endeavor was coordinated with industrial hygienists to ensure subject matter experts were engaged in the process. Air sampling was conducted to evaluate workers’ exposure to the IMX-101 constituents and to determine if additional PPE and/or equipment modifications would be needed to reduce the risk to operators. As a final summary document, the Programmatic Environmental Safety and Occupational Health Evaluation (PESHE) was developed by ARDEC to capture all current ESOH practices and analysis. This document would then provide guidelines to manufacturers, test ranges and demil operations to ensure environmentally responsible decision are made when working with the M795 IM projectiles.

ESOH Plan Changes

At the time that the LCEA and PESHEs were being developed (but not yet approved), the Joint Munitions Command (JMC) Command Surgeon challenged the OELs that USAPHC provided; requested some industrial hygienists to stand down on IMX program execution; and requested that the established OEL values undergo independent peer reviews and further evaluation. This was an unusual scrutiny of the Army’s occupational health SMEs and
established procedures; however, as the JMC requests were made in the interest of the workforce, the PM agreed to halt the IMX production transition programs to implement the JMC recommendation. This unplanned halt and subsequent peer reviews did lead to cost and schedule growths. The PM witnessed similar edicts that ran outside of the historic requirements for munitions acquisition. Deviations from the normal process do not have an established review/decision point for the new desired data; these trigger multiple tests, peer reviews, and medical reports, but downstream distribution and decision making appeared haphazard, inefficient and lacked an overall DoD objective. The lack of cohesiveness in the distribution of information to the DoD and its contractors inadvertently led to rumors and confusion. One example in particular, the re-evaluations of OELs, and the possible modification of these very important exposure limits, were often misinterpreted and or anticipated prior to being made official. This would in turn bring into question whether PPE requirements at the contractor sites could be modified, or if engineering controls were no longer needed to meet the more limiting OEL. It became unclear who should get the new information, which group of decision makers should resolve conflicts, address risks and make a balanced decision for the U.S. Army, and how it would impact the current production environment. It was also unclear which organizations would be responsible for resourcing the additional efforts that were now being performed and were previously not required and thus, unbudgeted.

**Recommendations**

After many years of steady production with legacy explosives, PM-CAS appreciates that multiple parties are studying the effects of IMX-101, IMX-104 and its constituents in the environment and on occupational health, but recommends that cross-functional awareness and understanding be improved. This would increase the efficiency for utilizing DoD resources, allow different parties to leverage work already done, minimize unnecessary replicate efforts, and to send resources to the right endeavors. It would be very beneficial such that the studies can focus on areas that are meaningful to the life cycle of the munitions.

To provide a platform for this cross-functional awareness, an overarching structure identified within the DoD is needed to define which organization funds which mission, who has overarching decision-making authority, who will facilitate what type of discussions, when there are obstacles who will resolve it, risk evaluation, etc. This structure would then need to be reviewed to determine if there are responsibility gaps and, once identified, who will champion to close the gap.

PM-CAS believes that the efforts of its IM programs is worth the work needed to understand its impacts on the environment and occupational health, but needs the guidance and support of the SME communities to be able to gain that understanding in a supportive and trusting relationship. We need to align our goals in order to both protect workers and the environment while providing the Warfighters with safe and affordable munitions at the promised delivery timeframe. If this common goal is adopted, our communities can better work together toward efficiently accomplishing the DoD IM objective, and serve as the principle for any future new DoD energetics.
3.0 On the Importance of Environmental Testing of Munitions

Michael R. Walsh*1, Marianne E. Walsh1, and Charles A. Ramsey2
1U.S. Army Cold Regions Research and Engineering Laboratory
Hanover, NH USA
2Envirostat, Inc. Ft. Collins, CO USA

ABSTRACT

Munitions are subjected to a rigorous series of performance tests prior to acceptance. For ordnance designed to detonate, these tests focus on the detonation characteristics and safety of the munition. Environmental impacts and the toxicity of the components are also considered as part of the life cycle environmental assessment for munitions, focusing on manufacture and disposal. Environmental field-testing of munitions by the Cold Regions Research and Engineering Laboratory has been conducted as part of a series of Strategic Environmental Research and Development Program (SERDP) projects focused on energetics residues deposition resulting from training activities with live ordnance. These tests consist of evaluating the detonation efficiencies of munitions based on residues deposition for various live-fire training scenarios. Particles collected from inefficient detonations are also used in related fate and transport studies. As part of these SERDP-funded research projects, two series of field detonation tests have been completed on insensitive munitions. Tests of PAX-21-filled mortar cartridges in 2012 demonstrated that ammonium perchlorate was not efficiently consumed during high-order and blow-in-place detonations. The toxicity and extreme mobility of perchlorate along with the significant mass of residues following detonation resulted in the reclassification of these rounds, restricting them from use on CONUS training ranges. Future environmental liabilities in the billions of dollars have been avoided by this action. Tests on two types of mortar projectiles containing IMX-104 were conducted in 2013. The insensitive high explosive compound 3-nitro-1,2,4-triazol-5-one (NTO) did not detonate efficiently, leaving gram quantity residues following high-order detonation and hundreds of grams following blow in place (BIP) operations. Toxicological studies of the highly water-soluble NTO were incomplete and the status of the munitions is now under review. These tests illustrate the importance of environmental field-testing and the toxicological assessment of energetic compounds.

INTRODUCTION

Live-fire training is an essential element for the preparation of a military for combat operations. For ground forces, this involves the firing of weapons that may contain energetic materials. The ammunition community carries out an evaluation of all new munitions called a life cycle environmental assessment (LCEA), designed to determine the environmental impact of fielding a round from manufacture to use or disposal. The objective of the LCEA is to avoid future environmental liabilities and to ensure sustainable use of ranges so that future troops can continue to train. The LCEA process is well established and has been used for many years in the U.S. and overseas to evaluate the munitions that are currently in inventory.

Ecological assessments of munitions have been carried out since at least the mid-1970s. These were driven by the National Environmental Policy Act of 1969 (revised since) [1]. Army
Regulation AR 200-2 Section 651.33(f) established that an environmental assessment is required if “a new hazardous or toxic material” results from the fielding of a new weapon system [2]. The LCEA has evolved over the years to its present form. The LCEA examines possible environmental contaminants generated through live fire training through the analysis of combustion byproducts. This method of assessment is quite effective for air emissions, where particle sizes are quite small and the assumption of complete consumption is valid, but in instances where incomplete consumption or transformation of the energetics of a munition item occurs, valuable data may be overlooked.

In the 1990s, environmental contamination on several U.S. training ranges caused loss of the use of those ranges and massive, complex environmental cleanup efforts. At the Eagle River Flats impact area on Joint Base Elmendorf Richardson, AK, a 24-year, $50 M cleanup effort for white phosphorus was recently successfully completed [3, 4, 5]. Range use is still limited to winter training when the ice protects the underlying soil that still may contained buried white phosphorus. At the Massachusetts Military Reservation (MMR), cleanup continues on a groundwater plume that threatens the drinking water of the Cape Cod area [6]. Over $1.2 B has already been spent and predictions are that another 30 – 50 years will be required to complete the cleanup [7]. The ranges at Camp Edwards, located on MMR, are either closed or highly restricted in their use.

The U.S. Department of Defense has been funding the SERDP for over a decade to investigate munitions related issues that were the cause of range closures outlined above. The understanding in the ammunition community was that firing and detonation of munitions would leave little if any residues in the form of the original energetic material. SERDP research conducted by the U.S. Army Cold Regions Research and Engineering Laboratory has demonstrated that energetic residues always remain following the firing or detonation of munitions. Residues in excess of 70% have been found for some weapon systems, although the usual rates are well below 0.1% [8, 9].

With the advent of the new generation of insensitive munitions (IM), SERDP funded five research projects through the Environmental Restoration program to examine fate, transport, and toxicity of the insensitive high explosives (IHE) used in these munitions as well as taking a closer look at some of the IHE compounds that make up these explosives. This paper describes the results obtained to date for one of these projects, SERDP ER-2219: Characterization of Residues from the Detonation of Insensitive Munitions [10]. The focus of this research was deposition residues for IHE compounds, although for completeness we also characterized the deposition mass for other compounds such as hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX), octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine (HMX), and ammonium perchlorate (AP). A case will be presented for the expansion of the LCEA process to cover firing point and detonation residues as part of the certification process for new munitions.

METHODS

The detonation of munitions generally leaves very little residues composed of the original explosive compounds [9]. This is a good indication of the proper functioning of a round. The
common terminology for a fully functioning round is that it results in a high order detonation, in which 99.99% or more of the explosive filler is consumed during detonation. This obviously leaves very little residue, even for the larger artillery rounds, making single-round characterization difficult when the round is fired into an active impact area.

To resolve the problem of detecting very low concentrations of in an area of potentially high background concentrations, U.S. Army Cold Regions Research and Engineering Laboratory (CRREL) has devised a novel method of sampling for post-detonation residues on an active impact range. The range we use for detonation characterizations floods in the winter and freezes over, providing not only separation from potential background contaminants but also providing separation and protection from unexploded ordnance [11]. The ice cover is overlaid with snow, and the snow provides an ideal collection medium for residues (Figure 1). The detonation depositional plume consisting of dark residues on the snow cover is easily demarcated against the clean, bright white snow, giving us an accurate indication of the plume size. Sampling an annulus outside the plume allows quality assurance, as does replicate sampling of all sampled areas. Multi-Increment® (MI) sampling is used, consisting of ≈100 increments per sample with triplicate samples taken from the main deposition plume area. Randomly selected plumes are sampled below the normal 2.5-cm depth as an additional quality assurance procedure. Thus, we have triplicate MI plume samples with random subsurface samples to ensure adequate sampling depth with one or two 3-m wide annuli sampled in replicate outside the demarcated plume to ensure proper plume delineation.

![Figure 1. Detonation residues plume with visually demarcated outline](image)

Processing and analysis of the samples is straightforward because the sampling medium is snow (water). The samples are melted and vacuum filtered to separate the aqueous portion from the solids. The aqueous portion is subsampled into two to four 500-mL aliquots, of which one to three are concentrated using solid-phase extractions where appropriate. The solids are air-dried and extracted using the proper solvent. The solvent is then prepared for and analyzed using either gas or liquid chromatography [12]. The results are a concentration that can be worked
back through the extract size, sample size, and plume size to derive an estimate of the total mass of a particular compound resulting from a detonation.

Using this method, we have characterized detonation residues for nine weapon systems and 13 munitions. Fifteen munitions have been characterized for BIP detonation operations. Three types of engineering demolitions have also been characterized. For detonation characterization, we normally test seven or more of the same munition to enable statistical analysis of the results. For firing points, 18 weapon systems and 19 munitions have been characterized for propellant deposition. For firing points, we fire up to 100 rounds, depending on the size of the weapon system, and characterize the deposition plume resulting from the activity.

Munitions containing two IHE formulations have been tested to date. In March of 2012, 60-mm mortar cartridges drawn from stock and containing PAX-21, a mixture of 2,4-dinitroanisole (DNAN), RDX, and AP, were tested [13]. Residues depositions were characterized for high-order and BIP operations for seven rounds each. In February 2013, 60-mm and 81-mm mortar bodies containing IMX-104, a mixture of RDX, DNAN, and NTO were tested using the same procedure [14]. All seven rounds in each IMX-104 test contained the standard issue supplementary charge. High-order and BIP detonations were characterized. In all tests, the rounds were placed 40- to 60-m apart and command detonated using a screw-in fuze simulator containing a C4 booster charge and blasting cap.

RESULTS

PAX-21 Results

The test site consisted of frozen soil and shallow ice (<1 m) overlain with a 1-m deep snow pack. Low-order detonation tests using various amounts of C4 in the booster cup indicated that a reliable high-order detonation would occur with a booster load of 10 g of C4 or more. All cups for the tests were therefore loaded with a mass of 12 g of C4. The loaded fuze simulators were screwed into the nose of the rounds until the booster material contacted the PAX-21 IHE load. The temperatures at the site ranged from -18°C in the morning to -1°C in the afternoon. Skies were clear and the sun strong, causing some melting of the snowpack during testing. The 673rd Civil Engineering Squadron, JBER-Elmendorf, provided explosive ordnance disposal (EOD) support for these tests.

The results of the high-order detonation tests indicate that although the RDX and DNAN detonated efficiently, the AP did not (Table 1). The average detonation efficiency for the two organic explosive compounds was 99.993% for the RDX and 99.994% for the DNAN. This high consumption level is indicative of a high-order detonation. However, the efficiency of the AP consumption was only 85%. Samples were analyzed in replicate at three independent labs to confirm the results. The relative percentage differences (RPDs) between the three high-order detonation sample analyses data sets averaged 7%. The mass of organic explosives residues estimated in the outside the plume (OTP) areas was <1.2% of the total estimated mass of all areas combined. Only two OTP samples had detectable concentrations of RDX in them, and both were below the reporting limits of the instrumentation.
Table 1. Mean high-order detonation IM deposition results: PAX-21

<table>
<thead>
<tr>
<th>Plume Area (m²)</th>
<th>OTP Area (m²)</th>
<th>Analyte (c)</th>
<th>Est. Plume Mass (mg)</th>
<th>Est. OTP Mass (a) (mg)</th>
<th>% of Original Analyte Mass</th>
</tr>
</thead>
<tbody>
<tr>
<td>330</td>
<td>250</td>
<td>DNAN</td>
<td>7.1</td>
<td>0.023</td>
<td>0.006%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>RDX</td>
<td>9.2</td>
<td>BDL (b)</td>
<td>0.007%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>ClO₄</td>
<td>14,000</td>
<td>5.5</td>
<td>15%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Organics (d)</td>
<td>16</td>
<td>0.02</td>
<td>0.006%</td>
</tr>
</tbody>
</table>

(a) Values estimated from analytical results below the instrument’s reporting limit for RDX
(b) BDL: Below Detection Limits for analyte
(c) RDX includes HMX as well (<9% of original mass), ClO₄ is perchlorate from AP
(d) DNAN, RDX, and HMX

Only one sample per high-order detonation test plume was collected for perchlorate analysis. Over 99% of the perchlorate was recovered from the aqueous portion of the processed sample. Replicate analyses for the perchlorate were conducted on the filtrate portion of the archived organic explosives samples only, as there were no archived solids remaining after processing of the plume samples.

Perchlorate in the OTPs was checked for only two plumes, H3 and H5. Both had an estimated mass of <0.05% of the combined plume/OTP mass. Analysis of the remaining OTPs for perchlorate was not performed.

BIPs were analyzed originally only for the organic explosive compounds (DNAN, RDX, HMX). No perchlorate samples were specifically taken for the PAX-21 BIPs, and results are for aliquots taken from archived filtrate samples. However, data from the high-order detonation samples indicate that >99% of the perchlorate is recovered from the aqueous portion of the sample so the perchlorate data should be fairly accurate.

The results for the BIPs follow the same general trend as found for the high-order detonation results, although deposition rates were higher [15]. The organic explosives were consumed fairly efficiently (99.8%) but the AP was not (Table 2: CRREL analyses). For the BIP operation tests, the perchlorate residues averaged 36% of the original perchlorate load. BIPs were analyzed at two labs, CRREL and MAXXAM, with an average RPD between the data sets of 1.2%. Perchlorate data was derived from the archived filtrate samples. No OTP samples were analyzed for perchlorate.

Table 2. Mean BIP results for organic explosives and the perchlorate in BIP filtrate aliquots: PAX-21 (CRREL data)

<table>
<thead>
<tr>
<th>Plume Area (m²)</th>
<th>OTP Area (m²)</th>
<th>Analyte (c)</th>
<th>Est. Plume Mass (a) (mg)</th>
<th>Est. OTP Mass (c) (mg)</th>
<th>% of Original Analyte Mass</th>
</tr>
</thead>
<tbody>
<tr>
<td>330</td>
<td>250</td>
<td>DNAN</td>
<td>720</td>
<td>17</td>
<td>0.61%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>RDX</td>
<td>860</td>
<td>19</td>
<td>0.13%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>ClO₄</td>
<td>35,000 (b)</td>
<td>—</td>
<td>36%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Organics (d)</td>
<td>1600</td>
<td>37</td>
<td>0.21%</td>
</tr>
</tbody>
</table>

(a) RDX (and HMX) + DNAN
(b) Estimated total ClO₄ residues mass for plume samples: Archived triplicate aqueous samples
(c) Mean of the combined plume or OTP estimated masses
**IMX-104 Results**

The test site was frozen over with a 60-cm deep snow pack. Low-order detonation tests using various amounts of C4 in the booster cup indicated that a reliable high-order detonation would occur with a booster load of 10 g of C4 or more. All cups for the tests were loaded with a mass of 12 g of C4 to compensate for any variation in packing density in the fuze simulator booster well. The loaded fuze simulators were screwed into the nose of the rounds until the booster material contacted the supplementary charge inside the rounds. The mortar bodies were screwed into aluminum plates to orient them vertically for the high-order tests (Figure 2). The temperatures at the site hovered around -2°C during the tests. Skies were generally overcast with one short period that was clear with a strong sun, causing difficulties with delineating a few high-order plumes because of residues melting partially into the snow pack. The 716th EOD Detachment, JBER-Richardson, provided support for these tests.

![Figure 2. Assembly of 81-mm round for testing](image)

The results of the high-order detonation tests indicate that although the RDX and DNAN detonated efficiently for both size rounds, the NTO did not (Table 3). The average detonation efficiencies for the 60s and 81s were 99.994% and 99.99% for the RDX and 99.995% and 99.99% for the DNAN. This high consumption level is indicative of a high-order detonation (99%+) and strongly indicates the functioning of the supplemental charge in the nose of the round. However, the efficiency of the NTO consumption was only 98.8% and 99.6% for the two rounds. These results are for the combined average plume and OTP data. OTPs contained 0.009% of the total estimated residues mass for the 60s and 16% for the 81s. The unusually high number for the 81-mm OTPs is attributable to the difficulty in delineating the plume for four of the seven plumes because of the strong sun melting the residues into the snow pack. There was no significant difference between the plumes delineated incorrectly and the correctly delineated plumes when the OTPs were added to the plume totals.
Table 3. Mean high-order detonation IM deposition results: IMX-104

<table>
<thead>
<tr>
<th>Round</th>
<th>Plume Area (m²)</th>
<th>OTP Area (m²)</th>
<th>Analyte(a)</th>
<th>Est. Total Mass(b) (mg)</th>
<th>Range (mg)</th>
<th>% of Original Analyte Mass(c)</th>
</tr>
</thead>
<tbody>
<tr>
<td>60-mm</td>
<td>250</td>
<td>200</td>
<td>DNAN</td>
<td>5.3</td>
<td>3 – 6</td>
<td>0.005%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX &amp; HMX</td>
<td>4.5</td>
<td>1 – 18</td>
<td>0.006%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>NTO</td>
<td>2200</td>
<td>1300 – 2800</td>
<td>1.2%</td>
</tr>
<tr>
<td>81-mm</td>
<td>350</td>
<td>230</td>
<td>DNAN</td>
<td>27</td>
<td>2.1 – 71</td>
<td>0.01%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX &amp; HMX</td>
<td>16</td>
<td>0.6 – 47</td>
<td>0.01%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>NTO</td>
<td>1900</td>
<td>900 – 2800</td>
<td>0.4%</td>
</tr>
</tbody>
</table>

(a) RDX includes some HMX as well (<9% of original mass)
(b) Mean of the combined plume, OTP, and subsurface estimated masses
(c) Based on mid-range value of formulation specifications.

The results for the BIPs for the IMX-104 rounds differ significantly from the high-order results (Table 4). The RDX/HMX residues are in the 1% range, but the DNAN is an order of magnitude higher, indicating an inefficient detonation. The NTO is quite problematic, with about 50% of the original mass of the compound estimated to be in the residues. Particles of IMX-104 were recovered on the filters when processing one of the samples. For that sample, over 98% of the NTO was estimated not to have reacted during the detonation event. That detonation is not included in the data for the 81-mm BIPs in the table below (See Table 4, note (d)). The wide range of residue mass values is also indicative of a poorly propagated detonation.

Table 4. Mean BIP detonation IM deposition results

<table>
<thead>
<tr>
<th>Round</th>
<th>Plume Area (m²)</th>
<th>OTP Area (m²)</th>
<th>Analyte(a)</th>
<th>Est. Total Mass(b) (mg)</th>
<th>Range (mg)</th>
<th>% of Original Analyte Mass(c)</th>
</tr>
</thead>
<tbody>
<tr>
<td>60-mm</td>
<td>580</td>
<td>300</td>
<td>DNAN</td>
<td>20,000</td>
<td>2700 – 34,000</td>
<td>19%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX &amp; HMX</td>
<td>8300</td>
<td>1000 – 17,000</td>
<td>1.4%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>NTO</td>
<td>89,000</td>
<td>54,000 – 124,000</td>
<td>50%</td>
</tr>
<tr>
<td>81-mm</td>
<td>610</td>
<td>320</td>
<td>DNAN</td>
<td>29,000</td>
<td>20,000 – 49,000</td>
<td>11%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX &amp; HMX</td>
<td>11,000</td>
<td>7100 – 19,000</td>
<td>1.6%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>NTO</td>
<td>190,000</td>
<td>160,000 – 250,000</td>
<td>45%</td>
</tr>
</tbody>
</table>

(a) RDX includes some HMX as well (<9% of original mass)
(b) Mean of the combined plume, OTP, and subsurface estimated masses
(c) Based on mid-range value of formulation specifications. Includes 520 g RDX/HMX for BIP donor block.
(d) Excludes Plume 4, which almost failed to detonate

QUALITY ASSURANCE

The background samples taken before each series of tests indicated no presence of the energetic analytes prior to testing. Detonation plumes were sampled in triplicate, with the range of estimated mass values generally within an order of magnitude. The 0-3 m OTP samples were within the <5% of total mass target except for the 81-mm high order detonations when a strong sun made delineation of the plumes difficult. The 3-6 m OTPs were all under 1%. Replicate SPEs were in very good agreement as were replicate injections. All analytical quality assurance was within laboratory standards. There was no carryover with the blank water samples, indicating the cleaning process for the lab glassware was adequate.
DISCUSSION

The field characterization of munitions detonations is an important method for determining the environmental impact from training with new ordnance. The past practice of monitoring air emissions does not adequately address the generation of particulate residues that may result from the detonation of munitions. Monitoring only combustion products ignores the possibility that significant residues of the original energetic material may result from detonations. We have tested scores of weapon systems and munitions and very few of them have left no detectable energetics when used.

Characterization of the two IHE formulations has demonstrated that there will be environmental issues associated with munitions containing these formulations. For the PAX-21, the large mass of the perchlorate residues were unexpected and a munition system that was in inventory had to be reclassified. It had been used on at least 36 ranges in the U.S. and had been successfully used in combat abroad. Over $150 M worth of munitions were affected by the discovery of high concentrations of perchlorates resulting from high-order detonation of the rounds. The costs associated with continued use of these rounds in the U.S. could easily have been in the billions of dollars. The Army acted quickly in reclassifying the rounds, avoiding significant future environmental liabilities.

A closer look is now being taken at the IMX-104 formulation, specifically at the NTO component of the IHE formulation. The toxicity studies for NTO have not been completed, and it is not known if rounds containing NTO will move forward at this time. There are indications from toxicity studies in the U.S. and Canada that adverse effects have been observed in rats, while phytotoxicity and avoidance by earthworms in spiked soils has been observed [16, 17]. The solubility and acidity of NTO, a pH of 3, may also be a factor as metals may be mobilized on ranges that have NTO residues. However, NTO is not perchlorate, and if the toxicity studies prove promising, we will have a solid insensitive compound upon which to base IHE formulations.

We were quite surprised by the results of the IM tests. The common assumption was that because so much research had been done on AP, there would not be a problem with perchlorate in the detonation residues. The statement of need in the SERDP solicitation for IM specifically stated that the investigation of AP was not necessary. We were very close to not analyzing our PAX-21 residue samples for perchlorate, but did so because it was a major component of the IHE formulation. We did not fully believe the results when they came back from the lab and reran them, a few in triplicate, to verify our findings. Problematic munitions are not unprecedented, as demonstrated by the “green” bullets of a decade ago, the white phosphorus at Eagle River Flats, and poor consumption of propellants for the AT4 cartridge [3, 18, 19]. Environmental and thorough toxicological testing, without preconceived assumptions, is likely the best solution to avoiding future liabilities associated with fielding munitions and weapon systems.
SUMMARY AND CONCLUSIONS

Environmental field-testing of munitions containing insensitive high explosive formulations were conducted over the last 2 years. Consumption inefficiencies were revealed for one of the IHE components in each formulation. These inefficiencies were not discovered using the current life cycle environmental assessment process, indicating a shortcoming in that process. Having the necessary data to determine the fate of energetic compound in explosive formulations is critical in the evaluation of the environmental impact of munitions and the consequential effects on range sustainability. The recent experience with the PAX-21 munitions has established this need, and the tests on the IMX-104 munitions reinforce that point. In both cases, a single component of the IHE formulation did not detonate efficiently, leading to high levels of contamination. Based on the mass of NTO remaining on the range following detonation of rounds containing this compound, the lack of complete toxicological data for NTO in the IMX-104 has the potential to delay the certification of all munitions containing NTO. The time and cost associated with running a battery of detonation tests in the field is small compared to the potential consequences of fielding a problematic munition.

FUTURE WORK

CRREL reran the high-order and BIP tests on the 81-mm IMX-104-filled mortar rounds this winter. ARDEC and NSWCIHEODTECHDIV have both provided valuable test design input and were invited to participate in the tests. Detonation tests on IMX-101 rounds were also conducted with 155-mm practice munitions. ARDEC provided the munitions (M1122 practice rounds containing 1 kg IMX-101) and on-site technical expertise during the tests. Samples were being analyzed at the CRREL analytical chemistry lab at the time of this manuscript. A new munition containing iRDX is being considered by CRREL and ARDEC for testing. The Air Force is also interested in testing of the AFX-757 IHE aerial bombs. ARDEC and CRREL are exploring future opportunities in collaborating on the assessment of the environmental performance of future munitions.

ACKNOWLEDGEMENTS

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4.0 Deposition of DNAN and RDX from PAX-21 and IMX-104 Detonations

Marianne E. Walsh, Michael R. Walsh, Susan Taylor, and Charles A. Ramsey

1U.S. Army Cold Regions Research and Engineering Laboratory. Hanover, NH USA
2Envirostat, Inc., Ft. Collins, CO USA

ABSTRACT

Post-detonation residues from mortar projectiles were characterized. The projectiles contained either PAX-21 or IMX-104, insensitive high explosive formulations. Projectiles were detonated on a pristine snow-covered ice surface that served as a collection medium for the residues. IMX-104 is composed of 2,4-dinitroanisole (DNAN) (32%), hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX) (15%), and 3-nitro-1,2,4-triazol-5-one (NTO) (53%). DNAN is a nitroaromatic, like the conventional energetic TNT. RDX is a nitramine and is a conventional energetic. NTO is a nitrotriazolone, a heterocyclic organic compound. It is present in IMX-104 as large, irregularly shaped crystals within a matrix of RDX and DNAN. PAX-21 is composed of three major energetics, two of which are organic: DNAN (34%) and RDX (36%). The third component is the inorganic salt ammonium perchlorate (30%) that, like NTO, is present as large crystals. Detonation efficiencies were estimated from the masses of energetics recovered in the post-detonation residues compared to the initial masses in each projectile. We also measured the efficiencies for blow in place (BIP) detonations, which included a donor block of C4 (520 g of RDX).

This paper discusses the methods used to determine the mass deposition rates for the energetic compounds and summarize the results for the RDX and DNAN. A companion paper (Chaper 3) presents the results for perchlorate and NTO.

INTRODUCTION

The U.S. Army Cold Regions Research and Engineering Laboratory and Envirostat, Inc., have developed methods for obtaining energetics residues samples on pristine snow surfaces that allows the determination of energetic mass deposition on a per-round basis. The method was used to characterize the residues from several conventional munitions under Strategic Environmental Research and Development Program (SERDP) ER-1155 and ER-1481. The same methods are currently being used for SERDP ER-2219 to measure the residues from the newly fielded Insensitive Munitions (IM). Our hypothesis is that IM energetic formulations will have higher residue deposition rates per round because of the high shock forces required to initiate and maintain the detonation train in these less-sensitive formulations.

Tests were conducted using mortar projectiles containing either PAX-21 or IMX-104 [1,2]. Rounds were detonated under two modes: high order (HO) detonations and single-round BIP operations. High-order detonation is the most common mode of detonation for fired munitions. HO detonations were performed by removing the original fuze from the munitions and installing a fuze simulator that was developed by U.S. Army Cold Regions Research and Engineering Laboratory (CRREL) under SERDP ER-1481 that enables high-order detonation without the
need for fuze activation through the live firing of a projectile. This process is known as command detonation. Similar procedures are commonly used during life-cycle environmental assessments of new munitions to determine air emissions from detonations. The booster cup of the fuze simulator was filled with C4, and a blasting cap inserted through the simulator into the booster charge was used to initiate the detonation train. Use of the fuze simulator has advantages over live fire. The detonation site is controlled and can be selected for a reduced danger zone. Detonations can be placed to ensure that there is no spatial overlap of the plumes. Duds are avoided through the command detonation process. BIP operations are normally performed either to render munitions safe or to destroy unexploded ordnance on site during range clearance operations. BIP destructions use an explosive donor charge, typically a block of C4, to initiate a detonation within the unexploded ordnance. Past research has shown that the latter can be a relatively non-polluting operation for conventional munitions containing TNT or Comp B, but a misfire or insufficient shock will result in a low order detonation that will spread large pieces of explosives in the environment. BIP operations are of particular concern for IM, given that the rounds are designed resist detonation initiation from an external shock source and to detonate only when fired.

For these tests, the detonations were initiated on a snow-covered range that was underlain by ice [3]. Advantages of the snow cover are many. It serves as a pristine collection medium, the plume is easily visualized on the white background, the sample processing method is straightforward, and replicate samples may be collected. Ideal conditions are ambient temperatures slightly below 0°C, cloud cover, no wind, and no precipitation.

For each test, there were seven rounds simultaneously detonated in the same manner to measure the variability in the deposition masses. Following detonations, the plumes containing the residues were demarcated by walking the edge, which was determined qualitatively as the border between the area of soot deposition and clean snow. A geographical position system was used to record the outline of each plume, and the data were used to compute the area of deposition. To verify that the visually discernable plume on the snow surface contained the bulk of the residues (>95% goal), we demarcated a 3-m annulus around the main plume area and another annulus extending from 3- to 6-m outside the plume (OTP). To obtain the snow samples, we used 10- x 10-cm polytetrafluoroethylene-lined scoops to collect multi-increment (MI) samples in each plume. We sampled to a depth of 2 cm or deeper if residue was apparent below the surface. A systematic-random approach was used to collect triplicate samples from each plume. Each MI sample within the plume (ITP) consisted of approximately 100 increments that were combined in a special clean polyethylene (PE) lab-grade bag. For OTP units, we collected between 40 and 100 increments. One OTP sample was collected per plume, and triplicate OTPs were taken around randomly selected plumes for quality assurance.

To estimate the mass of energetics in each plume, the snow samples were melted and the solids separated from the aqueous fraction by filtration. Then, deposition rates are estimated from the extrapolation of the mass derived from the analysis of the residues in both the aqueous and solid phases. DNAN is a nitroaromatic with properties (Log Kow, aqueous solubility, vapor pressure) similar to other energetic nitroaromatics, and it was determined using reversed phase HPLC parameters in SW-846 Method 8330, the standard analytical method for explosives [4].
Solid phase extraction using Waters Sep-Pak Porapak RDX cartridges and elution with acetonitrile were used to pre-concentrate the RDX and DNAN residing in the melted snow. Solid residues from the filtration process were extracted with acetonitrile. All of the extracts were analyzed by high-performance liquid chromatography and some of the extracts were analyzed by gas chromatography to confirm the presence of DNAN.

RESULTS AND DISCUSSION

PAX-21

The PAX-21 tests were conducted in March of 2012 using 60-mm mortar cartridges (DODIC BA17) that were drawn from inventory. For these tests, 12 g of C4 (91% RDX) were packed into each booster cup of the fuze simulators prior to installation into the nose of the mortar round. Blasting caps were inserted through an access hole in the top of the simulator into the C4 booster to initiate the explosive train for the HO detonations. The rounds used for the BIP tests had the issued fuze attached (8g HMX). One donor charge block was used for each BIP detonation. Further details of the tests are given in the companion paper by Michael Walsh.

The masses of DNAN and RDX deposited in the plumes (ITPs) and the plume annuli (OTPs) from the HO and BIP detonations are given in Tables 1 a and b. The mean residual masses of the DNAN and RDX/HMX for the seven HO shots were milligram quantities or only 0.006% and 0.007% respectively of the original mass of the compounds in each round. These low residual masses are indicative of high-order detonations based on these two organic compounds. The BIP detonations were far less efficient and yielded near gram quantities of DNAN and RDX. There is no overlap in the mass estimates of residues for RDX or DNAN between the high-order and BIP detonations.

IMX-104

The IMX-104 tests were conducted in February of 2013 using both 60-mm and 81-mm mortar bodies. The tests were conducted in a similar manner to the PAX-21 tests; all rounds contained fuze simulators with booster cup loads of 12 g C4 explosive, and one block of C4 was used as a donor charge for each of the BIP detonations. The exception was that standard-issue fuzes were not used for the BIP tests.

The RDX and DNAN mass deposition from the 60-mm projectiles containing IMX-104 were similar in magnitude to those of the PAX-21 for the HO detonations (Table 2a). Mean deposition masses were 5.3 mg and 4.5 mg for DNAN and RDX, respectively, per round or only 0.005% and 0.006% of the original masses. This compares to 0.07 mg/round of RDX residues for a high-order detonation of a 60-mm Comp-B round [2]. The BIP detonations, however, left significant residues of DNAN (Table 2b); the mean deposition mass was 20 g or nearly 20% of the original mass in the round. RDX mass deposition was also high (8 g), indicating an inefficient (low-order) detonation. RDX mass deposition from a similar operation on a round containing Comp B was 0.2 g. [2]
Deposition from the HO detonations of the 81-mm projectiles was variable, ranging from milligram to tens of milligrams (17 mg average) for RDX and DNAN (Table 3a). The weather conditions for these tests were not ideal in that there was a bright sun, which may have contributed to a higher uncertainty in the plume demarcation and the residues melting into the snow pack. The mass depositions for the BIP detonations were high for these rounds (Table 3b). The mean deposition of DNAN was 44 g per round, which is again nearly 20% of the original mass. The RDX deposition was 20 g per round. For an 81-mm mortar projectile with Comp B HE filler, the RDX deposition mass for HO and BIP operations averaged 8.5 mg and 150 mg respectively. The BIP operation RDX residues mass differs significantly between the conventional HE results and the IMX-104 results. There was no overlap between the high-order and BIP detonation residues masses for the 81-mm tests.

Comparative Efficiencies

Both the PAX-21 and IMX-104 rounds performed less efficiently than comparable rounds filled with Comp B, a conventional high explosive used in many of the current generation of munitions (Table 4). The average efficiencies of the high-order tests for both IHE compounds indicated a high-order detonation, with >99.99% consumption of DNAN and RDX. However, the mass deposition was higher in both cases than for Comp B high-order detonations. The BIP detonation residues for the two IM tests are much greater than those for the Comp B BIP detonations, up to two orders of magnitude for both of the rounds.

Table 1a. PAX-21 High Order Detonations

<table>
<thead>
<tr>
<th>Shot</th>
<th>ITP Area (m²)</th>
<th>OTP Area (m²)</th>
<th>Energetic</th>
<th>Estimated Mass (mg) Deposited Inside Plume</th>
<th>Estimated Mass (mg) Deposited in Annulus</th>
<th>Percent of Original Mass</th>
</tr>
</thead>
<tbody>
<tr>
<td>H1</td>
<td>350</td>
<td>270</td>
<td>DNAN</td>
<td>2.6</td>
<td>BDL*</td>
<td>0.0021%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>1.0</td>
<td>BDL</td>
<td>0.0007%</td>
</tr>
<tr>
<td>H2</td>
<td>350</td>
<td>260</td>
<td>DNAN</td>
<td>18</td>
<td>BDL</td>
<td>0.015%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>40</td>
<td>BDL</td>
<td>0.028%</td>
</tr>
<tr>
<td>H3</td>
<td>300</td>
<td>240</td>
<td>DNAN</td>
<td>17</td>
<td>BDL</td>
<td>0.014%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>16</td>
<td>BDL</td>
<td>0.012%</td>
</tr>
<tr>
<td>H4</td>
<td>460</td>
<td>280</td>
<td>DNAN</td>
<td>1.3</td>
<td>BDL</td>
<td>0.0011%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>0.34</td>
<td>BDL</td>
<td>0.0002%</td>
</tr>
<tr>
<td>H5</td>
<td>280</td>
<td>225</td>
<td>DNAN</td>
<td>5.0</td>
<td>BDL</td>
<td>0.0041%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>0.76</td>
<td>0.16</td>
<td>0.0007%</td>
</tr>
<tr>
<td>H6</td>
<td>260</td>
<td>230</td>
<td>DNAN</td>
<td>2.1</td>
<td>BDL</td>
<td>0.0017%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>4.3</td>
<td>BDL</td>
<td>0.0031%</td>
</tr>
<tr>
<td>H7</td>
<td>280</td>
<td>220</td>
<td>DNAN</td>
<td>2.9</td>
<td>BDL</td>
<td>0.0024%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>1.8</td>
<td>BDL</td>
<td>0.0013%</td>
</tr>
<tr>
<td>Means</td>
<td>330</td>
<td>250</td>
<td>DNAN</td>
<td>7.1</td>
<td>0.023</td>
<td>0.006%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>9.2</td>
<td>BDL</td>
<td>0.007%</td>
</tr>
</tbody>
</table>

*Original analyte masses are 120 g DNAN and 130 g RDX

BDL: Below Detection Limits for analyte
Table 1b. PAX-21 60-mm Blow in Place

<table>
<thead>
<tr>
<th>Shot</th>
<th>ITP Area (m²)</th>
<th>OTP Area (m²)</th>
<th>Energetic</th>
<th>Estimated Mass (mg) Deposited Inside Plume</th>
<th>Estimated Mass (mg) Deposited in Annulus</th>
<th>Percent of Original Mass</th>
</tr>
</thead>
<tbody>
<tr>
<td>BIP 1</td>
<td>370</td>
<td>250</td>
<td>DNAN</td>
<td>720</td>
<td>4.5</td>
<td>0.59%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>860</td>
<td>6.5</td>
<td>0.13%</td>
</tr>
<tr>
<td>BIP 2</td>
<td>380</td>
<td>250</td>
<td>DNAN</td>
<td>460</td>
<td>9.5</td>
<td>0.39%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>430</td>
<td>13</td>
<td>0.07%</td>
</tr>
<tr>
<td>BIP 3</td>
<td>330</td>
<td>250</td>
<td>DNAN</td>
<td>610</td>
<td>59</td>
<td>0.55%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>730</td>
<td>59</td>
<td>0.12%</td>
</tr>
<tr>
<td>BIP 4</td>
<td>280</td>
<td>240</td>
<td>DNAN</td>
<td>1,400</td>
<td>13</td>
<td>1.2%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>1,700</td>
<td>13</td>
<td>0.26%</td>
</tr>
<tr>
<td>BIP 5</td>
<td>310</td>
<td>250</td>
<td>DNAN</td>
<td>780</td>
<td>23</td>
<td>0.66%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>1,000</td>
<td>29</td>
<td>0.16%</td>
</tr>
<tr>
<td>BIP 6</td>
<td>290</td>
<td>240</td>
<td>DNAN</td>
<td>420</td>
<td>3.8</td>
<td>0.34%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>550</td>
<td>4.4</td>
<td>0.08%</td>
</tr>
<tr>
<td>BIP 7</td>
<td>360</td>
<td>260</td>
<td>DNAN</td>
<td>650</td>
<td>8.0</td>
<td>0.54%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>710</td>
<td>10</td>
<td>0.11%</td>
</tr>
<tr>
<td>Means</td>
<td>330</td>
<td>250</td>
<td>DNAN</td>
<td>720</td>
<td>17</td>
<td>0.61%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>860</td>
<td>19</td>
<td>0.13%</td>
</tr>
</tbody>
</table>

*Original analyte masses are 120 g DNAN and 130 g RDX in round and 520 g RDX in donor charge.

Table 2a. IMX-104 60-mm High Order Detonations

<table>
<thead>
<tr>
<th>Shot</th>
<th>Plume Area (m²)</th>
<th>OTP Area (m²)</th>
<th>Energetic</th>
<th>Estimated Mass (mg) Deposited Inside Plume</th>
<th>Estimated Mass (mg) Deposited in Annulus</th>
<th>Percent of Original Mass</th>
</tr>
</thead>
<tbody>
<tr>
<td>H1</td>
<td>193</td>
<td>177</td>
<td>DNAN</td>
<td>5.0</td>
<td>BDL</td>
<td>0.0046%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>1.4</td>
<td>BDL</td>
<td>0.0019%</td>
</tr>
<tr>
<td>H2</td>
<td>239</td>
<td>195</td>
<td>DNAN</td>
<td>5.0</td>
<td>BDL</td>
<td>0.0046%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>1.1</td>
<td>BDL</td>
<td>0.0015%</td>
</tr>
<tr>
<td>H3</td>
<td>206</td>
<td>182</td>
<td>DNAN</td>
<td>6.1</td>
<td>BDL</td>
<td>0.0057%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>18</td>
<td>BDL</td>
<td>0.0242%</td>
</tr>
<tr>
<td>H4</td>
<td>266</td>
<td>203</td>
<td>DNAN</td>
<td>5.2</td>
<td>BDL</td>
<td>0.0049%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>7.1</td>
<td>BDL</td>
<td>0.0095%</td>
</tr>
<tr>
<td>H5</td>
<td>256</td>
<td>199</td>
<td>DNAN</td>
<td>3.3</td>
<td>BDL</td>
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</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>1.3</td>
<td>BDL</td>
<td>0.0018%</td>
</tr>
<tr>
<td>H6</td>
<td>271</td>
<td>204</td>
<td>DNAN</td>
<td>5.3</td>
<td>BDL</td>
<td>0.0050%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>1.7</td>
<td>BDL</td>
<td>0.0023%</td>
</tr>
<tr>
<td>H7</td>
<td>304</td>
<td>214</td>
<td>DNAN</td>
<td>7.0</td>
<td>BDL</td>
<td>0.0065%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>1.0</td>
<td>BDL</td>
<td>0.0013%</td>
</tr>
<tr>
<td>Means</td>
<td>248</td>
<td>196</td>
<td>DNAN</td>
<td>5.3</td>
<td>BDL</td>
<td>0.0049%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>4.5</td>
<td>BDL</td>
<td>0.0061%</td>
</tr>
</tbody>
</table>

*107 g DNAN and 75 g RDX. Based on mid-range value of formulation specifications.
### Table 2b. IMX-104 60-mm Blow in Place

<table>
<thead>
<tr>
<th>Shot</th>
<th>Plume Area (m²)</th>
<th>OTP Area (m²)</th>
<th>Energetic</th>
<th>Estimated Mass (mg) Deposited Inside Plume</th>
<th>Estimated Mass (mg) Deposited in Annulus</th>
<th>Percent of Original Massa</th>
</tr>
</thead>
<tbody>
<tr>
<td>BIP 1</td>
<td>544</td>
<td>285</td>
<td>DNAN</td>
<td>34,000</td>
<td>220</td>
<td>32%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>17,000</td>
<td>89</td>
<td>2.9%</td>
</tr>
<tr>
<td>BIP 2</td>
<td>567</td>
<td>299</td>
<td>DNAN</td>
<td>16,000</td>
<td>160</td>
<td>15%</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>5,400</td>
<td>44</td>
<td>0.9%</td>
</tr>
<tr>
<td>BIP 3</td>
<td>529</td>
<td>295</td>
<td>DNAN</td>
<td>29,000</td>
<td>100</td>
<td>27%</td>
</tr>
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<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>12,000</td>
<td>47</td>
<td>2.0%</td>
</tr>
<tr>
<td>BIP 4</td>
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<td>284</td>
<td>DNAN</td>
<td>2,600</td>
<td>23</td>
<td>2.4%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>990</td>
<td>7</td>
<td>0.2%</td>
</tr>
<tr>
<td>BIP 5</td>
<td>679</td>
<td>320</td>
<td>DNAN</td>
<td>15,000</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>5,500</td>
<td>46</td>
<td>0.9%</td>
</tr>
<tr>
<td>BIP 6</td>
<td>609</td>
<td>305</td>
<td>DNAN</td>
<td>18,000</td>
<td>120</td>
<td>17%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>8,500</td>
<td>44</td>
<td>1.4%</td>
</tr>
<tr>
<td>BIP 7</td>
<td>577</td>
<td>296</td>
<td>DNAN</td>
<td>24,000</td>
<td>82</td>
<td>22%</td>
</tr>
<tr>
<td></td>
<td></td>
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<td>RDX</td>
<td>8,200</td>
<td>24</td>
<td>1.4%</td>
</tr>
<tr>
<td>Means</td>
<td>576</td>
<td>298</td>
<td>DNAN</td>
<td>20,000</td>
<td>120</td>
<td>19%</td>
</tr>
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<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>8,200</td>
<td>43</td>
<td>1.4%</td>
</tr>
</tbody>
</table>

†107 g DNAN and 75 g RDX (plus 520 g RDX for BIP donor block). Based on mid-range value of formulation specifications.

### Table 3a. IMX-104 81-mm High Order Detonations

<table>
<thead>
<tr>
<th>Shot</th>
<th>Plume Area (m²)</th>
<th>OTP Area (m²)</th>
<th>Energetic</th>
<th>Estimated Mass (mg) Deposited Inside Plume</th>
<th>Estimated Mass (mg) Deposited in Annulus</th>
<th>Percent of Original Massa</th>
</tr>
</thead>
<tbody>
<tr>
<td>H1</td>
<td>396</td>
<td>241</td>
<td>DNAN</td>
<td>2.3</td>
<td>BDL</td>
<td>0.0009%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>0.63</td>
<td>BDL</td>
<td>0.0004%</td>
</tr>
<tr>
<td>H2</td>
<td>337</td>
<td>225</td>
<td>DNAN</td>
<td>2.1</td>
<td>BDL</td>
<td>0.0008%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>0.9</td>
<td>BDL</td>
<td>0.0006%</td>
</tr>
<tr>
<td>H3</td>
<td>347</td>
<td>228</td>
<td>DNAN</td>
<td>7.7</td>
<td>0.8</td>
<td>0.0033%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>2.5</td>
<td>0.5</td>
<td>0.0021%</td>
</tr>
<tr>
<td>H4</td>
<td>299</td>
<td>213</td>
<td>DNAN</td>
<td>17</td>
<td>8.5</td>
<td>0.010%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>10</td>
<td>4.7</td>
<td>0.010%</td>
</tr>
<tr>
<td>H5</td>
<td>332</td>
<td>224</td>
<td>DNAN</td>
<td>36</td>
<td>36</td>
<td>0.028%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>23</td>
<td>19</td>
<td>0.029%</td>
</tr>
<tr>
<td>H6</td>
<td>366</td>
<td>234</td>
<td>DNAN</td>
<td>28</td>
<td>29</td>
<td>0.022%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>20</td>
<td>18</td>
<td>0.026%</td>
</tr>
<tr>
<td>H7</td>
<td>341</td>
<td>227</td>
<td>DNAN</td>
<td>11</td>
<td>9.4</td>
<td>0.008%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>7.6</td>
<td>5.2</td>
<td>0.009%</td>
</tr>
<tr>
<td>Means</td>
<td>345</td>
<td>227</td>
<td>DNAN</td>
<td>15</td>
<td>12</td>
<td>0.011%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RDX</td>
<td>9.2</td>
<td>6.8</td>
<td>0.011%</td>
</tr>
</tbody>
</table>

†147 g RDX and 256 g DNAN. Based on mid-range value of formulation specifications.
Table 3b. IMX-104 81-mm Blow in Place

<table>
<thead>
<tr>
<th>Shot</th>
<th>Plume Area (m²)</th>
<th>OTP Area (m²)</th>
<th>Energetic</th>
<th>Estimated Mass (mg) Deposited Inside Plume</th>
<th>Estimated Mass (mg) Deposited in Annulus</th>
<th>Percent of Original Mass*</th>
</tr>
</thead>
<tbody>
<tr>
<td>BIP 1</td>
<td>495</td>
<td>278</td>
<td>RDX</td>
<td>8,900</td>
<td>110</td>
<td>1.3%</td>
</tr>
<tr>
<td>BIP 2</td>
<td>572</td>
<td>305</td>
<td>DNAN</td>
<td>48,000</td>
<td>750</td>
<td>19%</td>
</tr>
<tr>
<td>BIP 3</td>
<td>575</td>
<td>301</td>
<td>RDX</td>
<td>18,000</td>
<td>280</td>
<td>2.8%</td>
</tr>
<tr>
<td>BIP 4</td>
<td>439</td>
<td>261</td>
<td>DNAN</td>
<td>130,000</td>
<td>1,200</td>
<td>53%</td>
</tr>
<tr>
<td>BIP 5</td>
<td>503</td>
<td>298</td>
<td>RDX</td>
<td>74,000</td>
<td>460</td>
<td>11%</td>
</tr>
<tr>
<td>BIP 6</td>
<td>617</td>
<td>321</td>
<td>DNAN</td>
<td>19,000</td>
<td>130</td>
<td>7.6%</td>
</tr>
<tr>
<td>BIP 7</td>
<td>901</td>
<td>420</td>
<td>RDX</td>
<td>7,600</td>
<td>46</td>
<td>1.1%</td>
</tr>
<tr>
<td>Means</td>
<td>586</td>
<td>312</td>
<td>DNAN</td>
<td>44,000</td>
<td>430</td>
<td>17%</td>
</tr>
</tbody>
</table>

*147 g RDX (plus 520 g RDX for BIP donor block) and 256 g DNAN Based on mid-range value of formulation specifications.

Table 4. Comparative Detonation Efficiencies RDX

<table>
<thead>
<tr>
<th>Scenario</th>
<th>Comp B Mass (mg)</th>
<th>Efficiency</th>
<th>PAX-21 Mass (mg)</th>
<th>Efficiency</th>
<th>IMX-104 Mass (mg)</th>
<th>Efficiency</th>
</tr>
</thead>
<tbody>
<tr>
<td>60 mm</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>High-order</td>
<td>0.073</td>
<td>99.9999%</td>
<td>9.2</td>
<td>99.993%</td>
<td>4.5</td>
<td>99.994%</td>
</tr>
<tr>
<td>BIP</td>
<td>200</td>
<td>99.97%</td>
<td>880</td>
<td>99.87%</td>
<td>20,000</td>
<td>98.6%</td>
</tr>
<tr>
<td>81 mm</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>High-order</td>
<td>8.5</td>
<td>99.999%</td>
<td>—</td>
<td>—</td>
<td>16</td>
<td>99.999%</td>
</tr>
<tr>
<td>BIP</td>
<td>150</td>
<td>99.98%</td>
<td>—</td>
<td>—</td>
<td>11,000</td>
<td>98.4%</td>
</tr>
</tbody>
</table>

SUMMARY AND CONCLUSIONS

The environmental performance of the PAX-21 and IMX-104 rounds has been shown to be inferior to the performance of comparable Comp B rounds. High-order detonations, while still containing low masses of both RDX and DNAN, were nevertheless higher than what can be expected of a high order detonation of a round containing conventional HE. The big difference, as we hypothesized, is in the BIP detonations, where three orders of magnitude difference in residues mass separate the conventional HE from the IHE results. Further investigation into BIP procedures for IM will need to be conducted before these results can be declared definitive.

SUBSEQUENT AND FUTURE WORK

Because of the high variability in the results of the 81-mm IMX-104 tests and the high deposition rates for the BIP operations, we reran the tests in March 2014, varying some of the
parameters. We increased the fuze simulator booster load from 12 g C4 to 18 g. For the BIPs, we consulted with the Armaments Research, Development and Engineering Center (ARDEC) Explosive Ordnance Disposal (EOD) specialist on site for the BIPs; he suggested that the C4 block be folded before it was attached over the supplemental charge on the rounds. Seven rounds were detonated for the BIP test and five for the high-order test. The plumes for the BIPs looked comparatively clean, lacking the greenish yellow tinge witnessed the previous year, and no yellow was obvious in the filtrate from the samples. We also observed no IMX particles on the filters during the lab processing. Analyses to determine the mass estimates are underway for these samples at the time of the writing of this manuscript. Further optimization of BIP procedures may be necessary to try to minimize energetics loading from these operations.

Live-fire exercises with these rounds followed by sampling the residues in the detonation plumes would verify the results from the high-order detonation tests. Past results for conventional HE-filled 81-mm mortar rounds indicated that there was no difference between live-fire and command detonations using the fuze simulators. Live-fire exercises would also generate data for estimates of low-order and dud rates for this new generation of munitions.

Tests such as those described in this paper to assess mass deposition should be conducted on all new rounds being developed for military use. Similar tests need to be conducted on the IMX-101 rounds nearing certification to ensure impacts to the environment will not adversely affect range sustainability. These test are relatively quick and can return results in a matter of a few weeks, giving a much better indication of not only the environmental loading of energetics from the use of the rounds but also how efficient the rounds are in the consumption of the energetic compounds in the explosive filler.

Integration of this testing into the life cycle environmental assessment of all new rounds needs to be investigated. Currently, the only residues regularly monitored for the Life Cycle Environmental Assessment (LCEA) process are in air emissions. A comparison of air emissions to residues deposition will indicate whether the current procedure adequately characterizes detonation residues. If not, mass deposition measurements from the rounds should be integrated into the life cycle environmental assessment.

ACKNOWLEDGEMENTS

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REFERENCES


5.0 Dissolution of NTO, DNAN and Insensitive Munitions Formulations and their Fates in Soils

Katerina Dontsova, PhD, Mark Brusseau, PhD, Jennifer Arthur, Noah Mark
University of Arizona
845 N. Park Avenue
Tucson, AZ 85721-0158

Susan Taylor, PhD, James Lever, PhD, PE, Marianne Walsh
Cold Regions Research and Engineering Laboratory
U.S. Army Engineer Research and Development Center
72 Lyme Road
Hanover, NH 03755-1290

Rose Pesce-Rodriguez, PhD
Army Research Laboratory (ARL), Weapons and Materials Research Directorate (WMRD)
U.S. Army Research Laboratory
Aberdeen Proving Ground, MD 21005-5006

Jiri Simunek, PhD
Department of Environmental Sciences
University of California, Riverside, CA 925021

ABSTRACT

The U.S. Military trains as it fights and uses live munitions during training. To save Soldiers lives both during training and war, the military is developing insensitive munitions (IM) that minimize unintentional detonations. New explosive compounds that are less sensitive to shock and high temperatures are being tested as replacements for TNT and RDX. Two of these explosives, DNAN (2,4-dinitroanisole) and NTO (3-nitro-1,2,4-triazol-5-one), have good detonation characteristics and are the main ingredients in a suite of IM explosives that are being, or soon will be, fielded. Both compounds, however, are more soluble than either TNT or RDX and have been shown to have some human and environmental toxicity. Data on their fate and transport is needed to determine if DNAN and NTO have the potential to reach groundwater and be transported off base, an outcome that could create future contamination problems on military training ranges and trigger regulatory action. In this study, we measured how quickly DNAN and NTO dissolve in IM formulations, and how solutions of these IM explosives interact with different types of soils. Because both dissolution and solution-soil interactions are determined by a suite of parameters we are using a multifaceted approach to study these processes. Given a mass of IM compounds scattered on the range, our work will help determine the dissolved IM masses, their subsequent transport and fate, and their likelihood of reaching groundwater.
INTRODUCTION

A series of steps is involved in transforming solid IM explosives, scattered onto range soils as a result of incomplete detonations, into solutions of explosive components that reach groundwater. These steps include dissolution, photo-transformation (both of the solid and of aqueous solutions formed by precipitation interacting with the solid), and complex interactions of the dissolved explosives with soil constituents and transport from the surface to the groundwater. If these compounds and their degradation products are innocuous, their transport to groundwater might not matter. Unfortunately toxicology data for DNAN and NTO show that while NTO has low mammalian toxicity (London and Smith, 1985), DNAN is more toxic than TNT to mammals (Davies and Provatas, 2006), can inhibit seed germination and plant growth, and even is used as a pesticide (Dumitras-Hutanu et al., 2009). It was also shown to be toxic to bacteria and earthworms (Doddard et al., 2013). Furthermore, both DNAN and NTO can form toxic transformation products (Le Campion et al., 1999; Davies and Provatas, 2006).

DNAN and NTO are readily soluble in water and their solubility is considerably higher than those of TNT and RDX (Table 1). When these compounds are part of a formulation, their dissolution will depend not only on their individual solubility and dissolution rates, but also on the fraction of each component exposed to water (Lever et al., 2005; Dontsova et al., 2006; Taylor et al., 2009b, a).

Table 1. Selected environmentally relevant chemical and physical properties of DNAN, NTO, TNT, and RDX, including solubility at 25 °C, octanol-water partitioning coefficient ($K_{ow}$), and soil organic carbon partitioning coefficient ($K_{oc}$).

<table>
<thead>
<tr>
<th>Property</th>
<th>DNAN ($a$)</th>
<th>NTO ($b$)</th>
<th>TNT ($c$)</th>
<th>RDX ($d$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Solubility, mg L$^{-1}$</td>
<td>276.2$^a$</td>
<td>16642.0$^b$</td>
<td>100.5$^c$</td>
<td>59.9$^d$</td>
</tr>
<tr>
<td>Log $K_{ow}$</td>
<td>1.7 – 1.92$^a$, 1.64$^d$</td>
<td>0.37 – 1.03$^b$, 1.6 – 1.84$^d$</td>
<td>0.81 – 0.87$^d$</td>
<td></td>
</tr>
<tr>
<td>Log $K_{oc}$</td>
<td>3.11$^a$, 2.2$^b$</td>
<td>3.03$^a$, 2.1$^b$</td>
<td>3.2$^c$, 3</td>
<td>0.88 – 2.4$^d$</td>
</tr>
</tbody>
</table>

$^a$ measured; $^b$ estimated; $^c$ interpolated from measured values.

Once in solution, DNAN, NTO, and their transformation products will experience reactive transport through the soil. En route to groundwater they can undergo irreversible and reversible adsorption by different mineral and organic phases in the soil, transformation, volatilization, and bio-uptake. The importance of each of these processes for the fate of a compound can be evaluated using the soil partition coefficient ($K_d$), soil organic carbon partition coefficient ($K_{oc}$), octanol-water partition coefficient ($K_{ow}$), Henry’s Law constant, the one-electron standard reduction potential, and transformation rate constant (k), among others. Values for some of these parameters for DNAN and NTO, as well as for common explosives, TNT and RDX, can be found in Table 1. Unlike most other explosives, which are polar but non-ionic compounds, NTO is an acid with pKa of 3.7-3.76 (Chipen et al., 1966; Le Campion et al., 1997), and will be negatively charged at environmentally relevant pHs (Smith and Cliff, 1999). As both organic and mineral soil surfaces tend to have net negative charge, we would predict low affinity of NTO for soils and greater mobility in the environment.
This paper describes: the dissolution of detonation residues of IM formulations (IMX-101, IMX-104, PAX-21) under controlled laboratory setting and in the field where they are exposed to rainfall and sunlight; the adsorption behavior of IM compounds in a range of soils; and the transport behavior of DNAN and NTO in representative soils. Combined these data will help us correlate their transport and fate behavior to soil properties.

RESULTS AND DISCUSSION

Dissolution

All tests particles came from partially detonated rounds and were collected in the field. Figure 1 shows optical images of DNAN, NTO and the IM formulations we studied along with the compositions of the formulations. Our experimental design is described in (Taylor et al., 2009a) and mimics field conditions on training ranges, where spatially isolated particles and chunks of explosives scattered on the soil surface by partial detonations are dissolved by rainfall. For the lab tests water was dripped on a well-characterized particle at a given rate (0.5 mL hr$^{-1}$ drip rate $= 0.55$ cm hr$^{-1}$ rainfall rate). For the outdoor tests the precipitation and temperature were measured with a weather station. No soil was involved in any of the tests allowing us to determine the dissolution as a function of the particle mass and the water volume interacting with particle.
The IM formulations differ from high explosives in several important ways. First, most of their crystal constituents are much more soluble than their DNAN matrices. Consequently, as NTO, Nitroguanidine (NQ) and AP dissolve they leave holes in the DNAN matrix (Fig. 2), increasing DNAN’s surface area and its dissolution rate. This is unlike Comp B where the RDX is less soluble than the TNT matrix. Second, because the solubility of NTO, NQ, AP and DNAN differ from each other by orders of magnitude, these compounds dissolve at different rates based on their solubility and the fraction of their surface exposed to water. We found that the dissolution of AP>NTO>NQ>DNAN>RDX. The 3D relationship of the constituents and their relative crystal sizes in IM formulations is, therefore, important for understanding how IM particles dissolve (Figure 2).

Figure 2. Micro computed tomography cross-sections of an IMX101, IMX104, and PAX21 particle after being wetted by 0, 1, 2, 8 and 12 mL of water and plots showing mass dissolved/ mass expected versus water volume for four particles of each formulation, one of which is shown.

We used micro computed tomography to image the three dimensional structure of IM formulations and learned that IM particles are fractured during detonation (Taylor et al. 2013). The fractures occur mainly in the matrix and go around the crystal boundaries of the NTO, NQ and AP crystals. We think that de-bonding of the crystals from the matrix during detonation is responsible for the large amount of AP scattered after high order detonations of PAX21 (Walsh et al. 2013) and predict that some fraction of the NTO and NQ will also survive high order detonations of IMX101 and IMX104 rounds (Taylor et al. 2013).
Our outdoor dissolution study shows that these IM formulations are much more friable than TNT or CompB. The particles crumb and break easily (Figure 3). These tests also suggest that photo-transformation of the IM formulations is occurring because: 1) the IM pieces changed color from cream or white to orange or brick red; 2) an unknown peak develops in the high performance liquid chromatograph (HPLC) where polar substance(s) elute; 3) this peak is largest in samples collected during the summer months when sun irradiance is highest; and 4) indoor (no sunlight) tests on IM explosives showed good mass balance (100±5%) and no unaccounted for HPLC peaks. As the tests are not yet complete we have not yet calculated a mass balance for these samples but we expect that it will be low because the IM compounds are transforming into products that are unknown and not analyzed.

NTO is known to be acidic in solution and we measured the pH of the both the drip and the outdoor water samples to quantify how this changed during dissolution of the particle. The first samples have pH values in the low 3 range that increased to neutral as the NTO concentration decreased to ~10mg/L (Fig. 4). There is a good correlation between NTO concentration and pH values between 2 and 5 for all samples. At NTO concentrations below 20mg/L the pH can vary between 4 and 6 and at NTO concentrations below 10mg/L the pH of the solution is close to neutral. The color of the solution was also found to correlate with the NTO concentration— the most yellow colored solutions had the highest NTO concentrations and the lowest pH values.
Phototransformation

Phototransformation of NTO was strongly affected by the pH of the solution with rates being highest at extreme pH values and lowest at neutral pH (Fig. 5, Table 2). In the environmentally-relevant range (4.2 - 8.8), alkaline conditions doubled the transformation rates over those seen under acidic conditions. Measured NTO phototransformation rates were higher than rates measured previously for RDX the compound that NTO is replacing in explosive formulations (Bordeleau et al., 2013). NTO phototransformation was also strongly affected by the presence of natural organic matter (humic acids) with phototransformation rates being almost three times higher in the presence of organic matter compared to rates measured when no organic matter was present (Fig. 5, Table 2).

DNAN phototransformation was higher than that measured for NTO, it was also higher than that measured before for TNT in pure water (O’Sullivan et al., 2011; Prak and O’Sullivan, 2012) (Fig. 5). Unlike TNT and NTO, that have been shown to be affected by organic matter in solution, DNAN phototransformation was not influenced by the presence of organic matter. Measured phototransformation rates for DNAN agreed well with published data (Rao et al., 2013).
Figure 5. Phototransformation of NTO as affected by solution pH (a) and the presence of natural organic matter (b).

Figure 6. Phototransformation of DNAN as affected by the presence of natural organic matter.

Table 2. Determined phototransformation rates, $k$, and half-lives, $t_{1/2}$, for NTO and DNAN measured at 35 °C, 765 W/m²

<table>
<thead>
<tr>
<th></th>
<th>NTO</th>
<th>DNAN</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>k (h⁻¹)</td>
<td>$t_{1/2}$ (days)</td>
</tr>
<tr>
<td>Direct</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1.5</td>
<td>0.294</td>
<td>0.10</td>
</tr>
<tr>
<td>4.2</td>
<td>0.156</td>
<td>0.19</td>
</tr>
<tr>
<td>7.0</td>
<td>0.084</td>
<td>0.34</td>
</tr>
<tr>
<td>8.8</td>
<td>0.306</td>
<td>0.10</td>
</tr>
<tr>
<td>Indirect</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7.0</td>
<td>0.246</td>
<td>0.12</td>
</tr>
</tbody>
</table>
A set of experiments was performed to determine the effect of temperature on phototransformation rates of NTO and DNAN and to calculate activation energies ($E_a$) of phototransformation reactions for these compounds. Phototransformation of DNAN was temperature dependent (Fig. 7) with $E_a = 27.8$ kJ/mol; measured $t_{1/2}$ of 0.13 days at 70°C, 0.23 days at 35 °C and predicted half-life of 0.50 days at 27°C. Determined activation energy ($E_a$) for DNAN was 27.8 kJ mol$^{-1}$. Knowing the activation energies of the phototransformation reactions allow us calculate transformation rates for local conditions. NTO transformation rates did not strongly depend on temperature indicating catalysis by decomposition products.

Figure 7. Effect of temperature on DNAN and NTO phototransformation rates. Natural logarithm of the phototransformation rate constant plotted as a function of temperature for DNAN and NTO

Soil Interactions

Eleven different soils representing a range of climates and conditions on active military ranges were used for this study. Ten of these soils were collected at military training ranges and Catlin silt loam was collected on the University of Illinois at Urbana-Champaign University farm (Figure 8). They had a wide range of physical and chemical properties (Table 3), and encompassed multiple soil orders with different moisture regimes, from very dry to wet.
NTO experienced very little adsorption in the studied soils. The largest observed $K_d$ was 0.51 cm$^3$ g$^{-1}$. For most soils linear adsorption isotherm described the observed adsorption well, confirming that $K_d$ can be used as a process descriptor for this compound (Table 4). Soil pH was

---

**Table 3. Measured physical and chemical properties of soils used in adsorption and transport studies with NTO, DNAN, and IM formulations**

<table>
<thead>
<tr>
<th>Soil</th>
<th>Texture</th>
<th>Clay</th>
<th>Silt</th>
<th>Sand</th>
<th>pH$^b$</th>
<th>EC$^b$</th>
<th>SSA$^c$</th>
<th>OC$^d$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Catlin</td>
<td>silt loam</td>
<td>25.6</td>
<td>65.5</td>
<td>8.9</td>
<td>7.31</td>
<td>492</td>
<td>6.4</td>
<td>5.28</td>
</tr>
<tr>
<td>Fort Harrison</td>
<td>sandy loam</td>
<td>8.7</td>
<td>36.5</td>
<td>54.9</td>
<td>6.67</td>
<td>449</td>
<td>7.4</td>
<td>3.88</td>
</tr>
<tr>
<td>Arnold AFB</td>
<td>silt loam</td>
<td>11.4</td>
<td>65.5</td>
<td>23.1</td>
<td>6.66</td>
<td>131</td>
<td>7.8</td>
<td>2.68</td>
</tr>
<tr>
<td>Plymouth</td>
<td>loamy sand</td>
<td>4.4</td>
<td>20.4</td>
<td>75.2</td>
<td>4.23</td>
<td>206</td>
<td>1.7</td>
<td>2.45</td>
</tr>
<tr>
<td>Camp Butner</td>
<td>sandy loam</td>
<td>7.7</td>
<td>25.9</td>
<td>66.4</td>
<td>6.69</td>
<td>219</td>
<td>4.8</td>
<td>2.42</td>
</tr>
<tr>
<td>Limestone Hills</td>
<td>sandy loam</td>
<td>11.2</td>
<td>35.7</td>
<td>53.1</td>
<td>7.54</td>
<td>539</td>
<td>10.5</td>
<td>1.99</td>
</tr>
<tr>
<td>Sassafras</td>
<td>loam</td>
<td>16.4</td>
<td>42.3</td>
<td>41.4</td>
<td>4.40</td>
<td>212</td>
<td>7.17</td>
<td>1.30</td>
</tr>
<tr>
<td>Camp Gruber</td>
<td>loamy sand</td>
<td>32.3</td>
<td>44.9</td>
<td>22.8</td>
<td>5.39</td>
<td>74</td>
<td>38.3</td>
<td>0.83</td>
</tr>
<tr>
<td>Camp Guernsey</td>
<td>loam</td>
<td>4.1</td>
<td>12.5</td>
<td>83.4</td>
<td>8.21</td>
<td>477</td>
<td>3.9</td>
<td>0.77</td>
</tr>
<tr>
<td>Florence MR</td>
<td>loam</td>
<td>26.8</td>
<td>33.5</td>
<td>39.7</td>
<td>8.00</td>
<td>417</td>
<td>33.0</td>
<td>0.45</td>
</tr>
<tr>
<td>Camp Swift</td>
<td>sandy clay</td>
<td>23.7</td>
<td>20.8</td>
<td>55.6</td>
<td>7.83</td>
<td>203</td>
<td>15.1</td>
<td>0.34</td>
</tr>
</tbody>
</table>

$^a$In 1:1 soil:water; $^b$EC=Electrical conductivity; $^c$SSA = specific surface area; $^d$OC = organic carbon.
the strongest indicator of NTO affinity for soil surfaces (Fig. 9). There was no relationship observed between \( K_{d,s} \) and other measured soil properties, such as OM, clay, or SSA. NTO is an ionic compound and at most of environmentally-relevant pH values the –N-H groups are dissociated, imparting a negative charge on the molecule. Dissociation of these groups increases with increasing pH. Because most soils also have net negative charge that increases with soil pH, NTO adsorption decreases with increasing pH of the soils. Compared to RDX, NTO adsorbed much less to high OM neutral soils (Dontsova et al., 2009), but more to soils with low pH and %OC (Dontsova et al., 2006).

Measured transformation rates were strongly related to the amount of organic carbon in soils. There was a highly significant (\( P=0.02 \)) positive relationship between percent OC present in the soil and the measured transformation rate \( k \); however, \( R^2 \) was low at 0.46 (Fig. 9). Half-life values ranged between 1.3 and 72.2 days.

![Figure 9. Measured NTO adsorption coefficients (\( K_{d,s} \)) plotted against soil pH (\( P = 0.00011 \)) and transformation rates (\( k_s \)) plotted as a function of soil OC (\( P = 0.02250 \))](image)

Table 4. Fate and transport parameters for NTO in studied soils: Freundlich adsorption parameters, \( K_f \) and \( n \), linear adsorption coefficient, \( K_d \), adsorption coefficient normalized to fraction of organic carbon in soils, \( K_{oc} \), transformation rate, \( k \), \( R^2 \) values for the fits, and half-lives, \( t_{1/2} \)

<table>
<thead>
<tr>
<th>Soil</th>
<th>( K_f ) (cm(^3) g(^{-1}))</th>
<th>( n )</th>
<th>( R^2 )</th>
<th>( K_d ) (cm(^3) g(^{-1}))</th>
<th>( R^2 )</th>
<th>( K_{oc} ) (cm(^3) g(^{-1}))</th>
<th>( k ) (h(^{-1}))</th>
<th>( R^2 )</th>
<th>( t_{1/2} ) (days)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Catlin</td>
<td>0.21</td>
<td>1.03</td>
<td>0.94**</td>
<td>0.21</td>
<td>0.92**</td>
<td>4.0</td>
<td>0.0221</td>
<td>0.84*</td>
<td>1.3</td>
</tr>
<tr>
<td>Fort Harrison</td>
<td>0.27</td>
<td>1.07</td>
<td>0.98**</td>
<td>0.35</td>
<td>0.95**</td>
<td>9.0</td>
<td>0.0021</td>
<td>0.98**</td>
<td>13.8</td>
</tr>
<tr>
<td>Arnold AFB</td>
<td>0.58</td>
<td>0.86</td>
<td>0.98*</td>
<td>0.34</td>
<td>0.94**</td>
<td>12.7</td>
<td>0.0044</td>
<td>0.94**</td>
<td>6.6</td>
</tr>
<tr>
<td>Plymouth</td>
<td>0.82</td>
<td>0.89</td>
<td>0.99</td>
<td>0.5</td>
<td>0.96**</td>
<td>20.4</td>
<td>0.0043</td>
<td>0.97**</td>
<td>6.7</td>
</tr>
<tr>
<td>Camp Butner</td>
<td>0.74</td>
<td>0.54</td>
<td>0.77</td>
<td>0.12</td>
<td>0.72**</td>
<td>5.0</td>
<td>0.0021</td>
<td>0.98**</td>
<td>13.8</td>
</tr>
<tr>
<td>Limestone Hills</td>
<td>0.33</td>
<td>0.88</td>
<td>0.92**</td>
<td>0.21</td>
<td>0.92**</td>
<td>10.6</td>
<td>0.0123</td>
<td>0.60*</td>
<td>2.3</td>
</tr>
<tr>
<td>Sassafras</td>
<td>0.9</td>
<td>0.86</td>
<td>0.99</td>
<td>0.48</td>
<td>0.96**</td>
<td>36.9</td>
<td>0.008</td>
<td>0.99**</td>
<td>3.6</td>
</tr>
<tr>
<td>Camp Gruber</td>
<td>0.54</td>
<td>0.99</td>
<td>1.00**</td>
<td>0.51</td>
<td>0.99**</td>
<td>61.5</td>
<td>0.0025</td>
<td>0.98**</td>
<td>11.6</td>
</tr>
<tr>
<td>Camp Guernsey</td>
<td>0.06</td>
<td>0.48</td>
<td>0.18*</td>
<td>0.02</td>
<td>0.21*</td>
<td>2.6</td>
<td>0.0004</td>
<td>0.75*</td>
<td>72.2</td>
</tr>
<tr>
<td>Florence MR</td>
<td>0.09</td>
<td>0.77</td>
<td>0.55*</td>
<td>0.06</td>
<td>0.57*</td>
<td>13.3</td>
<td>0.0005</td>
<td>0.91**</td>
<td>57.8</td>
</tr>
<tr>
<td>Camp Swift</td>
<td>0.1</td>
<td>0.84</td>
<td>0.92*</td>
<td>0.04</td>
<td>0.59**</td>
<td>11.8</td>
<td>0.0009</td>
<td>0.93**</td>
<td>32.1</td>
</tr>
</tbody>
</table>
DNAN exhibited much greater soil adsorption than NTO (Table 5, Fig. 10). Sorption was non-linear, with affinity of DNAN for soil surfaces decreasing as concentration increased due to saturation of the adsorption sites, similar to nitroaromatics, such as TNT and DNT. This becomes particularly relevant when high concentration pulses of DNAN are released during particle dissolution. Adsorption was positively correlated with amount of organic carbon in the soils (Fig 11). Average measured log $K_{OC}$ for studied soils was 2.3. DNAN transformation rates were significantly slower than ones observed for TNT (Donsova et al., 2006). Half-lives ranged between 4 and 48 days.

![Figure 10. DNAN adsorption isotherms for Catlin (OC = 5.28%) and Sassafras (OC = 1.30%) soils. Dashed lines represent fits of linear and solid lines Freundlich isotherms to the adsorption data. Mass balance of DNAN for the same soils with and without treatment to sterilize soils](image)

Table 5. Fate and transport parameters for DNAN in studied soils: Freundlich adsorption parameters, $K_f$ and $n$, linear adsorption coefficient, $K_d$, adsorption coefficient normalized to fraction of organic carbon in soils, $K_{OC}$, transformation rate, $k$, $R^2$ values for the fits, and half-lives, $t_{1/2}$

<table>
<thead>
<tr>
<th>Soils</th>
<th>$K_f$</th>
<th>n</th>
<th>$R^2$</th>
<th>$K_d$</th>
<th>$R^2$</th>
<th>$K_{OC}$</th>
<th>k</th>
<th>$R^2$</th>
<th>$t_{1/2}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Catlin</td>
<td>45.61</td>
<td>0.57</td>
<td>0.82</td>
<td>6.06</td>
<td>0.46</td>
<td>114.7</td>
<td>0.0021</td>
<td>0.85</td>
<td>14</td>
</tr>
<tr>
<td>Fort Harrison</td>
<td>13.86</td>
<td>0.77</td>
<td>0.9</td>
<td>6.32</td>
<td>0.93</td>
<td>162.9</td>
<td>0.0010</td>
<td>0.45</td>
<td>29</td>
</tr>
<tr>
<td>Arnold AFB</td>
<td>14.49</td>
<td>0.68</td>
<td>0.93</td>
<td>3.39</td>
<td>0.78</td>
<td>126.5</td>
<td>0.0022</td>
<td>0.92</td>
<td>13</td>
</tr>
<tr>
<td>Plymouth</td>
<td>10.05</td>
<td>0.83</td>
<td>0.98</td>
<td>4.38</td>
<td>0.89</td>
<td>178.6</td>
<td>0.0070</td>
<td>0.51</td>
<td>4</td>
</tr>
<tr>
<td>Camp Butner</td>
<td>15.35</td>
<td>0.56</td>
<td>0.97</td>
<td>2.05</td>
<td>0.87</td>
<td>84.8</td>
<td>0.0018</td>
<td>0.67</td>
<td>16</td>
</tr>
</tbody>
</table>

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Workshop Proceedings 
May 2014
Transport in Soils

In agreement with the batch tests, NTO experienced very little retardation in the soil columns. The retardation factor was less than 2 indicating that NTO will move to the groundwater at a velocity similar to that of water inflow. Recovery of NTO in solution, after NTO pulse moved through the soil column and NTO was no longer detectable in outflow, ranged between 96 and 39% (Table 6) indicating that in-situ transformation contributes to natural attenuation of this IM. The amount recovered decreased as the organic carbon content in the soil increased ($R^2 = 0.90$).

Breakthrough curves for soils with high OC content showed evidence of 2nd-order transformation rate, where the rate increased with time. Because this trend was not present if the soil was autoclaved before the tests we attribute the trend to microbial growth and resulting O$_2$ depletion in the soil.
Figure 12. Breakthrough curves for NTO in Camp Guernsey soil, with and without flow interruption (FI), and Catlin soil with and without treatment to sterilize the soil. Dashed grey vertical line indicates timing of 24-hour flow interruption. Tan vertical line indicates the time when the solution was reverted to a saturating solution to observe desorption phase of the isotherm.

Table 6. Percent recovery of NTO and bromide tracer in column transport experiments. Mean values are for two replications.

<table>
<thead>
<tr>
<th>Soil</th>
<th>OC, %</th>
<th>NTO</th>
<th>StDev</th>
<th>Bromide</th>
<th>StDev</th>
</tr>
</thead>
<tbody>
<tr>
<td>Catlin</td>
<td>5.3</td>
<td>39.4</td>
<td>1.1</td>
<td>103.9</td>
<td>8.8</td>
</tr>
<tr>
<td>Arnold AFB</td>
<td>2.7</td>
<td>52.5</td>
<td>2.5</td>
<td>101.7</td>
<td>6.8</td>
</tr>
<tr>
<td>Butner</td>
<td>2.4</td>
<td>59.8</td>
<td>0.3</td>
<td>100.8</td>
<td>0.9</td>
</tr>
<tr>
<td>Limestone Hills</td>
<td>2.0</td>
<td>67.1</td>
<td>8.4</td>
<td>99.0</td>
<td>0.7</td>
</tr>
<tr>
<td>Sassafras</td>
<td>1.3</td>
<td>85.6</td>
<td>4.8</td>
<td>103.4</td>
<td>5.9</td>
</tr>
<tr>
<td>Camp Guernsey</td>
<td>0.77</td>
<td>93.2</td>
<td>0.3</td>
<td>103.3</td>
<td>5.2</td>
</tr>
<tr>
<td>Florence MR</td>
<td>0.45</td>
<td>94.4</td>
<td>0.6</td>
<td>109.0</td>
<td>9.3</td>
</tr>
<tr>
<td>Camp Swift</td>
<td>0.34</td>
<td>95.9</td>
<td>4.2</td>
<td>103.0</td>
<td>2.1</td>
</tr>
</tbody>
</table>

Dissolution and transport of IM constituents from IMX-101 formulation (both manufactured and detonation residues) showed trends observed in dissolution experiments without soils and in soil adsorption and transport experiments. Concentrations observed were very high, with NTO appearing in the gram per liter range, followed by NQ, and DNAN, in agreement with their solubilities. Breakthrough of constituents also followed the same trend. NTO appears first in the
outflow followed by NQ, which was weakly adsorbed and finally DNAN. An amino-
transformation product of DNAN, 2-methoxy-5-nitroaniline (MENA) was also observed, but at a
much lower concentration than DNAN. No difference in dissolution was observed between
detonated and undetonated samples (Fig. 13).

![Figure 13. Breakthrough curves for NTO, NQ, DNAN, as well as DNAN transformation
product, MENA, from IMX 101 particle dissolution in Camp Swift soil: a) undetonated
particles, and b) residues of low order detonation. Flow rate is 0.01 mL min⁻¹. The black
vertical line marks the time, when the IMX 101 particles were removed from the soil
surface to observe desorption phase of the breakthrough](image)

**SUMMARY AND CONCLUSIONS**

We found that the components in these explosive formulations dissolve sequentially and in
the order predicted by their solubility. The good mass balances measured for the laboratory drip
tests indicate that the formulations are not being significantly photo-or bio-transformed under
laboratory conditions. IMX101 and IMX104 both contain NTO, which is very soluble and
produces low pH solutions. Due to NTOs solubility, the composition, concentration and pH of
water solutions entering the soil will vary with time as NTO-containing formulations dissolve.
Photo-transformation was observed in solutions and in solid IM particles under sunlight.

NTO was not significantly adsorbed by soils, while DNAN experienced soil adsorption that
was increasing with increase in soil OC (average log K_OC = 2.3). NTO half-lives ranged between
1.3 and 72 days and transformation rate increased with soil OC content; DNAN half-lives ranged
between 4.1 and 48 days. The high solubility of some of the IM constituents and their low
reactivity with the soil suggests that they have a high potential for reaching groundwater.

**ACKNOWLEDGMENTS**

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Armament Research, Development and Engineering Center (ARDEC), Picatinny Arsenal and
Michael Walsh, project SERDP ER-2219 for providing IM formulations and residues of low-
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Milose, Amibeth Sheridan, Laurie Stenberg, Sarah Gettier, Army National Guard–
Environmental Directorate and URS Corporation, Germantown, MD and Mike Heitmann,
CH2M HILL Englewood, CO for making possible collection of soils used in the experiments on Army National Guard installations.

REFERENCES


Spear, R.J., Louey, C.N., Wolfson, M.G., 1989. A Preliminary Assessment of 3-Nitro-1,2,4-Triazol-5-One (NTO) as an Insensitive High Explosive. DSTO Materials Research Laboratory, Maribyrnong, Australia, p. 38.


6.0 Factors Controlling Partitioning of Munition Constituents in Soil

R. Gonzalez, A. Miglino, D.M. DiToro, and H.E. Allen, University of Delaware, Newark, DE.

ABSTRACT (no manuscript received)

We studied adsorption and desorption of 6 munitions constituents (MCs) on a large number of soils with different properties and developed models describing the partitioning. Literature focuses on whether MC partitioning is controlled by organic matter or clay in soil. Our partitioning model incorporates both organic matter and clay with predictive capability significantly better than a model considering only one of the factors. Three different approaches have been tested to quantify the clay component: clay particle size, cation exchange capacity, and the sorption of cesium ions. The addition of iron oxides to the model provides further improvement. Desorption isotherms of many MCs from soil do not follow the adsorption isotherm. A portion of the adsorbed material resists desorption. The reversible/resistant model indicates that a large fraction of many compounds are not recovered in typical desorption tests. Our model assumes rapid equilibrium between soil solution and a surficial soil layer. Mass transfer resistance, controlled by diffusion, limits the rate of release of MC from the core of a soil particle. Although complete reversibility for the sorption process is assumed, the mass transfer-limited rate of release limits soil solution concentrations.
7.0 Development of an Environmental Fate Simulator for New and Proposed Compounds Military-unique Munition Compounds

Caroline Stevens¹, Kurt Wolfe¹, Rajbir Parmar¹, Mike Galvin¹, Gene Whelan¹, Said Hilal¹, Mitch Pelton², and Eric Weber¹

¹U.S. Environmental Protection Agency, Athens, GA
Ecosystems Research Division
National Exposure Research Laboratory
U.S. Environmental Protection Agency
Athens, GA
²Pacific Northwest National Laboratory, Richland, WA

ABSTRACT

The focus of this work is the development of an Environmental Fate Simulator (EFS) that will provide managers of military training and testing ranges with parameter estimates to assess the vulnerability of aquifers and surface waters to new and proposed energetic materials and their potential transformation products. Our working hypothesis is that there is a substantial amount of process science that has been published in the peer-reviewed literature concerning the transport and transformation of existing N-based munitions (e.g., TNT, 2,4-DNT and RDX) and related N-based chemicals (i.e., nitro aromatics, aromatic amines and substituted azobenzenes) that can be encoded through the use of cheminformatic applications. This process science can be leveraged to predict the transport and transformation of the emerging N-based munitions for which little fate data exists. The EFS provides the calculated physico-chemical (p-chem) properties of the parent chemical and transformation products, which are predicted as a function of the reaction system of interest. This is accomplished through the integration of cheminformatics applications for the encoding of process science underlying transformation pathways, computational chemistry tools for the calculation of physico-chemical properties, and software technologies that provide access to on-line databases for environmental descriptors required for estimating environmental concentrations.

INTRODUCTION

DoD is responsible for assessing the environmental exposure resulting from the testing and training activities associated with military munitions and other chemical constituents of concern. Of greatest concern is potential for off-site exposure to these materials and their degradation products primarily as a result of movement through surface waters and underlying aquifers. Modeling systems and databases currently exist where the user is responsible for defining the individual chemicals and their properties necessary to conduct chemical exposure and risk assessments. These data are required input into the U.S. Army Groundwater Modeling System (GMS) (http://chl.erdc.usace.army.mil/gms), the Adaptive Risk Assessment Modeling System (ARAMS) (http://el.erdc.usace.army.mil/arams/), and the Field Demonstration and Validation of the Training Range Environmental Evaluation and Characterization System (TREECS™) (http://el.erdc.usace.army.mil/TREECS/).
The objective of this work is to develop a web-based EFS that will provide the managers of military training and testing ranges with a tool to support vulnerability assessments of aquifers and surface waters to new and proposed energetic materials and their potential transformation products. The work will provide physico-chemical properties of parent and progeny for the eventual seamless consumption by modeling toolsets for assessing environmental exposure and subsequent human/ecological receptor health risks associated with loading and fate/transport of residual energetic materials (and their degradation products).

A major concern for assessing the environmental fate and transport of unexploded ordinances, other military relevant compounds (MRCs), and new military-unique munition compounds is the availability of the chemical and physical properties for not only the parent compounds but also for the degradation products. This is especially relevant at live fire ranges and impact zones, where the potential transport and offsite exposure of residual energetic materials are of potential concern. To investigate the environmental risk posed by new munition compounds, improved methods for the prediction of their physico-chemical properties are required, as they relate to their potential movement and impacts in a multiple media environment (e.g., aquifer, surface water, air, and soil). Coupled with improved predictive techniques for determining probable reaction pathways, this information allows for the assessment of media vulnerability and resulting exposure and risks to humans and ecological receptors, thus supporting defense environmental management and compliance requirements, as well as aiding in any fire range and impact zone sustainment. By providing the means to assess transport, exposure, and risk, these tools will provide endpoint metrics for exposure concentrations and health risks that can be compared to thresholds for determining if there is reason for concern, when this will occur, and to what level of severity. As an added benefit, these environmental data can be coupled with range-use characteristics to assess material loading, fate, environmental transport, exposure, and receptor risks, on- and off-site with quantified uncertainty. These tools could provide the information needed for managing range use and determining if, when, and to what extent ranges must be characterized, cleared or remediated.

RESULTS AND DISCUSSION

The EFS is composed of six major components as described below. The data flow diagram for the EFS based on these major components is illustrated in Figure 1. The user input is the chemical of interest. The user then has the option of executing the Reaction Pathway Simulator based on the environmental media of interest to generate potential transformation products, or execution of the Physicochemical Properties Calculator (PPC) to generate molecular descriptors for the parent chemical. The output of the PPC is stored in the Structure-Based Database. The user will have the option to generate reaction rate constants through the implementation of quantitative structure-activity relationships (QSARs) based molecular descriptors (e.g., pKa values or one electron reduction potentials stored in the database) and environmental descriptors (e.g., pH and Fe(II) concentrations) as a function of geographical location through execution of the Earth System Model. The calculated first-order rate constants will then be entered seamlessly into the database.
Description of the Major Components of the EFS

**Chemical Editor (CE):** The CE gives the user the ability to enter the chemical(s) of interest by providing a chemical structure, SMILES string, CAS# or common name. The entry of 2,4-dinitroanisole (2,4-DNAN) by drawing its chemical structure is illustrated in Figure 2.

**Reaction Pathway Simulator (RPS):** Based on chemical structure analysis, the RPS provides the dominant transformation products as a function of environmental conditions. The environmental conditions are defined by the user according to respiration conditions (aerobic vs. anaerobic) and the reaction media of interest (Figure 3). In this example, which is based on the selection of an anaerobic benthic sediment, the user would have the option to execute either (or both) the reaction libraries for abiotic hydrolysis or reduction.
Figure 3. User selects the transformation pathways to be executed through the selection of respiration conditions and the reaction medium of interest

The output of the RPS is based on the selection and execution of libraries of reaction schemes that represent one-step reactions for transformation (e.g., reduction or hydrolysis) of reactive functional groups based on peer-reviewed process science published in the literature. These reaction schemes are used to identify viable transformation products through the identification and subsequent transformation of reactive functional groups. Each scheme within a reaction library is assigned an integer rank ranging between 1 and 5 based on the relative rate of transformation. For molecules that are subject to more than one transformation pathway, these ranks are used to approximate the relative fractional formation of products. The functional group transformations illustrated in Figure 4 represent those most often observed for N-based munitions and other classes of chemicals of concern to range managers (e.g., halogenated solvents).

**Figure 4. Illustrated examples of reductive transformations representing one-step reactions for the transformation of reactive functional groups**
Physicochemical Properties Calculator (PPC): The PPC provides the necessary molecular descriptors through seamless linkage to existing p-chem calculators. The development of the PPC is based on a consensus approach for the calculation of p-chem properties that allows the user to compare output generated by a number of calculators that take different approaches to calculating specific physicochemical properties (Table 1). When fully functional, the user will have access to four p-chem calculators:

- SPARC (SPARC Performs Automated Reasoning in Chemistry) [1, 2] which uses a mechanistic-based approach
- EPI Suite™[3], which uses a fragment-based approach
- TEST (Toxicity Estimation Software Tool) [4, 5], which uses QSAR-based approaches through the calculation of structural, topological and electrostatic descriptors
- ChemAxon plug-in calculators [6], which use a blend of mechanistic- and QSAR-based approaches

The output from these calculators will give the user the ability to compare the calculated data with readily accessible measured data in web-based databases.

<table>
<thead>
<tr>
<th>Select Chemical Specific Parameters</th>
<th>ChemAxon</th>
<th>EPI Suite</th>
<th>TEST</th>
<th>SPARC</th>
</tr>
</thead>
<tbody>
<tr>
<td>All</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Melting Point (°C)</td>
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<td></td>
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<tr>
<td>Boiling Point (°C)</td>
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<td></td>
</tr>
<tr>
<td>Water Solubility (mg/L)</td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Vapor Pressure</td>
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<tr>
<td>Molecular diffusivity in water</td>
<td></td>
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</tr>
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<td>Ionization constant</td>
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<td>Henry's Law Constant (atm-m³/mol)</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Log Octanol Water Partition Coefficient</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Log Octanol Water Partition Coefficient (pH dependent)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Available
Not Available

Structure-Based Database (SBD): The SBD provides for the storage of the calculated and measured physicochemical properties required for estimating environmental concentrations (Table 2). The SBD can be searched by structure to compare molecules that contain the same structural fragments.
Table 2. An example spreadsheet for 2,4-DNAN and its potential reduction products. Computed values are shown for log Kow and pKa for each of the calculators where applicable

<table>
<thead>
<tr>
<th>Structure</th>
<th>Compound Name</th>
<th>Smiles String</th>
<th>Gas X</th>
<th>Octanol Water Partition Coefficient (log Kow)</th>
<th>Isolable Constant (pKa)</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="https://via.placeholder.com/150" alt="Image" /></td>
<td>2,4-DNAN</td>
<td>c1[Oc]<a href="=O">N</a>0]0)c1)0)0)c1</td>
<td>19277</td>
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<td>1.71</td>
<td>2.70</td>
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<tr>
<td><img src="https://via.placeholder.com/150" alt="Image" /></td>
<td>2-methoxy-5-nitroaniline</td>
<td>c1[Oc]<a href="=O">N</a>0]0)c1</td>
<td>99592</td>
<td>1.91</td>
<td>1.47</td>
<td>0.93</td>
</tr>
<tr>
<td><img src="https://via.placeholder.com/150" alt="Image" /></td>
<td>4-methoxy-3-nitroaniline</td>
<td>c1[Oc]<a href="=O">N</a>0]0)c1)0)c1</td>
<td>57720</td>
<td>2.99</td>
<td>1.55</td>
<td>0.93</td>
</tr>
<tr>
<td><img src="https://via.placeholder.com/150" alt="Image" /></td>
<td>4-methoxy-1,3-diaminobenzene</td>
<td>c1[Oc][N]0]0)c1</td>
<td>015054</td>
<td>-0.81</td>
<td>-0.31</td>
<td>0.36</td>
</tr>
</tbody>
</table>

**Environmental Systems Model (ESM):** The ESM provides the necessary environmental descriptors through linkage to web-accessible databases. This accomplished primarily through the execution of Data for Environmental Modeling (D4EM), which is an open source software system consisting of a library of utilities that can be used to access, retrieve and process model data automatically from sources on the internet. Figure 5 illustrates an example where D4EM was used to retrieve water quality parameters (e.g., water temperature and pH) from the USGS National Water Quality database (NAWQA) (http://water.usgs.gov/nawqa/) at a specific site based on a specified latitude/longitude provided by the user.

**Reaction Rate Calculator (RRC):** The RRC predicts transformation product formation based on the parameritization and execution of QSARs and algorithms.
Figure 5. An example of environmental descriptors that were accessed through D4EM from the USGS Water Quality database at a specific site based on a specified latitude/longitude

The parameterization of QSARs stored in Reaction Rate Calculator with the required molecular descriptors is accomplished through execution of the PPC. An example of this linkage is provided for aromatic amines that are known to sorb irreversibly to soil and sediments. Laboratory studies have provided evidence that this process occurs predominantly through 1,4-nucleophilic addition to quinone moieties found in the natural organic matter (NOM) [7] as illustrated for the nucleophilic addition of aniline to benzoquinone in Figure 6. In subsequent studies, QSARs for this covalent binding process were developed based on kinetic studies of the reaction of a series of anilines with a substituent in the 2-, 3-, or 4-position in an aerobic pond sediment [8]. The most robust QSAR generated for estimating the rate of this reaction process was based on $pK_a$ constants, which are a measure of a chemical’s nucleophilicity (Figure 6).

\[
\log k = \left( -0.411(\pm0.04) \right) pK_a - 3.14(\pm0.13)
\]

Figure 6. A plot of log $k$ for the irreversible binding to pond sediment versus $pK_a$ values for the nucleophilic addition of a series of mono-substituted anilines

Parameterization of the QSAR generated from the kinetic data is possible through execution of the ChemAxon $pK_a$ calculator, which is one of the calculators accessible through the PPC. An example of the calculation of $pK_a$ values for the aromatic amines that form from the reductive
transformation of nitroaromatics is shown in Figure 7 below. Reduction of 2,4-dinitroanisole (DNAN), which is one of the next generation low shock sensitive munitions, has been shown to be reduced in sediments to form the mono aminonitroanisoles and subsequently the diaminoanisole [9]. The calculated pKₐ values are shown adjacent to the amino groups. Based on the assumption that these aromatic amines have similar reactivity to the monosubstituted anilines and through parameritization of the QSAR with the calculated pKₐ values, we are able to determine that irreversible sorption will not become a significant process in aerobic sediments until reduction of both nitro groups has occurred resulting in the diaminoanisole with the amino group in the 4-position with a pKₐ value of 5.71.

**Figure 7. Reduction of the nitro groups in 2,4-dinitroanisole (DNAN) resulting in the formation of aromatic amines and covalent binding in soil and sediment systems**

**Workflow Options**

The selection and order of execution of the major components of the EFS is based on the user’s choice of one of three available workflows as described below.

1. **Generate Chemical Structure Information Workflow:**

   This workflow allows the user to enter an individual chemical through the Chemical Editor. Based on the selection of the available options for structure analysis, the user will be able to generate structure information including the dominant forms of ionized species and tautomers, as well as structures for possible stereoisomers.
2. **Calculate P-Chem Properties Workflow:**

   This workflow allows the user to enter an individual chemical through the Chemical Editor or through submission of a text file for multiple chemicals (i.e., batch mode). Based on the selection of the available p-chem calculators and p-chem properties, the user will be able to generate calculated p-chem properties of interest.

3. **Predict Transformation Product Formation Workflow:**

   This workflow allows the user to predict transformation products predicted based on the selection and execution of the reaction libraries that encode the process science for transformation processes. The user has the option to enter an individual chemical through the Chemical Editor or through batch mode. Entry into the RPS will prompt the user to select the reaction library of interest based on the selection of reaction conditions as described previously in Figure 3. The user will have the option to calculate p-chem properties for the predicted transformation products and reaction rate constants subject to the availability of QSARs.
SUMMARY AND CONCLUSIONS

The development of the major components of the EFS based on cheminformatics applications, as well as the integration of software technologies such as D4EM, has provided the required capabilities for each of these components. The use of ChemAxon software tools and the associated plug-in calculators provide the ability to encode the existing process science in the peer-reviewed literature, as well as the process science that is being generated from on-going SERDP projects. We are just beginning to explore the full potential of the plug-in calculators for the development of reaction rules governing reactivity and selectivity in support of the reaction schemes for transformation of reactive functional groups. Our sense moving forward is that we are no longer limited by the availability of technology, but by the lack of the process science supporting the environmental fate on the new munition compounds.

FUTURE WORK

A key challenge for us this coming year is to develop an automated process for the parameterization of QSARs for the estimation of first-order rate constants, which can then be used for calculating the percent formation of the transformation products in the RPS. This will allow us to move from the qualitative approach of assigning ranks for individual reaction schemes to a more quantitative approach based on the estimation of first order rate constants.

ACKNOWLEDGEMENTS

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REFERENCES


8.0 Chemical and Biological Degradation of Insensitive Munitions (IM) Mediated by Fe(III)-reducing Microorganisms

Kevin T. Finneran and Jolanta B. Niedzwiecka
Clemson University Environmental Engineering and Earth Sciences, Clemson, SC

ABSTRACT

The IM 2,4-dinitroanisole (DNAN) and nitroguanidine (NQ) were investigated to determine the capacity for biodegradation using mixed chemical-biological reactions mediated by Fe(III)-reducing microorganisms. Both were rapidly degraded by ferrous iron at pH 7, 8, and 9, with rates increasing as the pH increased. The electron shuttle anthrahydroquinone-2,6-dilsulfonate (AH$_2$QDS) degraded DNAN, but not NQ. DNAN degraded in one hour with AH$_2$QDS and ferrous iron at pH 8 and 9; >90% of the initial 100 micromolar DNAN was degraded in several minutes, suggesting that this pathway will be exceptionally effective at DNAN removal depending on pH. Rates were slower at pH 7, but DNAN degraded within 36 hours. The primary product of DNAN degradation by Fe(II) was 2-methoxy-5-nitroaniline (MENA), recovered at approximately 60-75% of initial DNAN carbon. 2,4-diaminoanisole (DAAN) was also detected, but at a lower final percentage. NQ degradation was much slower, with complete degradation taking 30 days at pH 8 and pH 9. Unlike DNAN, NQ degradation required solid surfaces (primarily as aggregated Fe(OH)$_2$(s)) for reduction – soluble ferrous iron alone did not reduce NQ. NQ reduction products have not yet been identified, but work continues to characterize the NQ degradation pathway. The ferrous iron ligand, trihydroxybutyric acid (THBA), increased DNAN and NQ reduction at pH 7 and 8, respectively. THBA amendment may be a reasonable alternative for ex situ reactions, where it cannot other metals. The Fe(III)-reducing bacterium Geobacter metallireducens also reduced DNAN. The cells reduced the compound directly, and via electron shuttling reactions with iron and extracellular quinone/hydroquinone couples.

INTRODUCTION

The Department of Defense has developed several new explosive composites that contain typical compounds such as the cyclic nitramines but that have been updated with insensitive munitions including 2,4-dinitroanisole (DNAN). DNAN is being used as a replacement for trinitrotoluene (TNT) in explosive formulations due to its higher stability and increased safety standards for explosives transport and storage. Nitroguanidine and 3-nitro-1,2,4-triazole-5-one (NTO) are another new insensitive munitions, which are evaluated to replace commonly used compounds, such as RDX and octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine (HMX).

RDX and HMX have been detected at many military sites and live-fire training installations [1]. When dissolved they can migrate in subsurface and cause groundwater contamination. RDX is a possible human carcinogen and HMX is toxic to central nervous system, hence their presence in groundwater is a concern. DNAN has not yet been detected at any major military installations; however, the DOD requires crucial information on bioremediation possibilities should it become an issue.
The most common in-situ strategies for explosive remediation include pump-and-treat systems and permeable reactive barriers using zero valent iron [1]. Pump-and-treat systems do not remove the cause of contamination and they involve pumping large volumes of groundwater; therefore, their effectiveness is limited. Permeable reactive barriers may be a good solution but only for a shallow plume. When the plume is located deep under the surface, the installation of reactive barrier and related costs can become an issue. Alternatively, in-situ bioremediation presents a big potential as the cheapest and most effective method. However, in this case, it is important to understand geochemical processes in subsurface and to analyze the microbial community present at the site.

Nitrated compounds have a strong electron-withdrawing effect, and as a result they are degraded by reduction rather than oxidation reactions. It has been reported that ferrous[2, 3] iron reduction mediates degradation of polyhalogenated alkanes [4] and nitroaromatic compounds such as pesticides [5] and munitions (19, Borch T, Inskeep WP, Harwood JA, Gerlach R. 2005). Also, other reduced extracellular electron shuttles like humic substances, organically complexed ferrous iron species or quinones can be employed for effective remediation of RDX and HMX [2, 3]. In order to establish reducing conditions, Fe(III)-reducing microorganisms must be present in the soil. These organisms are common in subsurface environment and, when active, they can mediate electron shuttles reduction.

As a result, reduced shuttles can transfer electrons to the contaminant and degrade it. The abundance of both electron shuttles and Fe(III)-reducing microorganisms demonstrates a great potential for development of effective on-site degradation strategies for different explosives. Moreover, due to the presence of nitro functional group in both RDX and DNAN and the fact that the two compounds are used together in new explosive formulations, DNAN degradation through direct electron transfer via shuttles was investigated in this study. The data presented below demonstrate the reaction kinetics and the role of different factors, which affect DNAN degradation rates.

RESULTS AND DISCUSSION

Results

DNAN degradation by ferrous iron was first investigated within the pH range 6.0-9.0 (Figure 1). At pH 6.0 only 10% of DNAN was reduced, whereas at neutral and alkaline conditions DNAN was completely removed within 24 hours for pH 7.0 and 2 hours for pH 8.0 and 9.0. In addition, more than 80% of DNAN was reduced within 6.0 hours at pH 7.0. The same effect was achieved for pH 8.0 and 9.0 in less than 1 hour. No solid phase was required to initiate the reaction; however, iron oxidation led to formation of insoluble iron hydroxides. Controls with DNAN were run at all pH values. Iron measurements taken before and after the experiment showed a loss of 10% the total iron(II) at pH 6.0, 50% loss at pH 7.0 and approximately 80% loss at pH 8.0 and 9.0. A slight drop in pH was observed over the course of the reaction (Table 1). Separate kinetics experiments were conducted to determine the reaction rates for different pH, yielding results presented in Table 2.
Figure 1. Ferrous iron mediated DNAN reduction at pH 6.0 to 9.0

Table 1. Final pH value for DNAN reduction by Fe(II)

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Final pH</th>
<th>Treatment</th>
<th>Final pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH 6, DNAN</td>
<td>6.43</td>
<td>pH 8, DNAN</td>
<td>8.05</td>
</tr>
<tr>
<td>pH 6, Fe(II) with DNAN</td>
<td>6.47</td>
<td>pH 8, Fe(II) with DNAN</td>
<td>7.80</td>
</tr>
<tr>
<td>pH 7, DNAN</td>
<td>7.23</td>
<td>pH 9, DNAN</td>
<td>8.99</td>
</tr>
<tr>
<td>pH 7, Fe(II) with DNAN</td>
<td>7.01</td>
<td>pH 9, Fe(II) with DNAN</td>
<td>8.38</td>
</tr>
</tbody>
</table>

Table 2. Reaction rates for DNAN reduction by Fe(II)

<table>
<thead>
<tr>
<th>pH</th>
<th>$k_{obs}$ [min$^{-1}$]</th>
</tr>
</thead>
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<tr>
<td>7</td>
<td>Not calculated</td>
</tr>
<tr>
<td>8</td>
<td>0.050</td>
</tr>
<tr>
<td>9</td>
<td>0.115</td>
</tr>
</tbody>
</table>

The hydroquinone AH2QDS was also used to reduce DNAN. DNAN was immediately reduced in less than one hour at pH 7.0, and no intermediates were detected (no MENA or DAAN) (Figure 2).
Since almost no DNAN degradation was observed at slightly acidic pH, the potential for organically complexed ferrous iron was investigated as an alternative treatment strategy (Figure 3). At pH 6, iron (II) forms the most reactive complexes with 2,3-DMSA, 2,3,4-THBA and gallic acid which showed complete DNAN degradation after 2 days. Also tiron and 3,4-DHBA showed 90% and 60% DNAN reduction respectively. In addition, the strong reductant AH$_2$QDS was used as a positive control and reduced DNAN almost completely in several minutes. L-cysteine and sulfide, which can be used as an electron donor instead of iron(II), had no effect on DNAN separately, although data indicate that they may reduce DNAN to MENA when combined. All of the ligands mentioned above were also successful in DNAN degradation at pH 7. In addition to that sodium citrate, malic acid, and oxalic acid demonstrated 100%, 90%, and 40% degradation at neutral pH respectively.
Investigations of the effect of different Fe(II) initial concentration indicated that Fe(II) concentration impacts reaction kinetics. For both pH 8 and 9, only 0.6mM Fe(II) was required to completely degrade 100uM DNAN. Treatments with 1.2mM Fe(II) demonstrated faster degradation rates; however, the difference is not significant at higher pH since DNAN reduction occurs on the order of minutes. Complete DNAN reduction at pH 9 was achieved with 6 consecutive re-amendments of Fe(II), each containing 100µM Fe(II) which in total delivered 600µM Fe(II). At pH 8 DNAN reduction started almost hour after the first spike of iron was added.

Experiments with Geobacter metallireducens strain GS-15 indicated that DNAN was reduced by cells alone most likely trough electron transfer from reduced membrane cytochromes. The same reaction rate was observed for treatment with GS-15 and iron gel – poorly crystalline Fe(III). Treatments with AQDS reduced by GS-15 to AH₂QDS and soluble ferric iron reduced to Fe(II) increased the reaction rate. In treatments with soluble Fe(III) DNAN was degraded in 6 hours, which is consistent with abiotic degradation in the presence of Fe(II) at pH 7. Treatment with AQDS (reduced to AH₂QDS) showed the fastest DNAN degradation rate.

In addition, ferrous iron measurements carried over the course of the biological experiment confirm Fe(III) transformation to Fe(II) by GS-15. Fe(II) citrate was completely reduced in 2 hours suggesting that iron reduction was not the rate limiting step in DNAN degradation. Less than 30% of iron gel was reduced and slight increase in Fe(II) concentration was observed after DNAN was degraded.

Discussion

This study reports for the first time DNAN degradation mediated by ferrous iron and electron shuttles. As was already demonstrated for RDX and HMX, extracellular electron shuttles can effectively reduce cyclic nitramines, which can be advantageous for designing a remediation strategy at sites where several explosives are present in the soil and groundwater [2]. The reactions can be catalyzed through Fe(III)-reducing microorganisms which reduce electron shuttles, promoting electron transfer. Data indicated that DNAN reduction by iron(II) is highly influenced by pH, as iron becomes stronger reductant at higher pH.

Direct DNAN reduction by Fe(II) is a fast reaction at neutral and alkaline pH but was inhibited by slightly acidic conditions (pH 6). Therefore, different organic ligands were used to complex iron(II) and enhance its reactivity at this lower pH (Figure 3). The ligands form more stable complexes with Fe(III) and, thus lower reduction potential for Fe(III)/Fe(II) redox pair making Fe(II) a stronger reductant. DNAN reduction at acidic pH is possible only when ligands with catechol or thiol donor groups form reactive complexes with ferrous iron (Figure 3). These moieties are components of natural organic matter and can play a key role in natural attenuation of contaminant. Strong chelating ligands like EDTA and NTA bind Fe(II) and, thus limit access to the metal’s surface preventing electron transfer.

Also, the subsequent Fe(II) oxidation can lead to formation of highly reactive solid phases, which consist of mixed Fe(II) and Fe(III) phases, such as magnetite. Abundance of ferric minerals in the subsurface environment is, therefore, critical from the remediation perspective.
IM degradation can be mediated not only by Fe(II) but also by reduced humic substances. Hydroquinone, a model compound that represents humic substances, degraded DNAN even faster than Fe(II) and similar results were reported for RDX (Figure 2).

To completely transform 100µM DNAN to DAAN, 1200µM of Fe(II) was required. To transform the same amount of DNAN to MENA only half of this amount (600µM) was required. When lower iron(II) concentrations were investigated, DNAN was degraded to MENA in the amount corresponding to oxidation of Fe(II). Ferrous iron is less strong reductant at pH 8 than at pH 9 (Figure 8 and 9), thus at pH 8 the concentration of Fe(II) has to increase in the solution to reach the reduction potential needed to initiate DNAN degradation.

Finally, to demonstrate that Fe(III) reducing microorganisms will mediate DNAN degradation, several conditions were investigated in suspensions of *Geobacter metallireducens* strain GS-15. Fe(III) reducers are ubiquitous in the subsurface environment; however, other microorganisms that reduce for example humic substances can be employed as well. DNAN was degraded most readily in treatments with reduced quinones and reduced Fe(III) citrate, a soluble form of Fe(III). Cells alone also donated electrons to DNAN most likely through membrane cytochromes as it was previously reported for RDX.

**SUMMARY AND CONCLUSIONS**

Microbially mediated DNAN reduction by Fe(II) and reduced electron shuttles may be of great significance to the natural attenuation of the contaminant. Iron(III) is one of the metals most commonly found in the subsurface environments as well as Fe(III)-reducing microorganisms that can catalyze the reactions. DNAN degradation by ferrous iron at neutral pH is complete within hours and even faster reaction rates were achieved for hydroquinone, a model compound representing humic substances. Hence in cases where iron(II) may not be available in the sufficient amount, DNAN degradation may be achieved by reduced humic substances.

**FUTURE WORK**

Future work includes understanding how the insensitive munition nitroguanidine (NQ) is degraded under Fe(III)-reducing conditions, how photosynthetic microorganisms can biodegrade the IM, and finally how electron shuttles can accelerate simultaneous degradation of the IM and cyclic nitramine compounds.

**ACKNOWLEDGEMENTS**

We thank Scott Drew (Geosyntec) and Clint Arnett (USACE-CERL) for suggestions with respect to presenting these data.
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9.0 Biotransformation of Insensitive Munition Components in Soil Microbial Cultures

Jim A. Field*, Reyes Sierra*, Christopher Olivares*, Stephan Cameron*, Mark Krzmarzick*, Hector Amezquita Garcia*, Leif Abrell†, Jon Chorover‡ Raju Khatiwada† and John Coffey II†

*Department of Chemical and Environmental Engineering, University of Arizona, Tucson, Arizona, United States
†Department of Soil, Water & Environmental Science, University of Arizona, Tucson, Arizona, United States
‡CH2M Hill International, Denver, Colorado, United States

ABSTRACT

The goal of this study was to evaluate the biotransformation of 2,4-dintroanisole (DNAN) and 3-nitro-1,2,4-triazol-5 one (NTO) in soil cultures. Each soil type was tested under aerobic incubations without any supplemental substrate and under anaerobic conditions with H2 as an electron donating substrate. Neither DNAN or NTO were significantly degraded in any soil under aerobic conditions. Both DNAN and NTO were readily biotransformed under anaerobic conditions. DNAN was converted to 2-methoxy-5-nitroaniline (MENA) and 2,4-diaminoanisole (DAAN) as intermediates which in turn were consumed and further converted to dimeric coupling products. The dimeric coupling products are hypothesized to be formed by a reaction between nitroso- and aromatic amine intermediates forming an azo bond. NTO was readily converted to 3-amino-1,2,4-triazol-5 one (ATO). While ATO was not transformed further in anaerobic conditions. Rapid biodegradation of ATO in dome soils under aerobic conditions was confirmed by release of ammonium and mineralization of ATO carbon to CO2. The results taken as a whole indicate that Insensitive munitions compounds (IMCs) are poorly biodegradable under aerobic conditions but can be rapidly biotransformed under anaerobic conditions. Biotransformation products of DNAN are converted to dimers. ATO, the reduced anaerobic biotransformation product of NTO, can be mineralized completely to CO2 if the conditions are changed to aerobic.

INTRODUCTION

IMCs are a new class of explosive that with a decreased risk of unintended explosions (Davies et al. 2006; Millar et al. 2008). The U.S. Military plans to increase the use of two new IMCs, NTO, and DNAN. Less information is available on the environmental fate of IMCs, DNAN and NTO, as compared to conventional munitions compounds such as 2,4,6-trinitrotoluene (TNT).

In order to better understand the environmental fate of IMCs at firing ranges, the goal of this study is to evaluate biotransformation processes in the soil that collectively contribute to their conversion and attenuation under a range of soil conditions with a focus on aerobic versus anaerobic conditions.
MATERIAL AND METHODS

Anaerobic experiments were carried out in 160 mL serum flasks with butyl rubber septa. In each flask, 100 mL of medium was utilized and the headspace was flushed with 80%:20% (v/v) N₂/CO₂. The composition of basal mineral medium was described previously (Olivares et al. 2013) and included 4000 mg/L NaHCO₃ as a buffer (in conjunction with CO₂ in the headspace). DNAN and NTO was incubated in the medium at 130-500 (depending on experiment) and 1500 μM, respectively. In experiments, where H₂ was used as an electron donating substrate, the H₂ was supplied by pressurizing the flask headspace to 1.5 atm with a gas mixture (H₂/CO₂, 80:20 v/v) after flushing with N₂/CO₂. In some experiments organic compounds such as glucose, acetate and pyruvate were used as C-source or electron donors.

Aerobic biotransformation assays were conducted in glass Erlenmeyer flasks (250 mL) capped with cotton gauze and placed on an orbital shaker at 180 rpm. The medium was the same as the anaerobic medium except they did not contain NaHCO₃ and 15 mM phosphate pH 7.2 buffer was used instead. Twice weekly water was added to the cultures to correct for evaporation (based on weight loss measurements). In some experiments, aerobic experiments were conducted in closed 160 ML serum flasks with 100 mL medium and the space flushed with 80%:20% He/O₂.

Most of the experiments were inoculated with soils at a concentration of 50 g L⁻¹, although in selected experiments a concentration of 5 to 10 g L⁻¹ was used. Most of the soils used were characterized (see Table 1). Some newer soil samples were also utilized but haven’t been characterized yet. One is Gortner from the Gortner garden on the University of Minnesota (enriched with compost), and Nibley soil located on Camp Navajo in Flagstaff, Az, one sample was from inside of a munitions disposal area (Nibley-In) and another came from outside of the disposal area (Nibley-out). Lastly two sludge samples were evaluated (anaerobic granular biofilm and aerobic return activated sludge (RAS)). As controls for experiments, heat killed inoculum was used and medium only without soil (or sludge). In the case of anaerobic experiments, endogenous controls (without addition of electron donating substrate) was also used. Heat-killed soil was prepared by autoclaving the sludge (121°C) for 4 consecutive days for 50 min the first day and 20 min the next 2 days.

The assays were incubated in the dark at 30°C. In the anaerobic biotransformation assays, the flasks were pre-incubated overnight to ensure that the sludge adapted to the medium conditions. DNAN was added to the flasks in the following morning. All assays were performed in duplicate.

DNAN, MENA, and DAAN in liquid phase were analyzed using an Agilent 1200 series high pressure liquid chromatograph – diode array detector (HPLC-DAD) (Santa Clara, CA, USA). Samples (20 μL injection) were separated using an Acclaim Explosives E2 RSLC, 2,2 μm, analytical column 2.1 x 100 mm (Thermo Scientific) at room temperature. The mobile phase (methanol/H₂O, 43/57% v/v) was run isocratically at a flow rate of 1 mL min⁻¹ for 30 min. The detector was set to scan for wavelengths of 210, 220, 254, 270, 280, 300, 325, and 360 nm. Detection of DNAN, MENA, and DAAN was performed at 300, 254 and 210 nm, respectively.
The concentration of NTO and ATO was determined by analyzing samples with the HPLC-DAD, using a mobile phase (water with 0.1% trifluoracetic acid and acetonitrile) as described in previous reports. Five microliters were injected into a Thermo Scientific Hypercarb column (150 x 4.6 mm, particle size = 5 um). The wavelength used for detection was 216.5 nm.

High resolution full scan mass spectra were obtained with a TripleTOF™ 5600 quadrupole TOFMS (AB Sciex, Framingham, MA) equipped with ESI source kept at 450 °C. The sample was infused at a flow rate of 10 µL min⁻¹. Spectra were obtained in ESI positive ion mode with a capillary setting of 5.5 kV and declustering potential of 50 V. Curtain gas, desolvation gas, and nebulizer gas levels were kept at 30, 35, and 35 psi, respectively, with nitrogen. High resolution mass spectra were obtained by averaging ca. 100 spectra acquired from a mass range of 35-600 m/z. Analyst TF 1.5.1 and Formula Finder 2.0.2.0 software applications were used to process spectral data and to identify molecular formulae.

The inorganic nitrogen species NH₄⁺, NO₂⁻, and NO₃⁻ were measured with ion chromatography (IC) for assays investigating the aerobic degradation of ATO. The IC analyses was performed on an ICS-3000 system (Dionex) with a split flow for simultaneous anion and cation analysis on an AG18 RFIC 4 × 50 mm column (Dionex) and IonPac CG16 RFIC 3 × 50 mm column (Dionex), respectively. The eluent flow rate for anion analysis was 1.0 mL min⁻¹ and for cation analysis was 0.5 mL min⁻¹.

ATO was synthesized from NTO by reducing it with Pd coated activated carbon in methanol supplied with H₂ (the reaction was stopped when no more H₂ was taken up).

RESULTS

Aerobic Incubations. DNAN and NTO biodegradation was tested under aerobic conditions with 10 different soil samples and aerobic activated sludge for incubations up to 112 days. Under fully aerobic conditions (O₂-unlimited), no biotransformation of DNAN and NTO was observed. An example of such an experiment is shown for the two Nibley soil samples incubated with DNAN (Figure 1). DNAN and NTO were on the other hand readily biotransformed under anaerobic conditions.

Anaerobic Incubations DNAN. Figure 2 shows rapid biotransformation of DNAN in soils under anaerobic conditions supplemented with the electron donating substrate, H₂. The rate of DNAN biotransformation was correlated with the soil organic carbon content. The DNAN was converted to MENA and DAAN (Figure 3). Significant (bio)transformation was also observed in endogenous and heat killed controls although the rates were slower when compared with live soil. The endogenous and heat killed controls, DNAN provided nearly stoichiometrically conversion of DNAN to MENA and DAAN. However, in the live treatment, MENA and to a lesser extent DAAN that initially formed, subsequently disappeared. Thus, new compounds were formed that were not feasible to measure on the HPLC DAD.

In order to determine what the new compounds were, UHPLC-QTOF-MS was utilized. Samples were collected in an anaerobic hood, spiked with 200 mg L⁻¹ ascorbic acid and
transferred to the QTOF-MS in an anaerobic box to eliminate any autoxidation reactions caused by air during sampling and handling. The results in Figure 4 show QTOF-MS determined products for an incubation with 10 g L\(^{-1}\) North Carolina soil for 40 d. A large number of the products were dimers. The most likely explanation for dimer formation is a nucleophilic attack between a nitroso intermediate and DAAN yielding a DAAN dimer with an azo linkage (m/z 273) as shown in Figure 5. A key intermediate for this reaction is 2-methoxy-5-nitrosoaniline (nitroso-MENA), which has been measured on multiple occasions in different experiments (although not detected in the sample of Figure 4). The dimer is then metabolized further by different reactions (Figure 5). Firstly there is a series of reduction reactions yielding hydrazo-dimer (m/z 275) and the hydrazo-group can also be reductively cleaved to regenerate the pool of DAAN. Or alternatively the metabolism may include O-demethylation reactions yielding free hydroxyl groups (m/z 259). The amino groups can become N-methylated (m/z 269 and 285). Some N-methyl groups are also oxidized to N-methylene groups (it not clear if the latter reaction occurs in the system or is artifact of the analysis). The hydroxyl groups can become dehydroxylated under the reducing conditions (m/z 269).

In additional experiments conducted in continuously fed anaerobic soil columns some additional metabolites were detected, including 2-amino-4-nitrophenol, N-acetylated dimers and three trimer compounds.

**Anaerobic Incubations of NTO.** In anaerobic microcosms with H\(_2\) added as an electron donor, NTO was fully biotransformed as shown in the examples of Camp Navajo and Florence soils (5 g L\(^{-1}\)) in Figure 6. In total seven soils were tested and all seven provided rapid rates of NTO biotransformation ranging from 0.23 to 1.25 mM d\(^{-1}\). No biotransformation was observed in killed controls or non-inoculated basal media (Figure 2). Endogenous controls, to which no electron donor was added, displayed very slow NTO bioconversion rates ranging from 0.008 tp 0.048 mM d\(^{-1}\). The lag phase prior to NTO degradation was less than 5 days in all soils except for Roger Road (AZ), and Maricopa (AZ) with a lag phases of approximately 12 days. No clear connection between the biotransformation rates and the organic carbon or total nitrogen content of the soil was observed.

ATO was the dominant product from NTO degradation under anaerobic conditions. The ATO formation was concomitant to the removal of NTO, and the yield of ATO production from NTO removal was stoichiometric, averaging 95.3 ± 9.4% in the H\(_2\) amended microcosms. The conversion of NTO to ATO was dependent on the presence of an electron donor; the conversion NTO to ATO was very low in endogenous microcosms although the yield per unit NTO removed was similar to that observed in H\(_2\)-amended microcosms. Many organic compounds also served as electron donors aside from H\(_2\) to drive NTO biotransformation to ATO. Some of the best electron donors aside from H\(_2\) were lactate lactose and glucose. A small supplement of yeast extract (10 mg L\(^{-1}\)) was found to highly stimulate biological NTO reduction rates especially when H\(_2\) or lactate was used as the electron-donor.

The main product of NTO biotransformation was ATO, and once formed it never was degraded further even after 34 days of incubation in the anaerobic NTO experiments. This was observed in seven soils. In two of the soils (Camp Butner (NC) and Camp Navajo, Flagstaff...
AZ), ATO was incubated directly for 62 days under anaerobic conditions under N-limiting conditions with either glucose or H₂. However, even after 62 days, there was no evidence of ATO degradation in the absence of O₂. Thus, experiments were conducted to determine if aerobic degradation of ATO was feasible.

**Aerobic Biodegradation of ATO.** ATO (1.5 mM) was incubated aerobically in soil (10 to 50 g L⁻¹ depending on soil organic carbon) in a minimal media with yeast extract (100 mg L⁻¹) and defined vitamins as the sole C and N source. A screening was conducted initially. At day 25 the residual ATO in a live soil or sludge sample was divided by the residual ATO in a heat killed control. The results in Figure 7 indicate that ATO was removed fully or in part in all of the soils. However, there were three soils in which the ATO in the live treatments were fully depleted. These included: Roger Road (AZ), Gortner (MN), and Camp Butner (NC).

Next experiments were designed in closed bottles with 80% He and 20% O₂ to demonstrate the formation of CO₂. In parallel, the release of inorganic nitrogen containing ions (NH₄⁺, NO₃⁻ and NO₂⁻) was also measured with an IC along with the decrease in ATO concentration using the HPLC-DAD. A typical experiment is shown in Figure 8 with Gortner soil supplied at 10 g L⁻¹. Between day 3 and 13, ATO was completely eliminated from the live treatment. No disappearance occurred in heat-killed or media only controls, indicating that the disappearance was caused by biodegradation. Simultaneously, there is a corresponding release of NH₄⁺ accounting for 71% of the N in ATO. Likewise, at the same time there is an acceleration of CO₂ production in the ATO-spiked soil above the background CO₂ production of soil alone. The increment in CO₂ in the headspace of 0.88% corresponded to full mineralization of ATO carbon. The value of 0.88% value was calculated based on mass balancing the carbon as CO₂ between the gas phase and the liquid phase with acid base equilibrium and Henry’s law. In conclusion, the results indicate a comprehensive biodegradation of ATO to mineral products under aerobic conditions. Similar results were also obtained with Roger Road (AZ) soil.

**DISCUSSION**

The IMC compounds were not biotransformed under aerobic conditions. The observed conversion of DNAN to MENA by an aerobic bacteria (Perreault et al. 2012) in a previous study does not necessarily contradict our finding. In our study no co-substrates were supplied; whereas in the previous study a medium rich in organic substrates was provided making the conditions limiting in O₂ and the biotransformation observed was reductive (not oxidative). In the case of nitroaromatic compounds (such as DNAN), the electron withdrawing character of the nitro-group makes such compounds less susceptible to an aerobic oxidative attack as the number of nitro-substitutions increases (Field et al. 1995). Highly nitrated nitroaromatics such as trinitrotoluene (TNT) are not microbially oxidized to CO₂ (Bruns-Nagel et al. 2000; Drzyzga et al. 1998). Instead such compounds are subject to reductive biotransformations forming reduced products such as aromatic amines (Boopathy et al. 1998; Gorontzy et al. 1993; Rafii et al. 1991). Likewise in this study we observed a rapid biotransformation of DNAN to corresponding aromatic amines (MENA and DAAN) under anaerobic conditions. Initial experiments in the literature with DNAN biodegradation confirm these general trends, DNAN was shown to be
readily reduced to corresponding aromatic amines (Olivares et al. 2013; Perreault et al. 2012; Platten et al. 2010).

Aromatic amine residues become polymerized and covalently bound into soil humus. Soil composting is one of the most accepted bioremediation strategies for treating soil contaminated with TNT (Jørgen and Woodhull 2000). A large fraction of $^{13}$C labeled TNT spiked into soil composting microcosms has been shown to be recovered in soil humus fractions (Bruns-Nagel et al. 2000; Drzyzga et al. 1998). In this study, the initial stage of the polymerization were observed through the formation of dimer compounds by reactions of reduced intermediates of DNAN. The formation of dimer compounds have previously been observed albeit with poor evidence for structural assignments (Platten et al. 2010). The dimers were rationalized as autoxidation products formed by exposure of samples to air. Also aerobic bacteria responsible for the reductive biotransformation of DNAN in $O_2$-limiting conditions were found to also form dimers (Perreault et al. 2012). In this study, the sampling and handling was carried out in an anaerobic hood and samples were spiked with 200 mg L$^{-1}$ of ascorbic acid in order to prevent autoxidation. Thus, the observed formation of dimers must be attributed to processes occurring in the absence of $O_2$. The most likely explanation for dimer formation is a nucleophilic attack between a nitroso intermediate and DAAN yielding an azo linked DAAN dimer. This reaction mechanism has been proposed during the synthesis of azo dyes by reduction of nitroaromatics with nano-iron (Moglie et al. 2008) or by reaction of aromatic amines with nitroaromatics (Zhao et al. 2011).

Less is known about nitrotriazoles. In this study, we found that NTO was only reduced under anaerobic conditions, the corresponding amine, ATO was not degraded further under anaerobic conditions. However, rapid mineralization of ATO to ammonium and CO$_2$ was observed under aerobic conditions in a few soils. Preliminary work with NTO has shown that bacteria reduce it to ATO. Additionally there is evidence that ATO subsequently can be mineralized by bacteria to CO$_2$ and NH$_4^+$ (Le Campion et al. 1998). Other highly nitrated heterocyclic explosive compounds such as the nitro-amine compound hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX) are more prone to being converted to mineralized products such as CO$_2$ compared to nitroaromatics (Hawari et al. 2000).

The results taken as a whole suggest that nitrated IMC compounds, DNAN and NTO, require anaerobic or $O_2$-limiting conditions to initiate biotransformation. Afterwards, reduced intermediates of DNAN are subject to polymerization reactions such as dimer and trimer formation by coupling reactions. On the other hand, ATO, the reduced intermediate of NTO can become mineralized by soil microorganisms under aerobic conditions.

ACKNOWLEDGEMENTS

This work was funded by SERDP grant ER-2221
Table 1. Selected properties of the soils used in this study

<table>
<thead>
<tr>
<th>Soils</th>
<th>pH</th>
<th>BET SA $^\dagger$</th>
<th>TN $^\S$</th>
<th>TC $^\¶$</th>
<th>TOC $^\‡$</th>
<th>Soil Texture</th>
</tr>
</thead>
<tbody>
<tr>
<td>Camp Ripley (MN)</td>
<td>5.96 ± 0.06</td>
<td>1.72 ± 0.03</td>
<td>1.27 ± 0.20</td>
<td>12.55 ± 1.50</td>
<td>12.5 ± 1.50</td>
<td>78.20 13.99 7.82</td>
</tr>
<tr>
<td>Camp Butner (NC)</td>
<td>6.36 ± 0.02</td>
<td>4.85 ± 0.07</td>
<td>1.33 ± 0.05</td>
<td>20.69 ± 1.20</td>
<td>20.69 ± 1.20</td>
<td>68.68 19.83 11.50</td>
</tr>
<tr>
<td>Florence (AZ)</td>
<td>6.96 ± 0.11</td>
<td>32.45 ± 1.73</td>
<td>0.81 ± 0.02</td>
<td>4.16 ± 0.20</td>
<td>4.16 ± 0.20</td>
<td>44.20 28.50 27.30</td>
</tr>
<tr>
<td>Camp Navajo (AZ)</td>
<td>6.32 ± 0.01</td>
<td>21.5 ± 0.56</td>
<td>3.65 ± 0.21</td>
<td>52.36 ± 3.70</td>
<td>52.36 ± 3.70</td>
<td>21.48 38.10 40.43</td>
</tr>
<tr>
<td>Maricopa (AZ)</td>
<td>7.75 ± 0.07</td>
<td>34.58 ± 1.70</td>
<td>0.80 ± 0.05</td>
<td>7.07 ± 0.40</td>
<td>4.65 ± 0.40</td>
<td>37.48 21.98 40.55</td>
</tr>
<tr>
<td>Roger Road (AZ)</td>
<td>7.75 ± 0.01</td>
<td>27.69 ± 0.80</td>
<td>1.54 ± 0.03</td>
<td>18.25 ± 0.10</td>
<td>7.07 ± 0.40</td>
<td>23.33 35.10 41.58</td>
</tr>
<tr>
<td>Catlin Soil (IL)</td>
<td>6.42 ± 0.06</td>
<td>5.05 ± 0.44</td>
<td>2.81 ± 0.18</td>
<td>45.44 ± 1.10</td>
<td>44.08 ± 1.10</td>
<td>13.50 54.98 31.53</td>
</tr>
</tbody>
</table>

$^\dagger$Brunauer, Emmett and Teller (BET) Surface area; $^\S$ TN = Total nitrogen; $^\¶$ TC = Total Carbon $^\‡$ TOC = Total Organic Carbon.
Figure 1. Lack of DNAN biodegradation during aerobic incubation with soil

Figure 2. Rapid anaerobic biotransformation of DNAN in four soils with high organic carbon content ranging from 12 to 52 g organic carbon kg\(^{-1}\) soil dwt. Soils (50 g L\(^{-1}\)) were supplied with H\(_2\) as electron donor. Rates of DNAN biotransformation were significantly lower in soils with low organic carbon content ranging from 4 to 7 g organic carbon kg\(^{-1}\) soil dwt.
Figure 3. DNAN (●), MENA (□), DAAN (▲) and sum HPLC-DAD measured compounds (----) during anaerobic DNAN biotransformation for North Carolina soil incubated as live soil with H₂ (A), endogenous control (B), and heat-killed control (C).

Figure 4. Dimer biotransformation products observed after 40 days of anaerobic incubation of North Carolina soil (10 g L⁻¹) with 20 mM pyruvate (as electron donor) using UHPLC-QTOF-MS.
Figure 5. Proposed pathway of dimer formation and further metabolism of the dimers
Figure 6. The anaerobic degradation of NTO to ATO in microcosms inoculated with (A) Camp Navajo, Flagstaff (AZ) and (B) Florence (AZ) soils (5 g L\(^{-1}\)) in H\(_2\) amended microcosms (black squares) endogenous controls (triangles), and killed controls (circles). NTO concentrations are shown with solid symbols and solid lines and ATO concentrations are shown with open symbols, dotted lines). Error bars indicate standard deviation of duplicate microcosms.
Figure 7. Screening of aerobic ATO degradation by soils, return activated sludge (RAS) and an enrichment culture derived from NC soil. The residual concentration of ATO remaining at day 25 in live cultures was divided by the residual concentration in heat killed controls.
Figure 8. Correspondence of ATO disappearance during aerobic degradation by Gortner soil (10 g L−1) with the release of ammonium and CO₂. NH₄⁺ release corresponds to 71% of nitrogen in ATO, and CO₂ release corresponds to 100% of carbon in ATO.
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10.0 Forecasting the Environmental Impacts of New Energetic Formulations

K.R. Glaesemann and E.J. Bylaska, Environmental Molecular Sciences Laboratory, Pacific Northwest National Laboratory; P.G. Tratnyek and A.J. Salter-Blanc, Institute of Environmental Health, Oregon Health & Science University

ABSTRACT

There is a need to have quantitative models that can predict the reactivity (i.e., transformation) of future explosive compounds. The environmental fates of explosives (both used and unused) are of great interest to the Department of Defense, because the cost of cleanup. Many redox reactions of importance in degradation chemistry involve multiple elementary steps that occur by single-electron transfer (SET). A SET step is often the first and rate limiting step in reactions of environmental contaminants, so there is a great deal of interest in the corresponding one-electron reduction potentials ($E^1$). Although $E^1$ can be obtained by experimental methods, calculation from first-principles chemical structure theory is an increasingly attractive alternative. Sufficient data are now available to perform a critical assessment of these methods for contaminant degradation reactions involving compounds present in explosives. Previous work involving datasets containing $E^1$'s for dehalogenation of chlorinated aliphatic compounds (CAC) by dissociative SET contained a variety of errors and inconsistencies, but the preferred datasets showed good agreement between values calculated from thermodynamic data and quantum mechanical models. The end goal will be quantitative structure-activity relationships (QSARs) that are accessible to non-specialists.

INTRODUCTION

There is a need to have quantitative models that can predict the reactivity (i.e., transformation) of future explosive compounds. The environmental fates of explosives (both used and unused) are of great interest to the Strategic Environmental Research and Development Program (SERDP), because the financial and political costs of the environmental fate of chemicals can be great. This work focuses on reaction property predictions. This work suggests the four major reaction pathways should be studies: Hydrolysis/Elimination, Homogeneous nitro reduction (in solution), Heterogeneous nitro reduction (on surfaces), and Oxidative coupling of nitro reduction products. This work will demonstrate the novel strategy for fully in silico calibration of predictive models for properties of chemicals. The end goal will be QSARs that are accessible to non-specialists. We use “in silico calibration” to overcome the practical restrictions on conventional methods of QSAR development that rely on calibration with experimental data, although extensive experimental data will be acquired.

RESULTS AND DISCUSSION

Introduction to Single Electron Transfers

The characteristic reactions of most major redox active species in the aquatic environment involve transfers of an even number of electrons between closed-shell donors and acceptors. The standard potentials for these reactions are readily available, often in critically evaluated reviews.
[1-4], usually calculated from standard thermochemical data, but occasionally measured directly by electrochemical or other methods. Meta-analyses of these standard potentials have been performed to investigate various fundamental aspects of environmental redox processes, such as the role of thermodynamically unstable intermediates as barriers to multi-electron redox process in biogeochemistry [5, 6]; the viability of various pathways of microbial metabolism [7-9]; feasibility of contaminant degradation pathways [10, 11], and prospects for extraterrestrial life [12].

However, the mechanisms of most redox reactions are composed of elementary steps that involve SET, usually forming radical intermediates, which react further by various mechanisms to form stable products. The initial SET step is often thermodynamically unfavorable, providing the barrier that inhibits the equilibration and controls the kinetics of multistep redox processes that are favorable overall [5, 6]. For overall reactions that are far from equilibrium under environmental conditions, such as those that result in degradation of explosive contaminants, the initial SET step is particularly important in controlling the kinetics of contaminant transformation. This is well established for nitro aromatics compounds (NACs) [e.g., 13, 14-16] and anilines [e.g., 17, 18, 19].

The quantitative property that is most often used to describe the rate-limiting SET steps in these redox reactions is the $E^1$ for the corresponding half-reaction [19, 20]. Experimental values of $E^1$ for these half-reactions are relatively scarce, but there are a number of methods by which they can be obtained [21, 22]. $E^1$ data can be obtained from voltammetry, but aprotic solvents are usually necessary to stabilize the radical intermediates [3, 21, 23, 24], or from pulse radiolysis, but this method usually requires the use of an intervening mediator compound [14, 21, 25]. These complications make theoretical calculations of $E^1$ especially important for studies of redox reactions of organic contaminants.

The most basic strategy for determining $E^1$’s is to calculate the free energy difference for the reaction, for example:

$$\text{R-NO}_2\text{(aq)} + 1\text{e}^- \rightarrow \text{R-NO}_2^\cdot\text{(aq)} \quad [1]$$

and then convert Δ$G$ to a potential using

$$E_{\text{abs}} = -\frac{\Delta G}{nF} \quad [2]$$

Where $n$ is the number of electrons transferred and $F$ is the Faraday constant. This difficult to measure type of potential is known in electrochemistry as an absolute potential ($E_{\text{abs}}$). Instead, redox potentials are usually measured relative to an inert electrode, such as the standard hydrogen electrode (SHE). It is conventional to report redox potentials relative to the SHE. This means that the free energy differences for the one-electron reduction half reaction given in eq. 1 is usually reported in terms of the following overall reaction.

$$\text{R - NO}_2\text{(aq)} + \frac{1}{2}\text{H}_2\text{(g)} \rightarrow \text{R - NO}_2^\cdot\text{(aq)} + \text{H}_\text{(aq)}^+ \quad [3]$$
Electronic structure and solvation methods can be used to estimate SHE potentials. However, the development of a computational scheme that can accurately predict $E^I$ requires care. A commonly used strategy, that does not use any experimental data, is to directly calculate the absolute potentials by using electronic structure methods and solvation models to calculate the free energy difference for the one-electron transfer reactions and then convert it to an SHE potential. For reactions in which the reaction energy difference is just an adiabatic electron affinity, this approach can provide reasonable estimates (errors less than 0.2 V) provided the solvation models are designed to correctly treat the radical anion product. Unfortunately, for reactions in which the one-electron transfer reaction involves the making or breaking of covalent bonds, this strategy is prone to large errors (i.e., greater than 0.5 V). This is because electronic structure methods need to include high-level treatments of the electronic correlation energy—which are very expensive to compute—in order to directly calculate bond energies. In addition to the issues associated with electronic structure methods, our objective (the prediction of redox potentials in solution) also requires the calculation of solvation energies. Several models exist for calculating solvation energies. Currently, the most computationally feasible models for estimating solvation energies are “continuum” reaction field solvation models [26]. Despite the approximate treatment of solvation in these approaches, they have been shown to give hydration energies of many neutral molecules within 1 kcal/mol (0.05 V) when compared to experiment results (for charged species, errors are typically larger, on the order of 0.1 – 0.2 V).

**Estimating Entropy and Free-energies**

In this study, the following strategy was used to calculate the solution phase $E^I$’s. The absolute potentials were directly calculated from gas-phase reaction energy, entropy and solvation energy differences using electronic structure calculations, gas-phase entropy estimates, and continuum solvation models.

Given an optimized structure and vibrational frequencies for a gas-phase polyatomic molecule, one can calculate thermodynamic properties using formulas derived from statistical mechanics [27]. In many cases, results obtained with these formulas, and accurate structures and frequencies, can provide values more accurate than those determined by measurements. Calculation of these formulas is straightforward and most electronic structure programs contain options for calculating them. In this work the authors use Density Functional Theory (DFT) to calculate optimized gas-phase geometries and vibrational frequencies. DFT has been shown to be a reliable predictor of both structure and energies for organic compounds. DFT methods are computationally efficient, which is essential given the large numbers of molecules that need studied. However, for compounds that contain internal degrees of freedom (e.g., molecules that contain fragments such as -NO$_2$ rotors) that are not well described by normal vibrations, these formulas need to be corrected. Estimating accurate entropies in this situation is very demanding, and several strategies for this have been developed [26, 28]. The strategy that has been used by the authors is to explicitly solve for the energy levels of the rotational Schrödinger equation for each rotor and then use this as input in a canonical partition function to estimate its entropy [28, 29]. The gas-phase free energy of formation $\Delta G^\circ$ can be determined using the values of $\Delta H^\circ$ and $S^\circ$. This is done by calculating the entropy of formation, $\Delta S^\circ$, found by subtracting off the entropies of the atomic standard states from the virtual entropy of the compounds.
Estimating Solvation Energies

Solvent effects are estimated using the COSMO method [30, 31]. The solvent is treated as a dielectric continuum with a permittivity \(\varepsilon\) surrounding the solute molecule’s molecular cavity. The charge distribution on the cavity is calculated using the quantum chemistry method. The polarization charges are derived from the continuum, caused by the polarity of the solute, from a scaled-conductor approximation. Despite the approximate treatment of solvation in this approach, it (and others like it) has been shown to give hydration energies for many neutral molecules that are within a few kcal/mol of the experimental values.

Calculated free energies of solvation cannot be compared directly to thermodynamic tables, because they use different standard states. The standard states for the COSMO model are 1 mol/L at 298.15 K in the gas phase and 1 M at 298.15 K in the solution phase. Thermodynamics tables define the standard state for the solute as 1 bar of pressure at 298.15 K in the gas phase and 1 M and 298.15 K in the solution phase. In order for the theory calculations to conform to the standard state of 1 bar of pressure at 298.15 K in the gas phase, a constant value of 1.90 kcal/mol must be added to the free energies of solvation.

For charged solutes, comparisons are less straightforward. Thermodynamic tables report free energies of formation for charged solutes or electrolytes in solution relative to \(\text{H}^+\) (aq) with the convention. The reader is directed to the references for a discussion of the derivation of the calculation of aqueous free energies of formation:

\[
\Delta G^\circ(X_{\text{aq}}) = \Delta G_{\text{SCRF}}(X) + \Delta G^\circ(X_{\text{aq}}) + \Delta G^\circ(H^+_{\text{aq}}) - \frac{1}{2} \Delta G^\circ(H_2\text{aq}) - E_H^0 + \Delta G^\circ(e^-_{\text{aq}}) - \frac{1}{2} \Delta G^\circ(e^-_{\text{aq}})
\]

[4]

Computational Descriptor Variables and Correlations to Experiments

A few select examples of correlations and results are presented in this report. An extensive database of simulations was developed for organizing the results into a usable framework for effective data mining. The framework is a set of Python scripts that the chemist inputs a SMILES structure and then the script submits an appropriate simulation to the supercomputer. The simulations perform a geometry optimization, a vibrational analysis, and finally a COMSO calculation. The Python scripts monitor the simulations and can automatically restart simulations that failed due to time limits on the computer. A variety of schedulers are supported (SLURM, MOAB, PBS). Once the calculations are completed, the results are gleaned from the NWChem output files and put into a MySQL database. These Python scripts can be requested from author Dr. Bylaska. Multiple reaction pathways were studied for each molecule because no assumption about which mechanism is dominant: proton abstraction, Meisenheimer complex formation, or direct nitro substitution.
Figure 1. The three different possible mechanisms for alkaline degradation

In order to determine which structural relationships are usable for developing usable QSARs many properties need to be analyzed including E1 for disassociation discussed above, HOMO energy, LUMO energy, coordination number, etc. Previous reports have focused on LUMO and HOMO relationships. All calculations used DFT and a plane wave basis set in the code NWChem. The COSMO solvation model is also applied to all results. A comparison of various DFT functionals is presented in Figure 2. This comparison (normalized to B3LYP) shows which molecules have results that should be investigated further, since at least one functionals results are significantly different.
Figure 2. Correlation of E1 using different DFT functionals (kcal/mol)

The hybrid exchange-correlation functional B3LYP have been the quantum chemistry gold standard for energetics for two decades, since its inclusion in Gaussian92/DFT. The PBE exchange correlation functional is also a gradient corrected but PBE does not include exact Hartree-Fock exchange. These Figures provide an example of all the data that has been collected. The E1 values reflect a first-order estimate of the relative stability of the compounds in the environment. This number alone should not be used to judge environmental friendliness: E1 is not a full mechanism rate and fast degradation is not useful if the products are dangerous. In Figure 3, individual E1 values using B3LYP are presented. In Figure 4 individual E1 values using PBE are presented. The missing values are values that were determined to be incorrect using the data in Figure 2 as a guide; the missing values are currently be recalculated with careful attention to the problems that occurred with the first series of calculations. The errors were generally the simulation following a non-optimal path.
Figure 3. E1 values calculated using B3LYP

Figure 4. E1 values calculated using PBE
SUMMARY AND CONCLUSIONS

Computational and experimental work provides an effective way to determine QSARs for a variety of compounds of interest to the explosives field. Reactions that are well defined by a single electron transfer (SEP) are best described. For other work, the reader is directed to previously published efforts by the authors.[32]

ACKNOWLEDGEMENTS

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11.0 Toxicity of IMX Formulations and Components: What We Know and Path Forward

L.R. Williams, W.S. Eck, and M.S. Johnson, U.S. Army Institute for Public Health, Toxicology Portfolio, Aberdeen Proving Ground, MD

ABSTRACT

New insensitive munitions are being developed to minimize the acute hazards associated with sympathetic and non-intentional detonation of warheads. Two components that are used in some insensitive munition formulations have limited toxicity data, i.e. 3-nitro-1,2,4-triazol-5-one (NTO) and 2,4-dinitroanisol (DNAN). Oral acute, subacute and subchronic studies have been completed on both compounds. The primary adverse effect from subchronic oral NTO exposure was hypospermia which followed a dose-dependent trend. Effects from DNAN exposure include anemia and testicular atrophy. Recent methods to ascertain a point of departure (POD) from toxicity data have migrated away from using no-observed adverse effect levels (NOAELS) to the U.S. Environmental Protection Agency (USEPA’s) benchmark dose (BMD) approach; a more robust method which uses the entire dose-response relationship. Occupational exposure levels (OEL) have been developed for NTO and DNAN using the BMD approach and have recently been peer-reviewed. However, other concerns regarding environmental issues have been raised. Here we present the latest toxicity data and interpretations for NTO, nitroguanidine (NQ) and DNAN and describe a process to assist with holistic Environmental, Safety, and Occupational Health (ESOH) assessment and sustained production and use.

INTRODUCTION

Unintentional detonation of munitions and munition stockpiles has caused losses to life, equipment, and infrastructure. The U.S. Department of Defense (DOD), therefore, has a focused initiative to improve the safety of munitions, developing insensitive munitions for use in future and existing weapon systems (Duncan 2002). Insensitive munitions are “munitions which reliably fulfill their performance, readiness and operational requirements on demand, and which minimize the probability of inadvertent initiation and severity of subsequent collateral damage to weapon platforms, logistic systems, and personnel when subjected to unplanned stimuli” (Beyard 2007). In addition to minimizing collateral damage from weapon or ordnance accidents, insensitive munitions offer logistical advantages on the battlefield; more munitions can be stored in a given area if quantity-distance requirements are reduced, resulting in more efficient use of available land and smaller targets for potential enemy action. As modern battlefields increasingly shift into populated urban centers, insensitive munition inventories represent a less desirable target for terrorists and minimize the threat to surrounding communities. Less-sensitive munitions could potentially be more cost effective and efficient to transport if granted reduced DOD/Department of Transportation hazard classification rankings. IMX-101 is planned for use in several weapons systems.

Military readiness depends on the sustainable use of materials at present ranges where testing and training activities occur. This requires an assessment of human health and environmental effects arising from exposure to substances in soil, surface water, and ground water. It is most
efficient to begin the assessment of exposure, effects, and environmental transport of military-related compounds/substances early in the research, development, testing and evaluation (RDT&E) process in order to avoid unnecessary costs, conserve physical resources, and sustain the health of our forces and others potentially exposed. Failure to identify problems early can lead to program delays or even cancellations, as well as unnecessary costs.

In an effort to support this preventive approach, the U.S. Army Public Health Command (USAPHC) has developed a phased process to assess environmental, safety and occupational health (ESOH) consequences which can adversely impact readiness, training, and development. This approach requires specific data for this assessment. This report provides the data available to assess ESOH impact of three insensitive munition (IM) materials, NTO, NQ, and DNAN.

RESULTS AND DISCUSSION

DNAN – Oral Toxicity

DNAN (dinitroanisole or 1-methoxy-2,4-dinitrobenzene) is also a component of IMX-101 and also in the explosive formulation known as PAX-21 which has been fielded for several years.

An acute oral LD$_{50}$ in rats was determined on DNAN and found to be 199 mg/kg in both sexes of rat. Clinical signs of toxicity included decreased activities, breathing abnormalities, salivation and soft stools. No remarkable clinical findings or gross lesions were discovered at necropsy (Dodd and McDougal 2002).

Lent et al., (USAPHC 2012b) reported an Approximate Lethal Dose (ALD) of DNAN of 300 mg/kg in rats. After 14 days of dosing, the primary adverse event was splenomegaly in female rats with a LOAEL of 50 mg/kg-day. In the subchronic 90-day study, mortality occurred in three male rats (days 50, 63, and 77) and one female rat (day 26) all from the 80 mg/kg-day dose group. Rats in the highest dose group (80 mg/kg-day) experienced lethargy, labored/rapid respiration, prostrate and/or recumbent posture, hunched posture, ear twitching, squinting, curled tail, and gait irregularities. Female rat splenomegaly was the primary adverse event; the benchmark dose corresponding to a 10 percent effect level (BMDL$_{10}$) at a 95% level of confidence was 2.3 mg/kg-day.

Splenomegaly and extramedullary hematopoiesis were also observed in rats treated 14 days with the various isomers of dinitrotoluene (Lent et al. 2012b). The mode of action is expected to be through the reduction of nitro groups causing red blood cell lysis similar to that reported from nitrate and nitroaromatic exposure. Therefore, the spleen is considered a secondary target of effect for many nitroaromatics (Lima et al. 2011; Chandra et al. 1995).

No chronic oral experimental data were found. TOPKAT® modelling predicts a chronic LOAEL of 17.3 mg/kg-day at high confidence.
**DNAN – Inhalation Toxicity**

DNAN’s 4-hour acute LC₅₀ is greater than 2000 mg/m³ (Dodd and McDougal 2002). Rats exposed for two weeks (6 hours/day; 5 days/week) to aerosol concentrations of DNAN ≥ 545 mg/m³ in an acetone vehicle experienced 80 percent mortality, lethargy, labored breathing, irregular gait, nasal discharge, decreased fecal volume, yellow staining of the vent, decreased food consumption, and decreased body weight gain; control (acetone vehicle only) animals also exhibited signs of Central Nervous System (CNS) depression (irregular gait). Mild signs of toxicity and non-specific minimal metaplasia of laryngeal epithelium were observed at 165 mg/m³. No NOAEL was determined (Dodd and McDougal 2002).

In the PAX-21 studies, an inhalation LOAEL of 165 mg/m³ was determined for DNAN. A proposed workplace Reference Concentration for of 0.14 mg/m³ was determined based upon uncertainty factors of 3 for not determining a NOAEL, 10 each for subchronic to chronic and animal to human conversions, 3 for database sufficiency, and 1 for sensitive subpopulation (because workers were assumed to be healthy adults), and a correction for exposure time for an 8-hour work shift (Dodd and McDougal 2002).

Rats were exposed nose-only to a 2.39 mg/L aerosol atmosphere of DNAN for a single 4-hour exposure in a study to compare inhalation versus oral exposure toxicokinetics. No test compound-related mortalities occurred in rats exposed during the study and no adverse toxic signs, body weight changes, or gross necropsy findings were observed in exposed rats. However, mortality occurred in 4/10 rats exposed to the oral inhalation equivalent dose (USAPHC 2013a). The results of the LC₅₀ portion of this study suggest that acute inhalation exposure to the highest-achievable concentration of DNAN aerosol falling within an acceptable particle size range (2.39 mg/L) does not result in adverse effects. The results of the multi-time point blood absorption portion indicated that, under the stated study conditions and limitations, acute exposure to DNAN via oral gavage appears to induce higher DNAN whole blood concentrations in laboratory rats compared to those exposed via inhalation. Oral exposure to DNAN also resulted in an insignificant increase in whole blood concentrations of the metabolite 2,4-dinitrophenol (2,4-DNP) compared to those animals exposed via inhalation and four rats died from oral exposures that did not from nominal equivalent inhalation exposures. Additionally, female rats appear to convert a greater proportion of DNAN to 2,4-DNP than male rats when exposed via inhalation while male rats convert a greater proportion of DNAN to 2,4-DNP than female rats when exposed orally (USAPHC 2013a).

**DNAN – Dermal Toxicity**

Skin irritation studies were conducted as a part of the PAX-21 studies. Rabbits exposed to DNAN with a penetration flux of 0.74 µg/cm²-hour exhibited slight dermal irritation that was reversible within 24-48 hours. Studies in guinea pig using a modified Buehler method indicated DNAN was not a sensitizer (Dodd and McDougal 2002).

McCain et al., (USAPHC 2012a) studied in vitro dermal penetration of NTO in static Franz diffusion cells. Human epidermal membranes were prepared from frozen cadaver skin according to OECD guidelines and were mounted on static diffusion receptor cells so that the visceral side
was in contact with the receptor fluid. Test chemicals at infinite dose (100 mg) powder were carefully placed on the skin in the donor chamber, and at different times (1, 2, 4, 6 and 8 hours) about 0.1mL of receptor fluid (buffer) was collected and quantified for component content by HPLC. The dermal penetration rate showed that the steady state flux of neat DNAN was 1.10 μg/cm²-hour.

**DNAN – Ocular Toxicity**

Horner (1942) reported a relatively low rate of cataract development in humans who were consuming dinitrophenol, a metabolite of DNAN, in order to lose weight. Takahashi and coworkers (Takahashi et al. 1988) reported that 63 percent of Japanese Quail receiving a single dose of 120 mg/kg of DNAN and 100 percent receiving a single oral dose of 150 mg DNAN/kg bw developed reversible cataracts within one to four hours after administration. Mortality of approximately 20% was also observed among the 120 mg/kg group and about 55% among the 150 mg/kg group.

**DNAN – Mutagenesis**

DNAN tested positive in the Ames *Salmonella* histidine reversion test in strain TA100 without activation (GENETOX 2009; McMahon et al. 1979; CCRIS 2010; Chiu et al. 1978). DNAN tested negative in Chinese Hamster Ovary (CHO) cells (AS52/XPRT) and the mouse micronucleus test (Dodd et al. 2002). DNAN was negative in the mouse micronucleus assay at exposures of 10-90 mg/kg in both males and females (Dodd and McDougal 2002). The SOS Chromotest is a biological assay to assess the genotoxic potential of chemical compounds. The test is a colorimetric assay which measures the expression of genes induced by genotoxic agents in *Escherichia coli*, by means of a fusion with the structural gene for β-galactosidase. DNAN had positive genotoxic effect on *E. coli* in the SOS chromotest (DRDC 2011).

**DNAN – Carcinogenesis**

No experimental data were found. TOPKAT modelling is indeterminate, with six models each predicting positive and negative outcomes. There is some suggestion in the modelling that rats may be more likely than mice to develop cancer.

**DNAN – Environmental Fate and Transport**

With a low log $K_{OC}$ value, DNAN is not expected to adsorb to soil as a parent compound; however, it is expected to adsorb to the humic fractions of wet soils following the reduction of nitro groups by microbes. Aqueous solubility is projected to be moderate, so it is only expected to be a moderate to low groundwater transport hazard. This was confirmed experimentally by Site (2009) in experiments where DNAN and the other components of IMX-101 (NTO and NQ) were allowed to percolate through three different types of soil—a muck-peat high organic carbon soil, a clay-loamy soil, and a sandy-quartzose soil. All soil samples were collected on active Army installations and represent the spectrum of potential soil types. Samples of DNAN were placed in lysimeter devices and subjected to a simulated 3-month rainfall exposure using synthetic rain water. DNAN was not found in leachate samples, but rather to be primarily
located in the top 5 centimeters of the lysimeter devices, indicating it was migrating very slowly, presumably due to low solubility. A result of this concentration in the top level of soil was that DNAN was concentrated by rye grass sprouts that were planted on the top level of soil. Bioconcentration and bioaccumulation are expected to be low based upon the relatively low log K\textsubscript{OW}. With its low aqueous solubility and relatively high K\textsubscript{OW}, DNAN will most likely remain close to the discharge point in manufacturing and training ranges sites (DRDC 2011).

Photolysis of DNAN using SolSim shows photodegradation of DNAN and the formation of its degradation products with time in a first order reaction. The t\textsubscript{1/2} of DNAN disappearance with SolSim was 3.1 day (DRDC 2011).

**DNAN – Ecotoxicity**

Administration of DNAN to Japanese Quail (\textit{Coturnix japonica}) resulted in rapid production of cataracts. Quail receiving single oral doses of 120 or 150 mg/kg developed cataracts within 4 hours of treatment (100 percent). Mortality was also noted in these groups with losses being 1 of 5 at the lower dose and 5 of 9 at the higher dose (Takahashi et al. 1988).

Carp (\textit{Cyprinus carpio}) exposed to DNAN at a concentration of 117-270 mg/L experienced mortality (ECOTOX 2009).

Acute and chronic aquatic toxicity bioassays were conducted using standard fish (\textit{Pimephales promelas}) and invertebrate (\textit{Ceriodaphnia dubia}) models. Chemical analysis of test water indicated that DNAN concentrations were relatively stable during the bioassays. Acute toxicity was similar for the two species tested, with 48-hr lethal median concentrations (LC50) ranging from 37 to 42 mg/L DNAN. Chronic toxicity tests indicated that fish survival (7-day LC\textsubscript{50} = 10 mg/L) was significantly more sensitive to DNAN relative to the invertebrate (no significant impact on survival at 24 mg/L). When assessing the most sensitive chronic endpoints, the two test species indicated similar chronic toxicity, with lowest observable adverse impacts ranging from 10 to 12 mg/L DNAN and median effects on sublethal endpoints (growth, reproduction) ranging from 11 to 15 mg/L DNAN. Chronic no-effect concentrations ranged from approximately 6 to 8 mg/L DNAN, which is less than that reported for TNT (USACE 2013).

ECOSAR modeling predicts a 96-hour LC\textsubscript{50} in freshwater fish of 9.00 mg/L, a 48-hour LC\textsubscript{50} of 72.78 mg/L for \textit{Daphnids}, and a 96-hour EC\textsubscript{50} for green algae of 0.82 mg/L. TOPKAT modelling projects an LC\textsubscript{50} in fathead minnow of 24.9 mg/L at high confidence, and an EC\textsubscript{50} in \textit{Daphnia} of 7.0 mg/L, also at high confidence.

In a 96-hr freshwater green algae (\textit{P. subcapitata}) inhibition test, DNAN had an EC\textsubscript{20} of 1.4 mg/L. The results obtained for DNAN are similar to TNT (EC\textsubscript{20} of 0.54 mg/L) (DRDC 2011).

**DNAN – Degradation/Treatment**

DNAN biotransformation rates were studied in sludge under aerobic, microaerophilic, and anaerobic conditions. The biotransformation of DNAN was most rapid under anaerobic conditions with H\textsubscript{2} as a co-substrate. The results showed that the \textit{ortho}- nitro group in DNAN is
regioselectively reduced to yield 2-methoxy-5-nitroaniline (MENA), and then the para-nitro group is reduced to 2,4-diaminoanisole (DAAN). Both MENA and DAAN were identified as important metabolites in all redox conditions. Azo and hydrazine dimer derivatives formed from the coupling of DNAN reduction products in anaerobic conditions. Secondary pathways included acetylation and methylation of amine moieties, as well as the stepwise O-demethylation and dehydroxylation of methoxy groups. Seven unique metabolites were identified which enabled elucidation of biotransformation pathways. The results taken as a whole suggest that reductive biotransformation is an important fate of DNAN leading to the formation of aromatic amines as well as azo and hydrazine dimeric metabolites (Olivares et al. 2012).

Hawari (DRDC 2011) found that soil microbes readily transformed DNAN in aerobic microcosms supplemented with a nitrogen (NH$_4$Cl) and carbon sources (glucose and succinate). DNAN disappeared at the rate of 1.7 ± 0.2 nmol h$^{-1}$ g$^{-1}$ and was completely removed in 8 days.

Perreault et al., (2012) reported the aerobic biotransformation of DNAN in artificially contaminated soil microcosms. DNAN was completely transformed in 8 days in soil slurries supplemented with carbon and nitrogen sources. DNAN was completely transformed in 34 days in slurries supplemented with carbons alone and persisted in unamended microcosms. A strain of Bacillus (named 13G) that transformed DNAN by co-metabolism was isolated from the soil. HPLC and LC-MS analyses of cell-free and resting cell assays of Bacillus 13G with DNAN showed the formation of 2-amino-4-nitroanisole as the major end-product via the intermediary formation of the aryl nitroso (ArNO) and arylhydroxylamino (ArNHOH) derivatives, indicating regioselective reduction of the ortho-nitro group. A series of secondary reactions involving ArNO and ArNHOH gave the corresponding azoxy- and azo-dimers. Acetylated and demethylated products were identified. Overall, this paper provides the evidence of fast DNAN transformation by the indigenous microbial populations of an amended soil with no history of contamination with explosives and a first insight into the aerobic metabolism of DNAN by the soil isolate Bacillus 13G (Perreault et al. 2012).

The effectiveness of the Anaerobic Fluidized-Bed Bioreactor (AFBB) was evaluated for treatment and transformation of DNAN. DNAN fed into AFBBs, and was monitored for removal and transformation. Ethanol was used as the electron donor. Results show that AFBB technology effectively removed DNAN, but produced secondary compounds (reduced analogs of DNAN), which may be as harmful as the feed material, but are no longer resistant to aerobic degradation (USACE 2011).

The alkaline hydrolysis of DNAN was investigated with coordinated experimental kinetic measurements and molecular modelling calculations. The results suggest that a Meisenheimer complex is an intermediate in the formation of 2,4-DNP. Despite these advances, the remaining uncertainties in the mechanisms of these reactions--and potential variability between the hydrolysis mechanisms for different NACs--mean that it is not yet possible to generalize the results into predictive models (e.g., quantitative structure-activity relationships, QSARs) for hydrolysis of other NACs (Salter-Blanc et al. 2013).

Saad et al., (2012) described the use of two commercially available lignins, namely, alkali and organosolv lignin, for the removal of DNAN from water. Sorption of DNAN on both lignins
reached equilibrium within 10 hours and followed pseudo second-order kinetics with sorption being faster with alkali than with organosolv lignin, i.e. $k_2$ 10.3 and 0.3 g/(mg hr), respectively. In a separate study the sorption of DNAN was investigated between 10 and 40°C and found that the removal of DNAN by organosolv lignin increased from 0.8 to 7.5 mg/g but reduced slightly from 8.5 to 7.6 mg/g in the case of alkali lignin. Sorption isotherms for either alkali or organosolv lignin best fitted the Freundlich equation with enthalpy of formation, $\Delta H_f$, equal to 14 or 80 kJ/mol. To help understand DNAN sorption mechanisms they characterized the two lignins by elemental analysis, BET nitrogen adsorption-desorption and $^{31}$P NMR. Variations in elemental compositions between the two lignins indicated that alkali lignin should have more sites (O- and S-containing functionalities) for H-bonding. The Brunauer-Emmett-Teller (BET) surface area and calculated total pore volume of alkali lignin were almost 10 times greater than that of organosolv lignin suggesting that alkali lignin should provide more sites for sorption. $^{31}$P NMR showed that organosolv lignin contains more phenolic –OH groups than alkali lignin, i.e., 70% and 45%, respectively. The variations in the type of OH groups between the two lignins might have affected the strength of H-bonding between DNAN and the type of lignin used (Saad et al. 2012).

DNAN is soluble in water and, thus can migrate through subsurface soil to cause groundwater contamination. DNAN has a weak Kow and, thus can bioaccumulation potential is considered negligible in biological receptors in the environment. DNAN can abiotically degrade with ZVI and with light. DNAN can also biotically degrade by soil indigenous bacteria. Under both abiotic and biotic conditions DNAN showed regioselective initial reduction of the ortho-nitro group to form 2-amino-4-nitroanisole. This in turn reduces to give the diamino derivative. In contrast, 2,4-DNT and TNT were selectively reduced at the para-nitro group. Taking all experimental evidences gathered this far we assume that DNAN can undergo natural attenuation (Perreault et al. 2012; Saad et al. 2012).

**DNAN – Metabolism**

Adult male rhesus monkeys received single oral doses of either DNAN at 50, 25, or 5 mg/kg followed by serial blood and urine sampling up to 48 hours post exposure (Hoyt et al. 2013). Results showed that DNAN had a complex temporal profile with consistently low blood, serum, and urine levels and without an evident peak. However, 2,4-DNP, the reductive metabolite of DNAN, appeared in blood at concentrations 10-fold higher than the parent compound at 50 and 25 mg/kg, but was not detected at doses of 5 mg/kg. Rodents dosed with DNAN showed a similar blood profile. Therefore, it appears that the primary metabolite for DNAN exposure is DNP in primates and cannot be detected currently in urine at low levels of exposure (i.e. levels approximately at the Occupational Exposure Levels).

During the 1930s, 2,4-DNP was used extensively as a diet pill. In 1938, the U.S. Food and Drug Administration announced that manufacturers of drugs such as 2,4-DNP that were known to have caused adverse effects even when used under medical supervision would be prosecuted by the FDA whenever the drugs were found in interstate commerce. The use of 2,4-DNP was essentially discontinued at this time. 2,4-DNP has an oral lethal dose of 14-43 mg/kg in humans with the cause of death generally attributed to the pyretic effect of 2,4-DNP, produced by an increase in metabolic rate due to uncoupling of oxidative phosphorylation in mitochondrial

NTO – Oral Toxicity

The oral LD_{50} for NTO is reported to be >5000 mg/kg in both the rat and (London and Smith 1985). Sarlauskas and coworkers (Sarlauskas et al. 2004) conducted an investigation into the mechanism of toxicity of NTO and the related ANTA (5-nitro-1,2,4-triazol-3-amine) by evaluating their reactions with one-electron and two-electron reductions by flavoproteins and oxyhemoglobin oxidation. Both NTO and ANTA were found to undergo cyclic oxidation-reduction reactions with production of superoxide (O_{2}^{•−}), but to a lesser degree than TNT. Less production of free radicals is correlated with the lower toxicity of NTO and ANTA compared to TNT (Sarlauskas et al. 2004).

Results from a 14-day subacute oral toxicity study in rats showed significantly decreased testes weights in the high dose groups (≥500 mg/kg bw-day) (USAPHC 2010).

A 90-day oral gavage study in rats was performed with doses of 0, 30, 100, 315, and 1000 mg NTO/kg bw-day. Testes and epididymides weights were reduced in the 315 and 1000 mg/kg-day treated rats. Moderate to severe testicular hypoplasia, characterized by interstitial degeneration and loss of spermatogenic epithelium in the seminiferous tubules, was observed in the testes in 66% and 100% of males from the 315 and 1000 mg/kg-d dose groups, respectively. Epididymal aspermia was also observed at these dose levels (USAPHC 2010).

NTO – Inhalation Toxicity

Rats were exposed nose-only to a 0.184 milligrams per liter (mg/L) aerosol atmosphere of NTO for a single 4-hour exposure. No test compound-related mortalities occurred in rats exposed during the study and no adverse toxic signs, body weight changes, or gross necropsy findings were observed in exposed rats. The results LC_{50} portion of this study indicate that acute inhalation exposure to the highest-achievable concentration of NTO aerosol falling within an acceptable particle size range (0.184 mg/L) did not result in any observed adverse effects to rats. The results from the multi-time point blood absorption study indicated that exposure to NTO via inhalation resulted in greater than 3-fold higher blood concentrations of NTO than via oral gavage (USAPHC 2013b).

NTO – Dermal Toxicity

The manufacturers’ MSDS suggests NTO may cause dermal irritation (BAE Systems, 2007). Administration of 500 mg NTO to the skin of a rabbit for 24 hours had a mild irritant effect (London and Smith 1985).

McCain et al., (USAPHC 2012a) studied in vitro dermal penetration of NTO in static Franz diffusion cells. Human epidermal membranes were prepared from frozen cadaver skin according to OECD guidelines and were mounted on static diffusion receptor cells so that the visceral side was in contact with the receptor fluid. Test chemicals at infinite dose (100 mg) powder were
carefully placed on the skin in the donor chamber, and at different times (1, 2, 4, 6 and 8 hrs) about 0.1mL of receptor fluid (buffer) was collected and quantified for component content by HPLC. The dermal penetration rate showed that the steady state flux of neat NTO, was 332 μg/cm²/hr.

**NTO – Ocular Toxicity**

No experimental data were found. TOPKAT modeling predicts NTO is likely to be a moderate to severe ocular irritant.

**NTO – Developmental/Reproductive Toxicity**

Previous studies have consistently identified the testes and epididymides as potential targets of NTO (USAPHC 2010, 2013). Effects to the testes have been reported to occur from repetitive exposures as brief as 14-days. Tests were then conducted to ascertain the possibility that these effects could be manifested through endocrine disruptive mechanisms. NTO was tested in nine endocrine disruptor bioassays. Five of these assays were *in vitro*: estrogen receptor binding, androgen receptor binding, estrogen transactivation, aromatase, and steroidogenesis. NTO had no effect on these endpoints. Using a Weight of Evidence (WoE) approach, NTO does not appear to directly affect testosterone- or estrogen-mediated signalling pathways. Metabolites were not directly tested with these methodologies (USAPHC 2012c).

NTO targeting of reproductive organs was tested *in vivo* using the Hershberger bioassay, a short-term screening test that evaluates the accessory tissues of the male, and the uterotrophic assay, a short term screening test used to identify substances with estrogen-active potential in female rats. Male and female rats were exposed to NTO in corn oil by oral gavage at 250, 500, and 1,000 mg/kg-day for 10 days. The results of these studies were negative; there was no evidence to suggest that NTO would act like an estrogenic or anti-androgenic endocrine disruptor at these dose levels (Quinn et al. 2014).

To determine whether NTO has the potential to interact with the endocrine system *in vivo*, the effects of NTO on puberal development and thyroid function were investigated in juvenile rats. Male and female Sprague-Dawley rats (15/sex/group) were administered NTO in corn oil via oral gavage from weaning through puberty. Females were given 0, 500, and 1000 mg/kg-day NTO from post natal day (PND) 22 through PND 42/43. Males were given 0, 250, and 500 mg/kg-day NTO from PND 23 through PND 53/54. Doses were adjusted based on daily body mass measurements. Females were examined daily for initiation of vaginal opening (VO). Estrous cyclicity was evaluated daily upon completion of VO. Males were examined for initiation of preputial separation (PPS) beginning on PND 30. Final body weight females in the 1000 mg/kg-day group was reduced 7% compared to the control (p=0.031). Body weight of male rats was unaffected by NTO treatment. Age and body weight at the time of VO as well as all measures of estrous cyclicity (age at first estrus, cycle length, percent cycling, percent cycling regularly) were not affected by treatment with NTO at 500 and 1000 mg/kg-day. Age at PPS and body weight at PPS did not differ among NTO treated rats and the control group. Biologically significant effects on organ weight were limited to reductions in testes and epididymides weights. Testis weights were reduced to 70% and 35% of control in the 250 and 500 mg/kg-day
groups (p<0.001 and p<0.001, respectively), while epididymides were reduced to 76% of control in the 500 mg/kg-day group (p≤0.001. There were no differences in hormone levels between NTO treatment groups and the control groups for males or females. These preliminary findings suggest that NTO is not acting as an estrogen or thyroid active compound under the test conditions (USAPHC 2013).

NTO was examined for reproductive developmental toxicity in a study conducted in a manner consistent with the methods outlined in OECD Test Guideline 422. Forty adult Sprague Dawley rats of each sex (N=80) were randomly distributed into 3 dose groups and a corn oil control group (10 rats of each sex per dose group (USAPHC 2013). Rats were exposed to nominal daily oral gavage exposures to 0, 31, 125, and 500 mg/kg-d NTO 7 days/week for 4 weeks. In addition to the main study, 20 male rats were added to serve as satellite recovery groups for the highest dose group and the control group. These animals were dosed concurrently with the main study animals for a 4-week period and held for an additional 4-week period to evaluate the reversibility, persistence, or delayed occurrence of the toxic effects associated with repeated exposure to NTO. Preliminary results indicate that significant adverse events were limited to the male reproductive organs. Testes mass, as well as testes to body and brain mass ratios, in the 500 mg/kg-day dose group was significantly reduced compared to all other dose groups, including controls. Epididymides mass, as well as epididymides to body and brain mass ratios in the 500 mg/kg-day dose group was significantly reduced compared to all other dose groups, including controls. The number of sperm per gram in the cauda epididymis in male rats in the 500 mg/kg-day group was significantly reduced compared to all other dose groups. The significant reductions in testes and epididymides mass and mass ratios, as well as sperm counts, continued to be observed in the 500 mg/kg-day male recovery group compared to the recovery control group following a 4-week recovery period. Absolute spleen mass and spleen to body mass ratios were significantly reduced in the female 500 and 31.25 mg/kg-day groups relative to controls. Absolute brain mass in the female 31.25 mg/kg-day group was reduced compared to controls. Sporadic significance between dose groups was also observed for male and female clinical chemistry and hematology parameters, body weight gain, and food consumption but was most likely random since it was not observed relative to controls and no dose-related trends were observed. However, no differences were observed in any of the reproductive or litter measures obtained during the study. These measures are summarized in the table below (USAPHC 2013).

NTO – Mutagenesis

NTO was evaluated for mutagenicity in Salmonella typhimurium and Escherechia coli plate incorporation assay both with and without S9 activation. Results were negative in Salmonella at levels of up to 500 µg/plate without activation, and up to 5000 µg/plate with activation. In E. coli, results were also negative at maximum concentrations up to 2500 µg/plate without activation and 5000 µg/plate with activation (Reddy et al. 2011).

NTO was also evaluated in the L5178Y TK+/- mouse lymphoma mutagenesis assay. Cells were treated with NTO at concentrations up to 5000 µg/mL, both with and without activation. Results of the assay were negative, either with or without activation (Reddy et al. 2011).
NTO was tested in CHO cells for clastogenicity. The test was conducted both with and without exogenous metabolic activation at concentrations up to 5000 µg/mL; results were negative (Reddy et al. 2011).

The SOS Chromotest is a biological assay to assess the genotoxic potential of chemical compounds. The test is a colorimetric assay which measures the expression of genes induced by genotoxic agents in E.coli, by means of a fusion with the structural gene for β-galactosidase. NTO was negative in the SOS chromatotest (DRDC 2011).

**NTO – Carcinogenesis**

No experimental data were found. TOPKAT modeling of carcinogenicity produced an indeterminate result.

**NTO – Environmental Fate and Transport**

NTO has a high solubility in water and a low log K\textsubscript{OC}, indicating it will bind poorly to soil and have a high mobility in ground water causing groundwater contamination (DRDC 2011). It typically liberates protons and becomes acidic in water. This was confirmed experimentally by SIT (2009) in experiments where NTO was allowed to percolate through three different types of soil—a muck-peat high organic carbon soil, a clay-loamy soil, and a sandy-quartzose soil. All soil samples were collected on active Army installations and represent the spectrum of potential soil types. Samples of NTO were placed in lysimeter devices and subjected to a simulated 3-month rainfall exposure using synthetic rain water. The amount of binding varied with soil type, as might be expected, with NTO being most strongly retained by the muck-peat, and least by the sandy-quartzose, but demonstrating high mobility in all soil types. The K\textsubscript{d} of NTO is <0.1 and demonstrated low soil sorption (DRDC 2011).

Photolysis of NTO was studied using artificial sunlight generated from a SolSim Solar Simulating Photoreactor (Luzchem Research, Inc, Canada). More than 90% of NTO was degraded in 7 days in a SolSim photo-reactor with concomitant formation of nitrite (NO\textsubscript{2}\textsuperscript{-}), nitrate (NO\textsubscript{3}\textsuperscript{-}) and ammonium (NH\textsubscript{4}\textsuperscript{+}) ions. Photodegradation rates of NTO in water and in water extract from soil were very similar suggesting less than 5% of variation (DRDC 2012).

Rye grass grown on the top level of the lysimeters was also collected in this experiment and analyzed for uptake of NTO (SIT 2009). Uptake by the grass was considerable, with the amount varying from 398 mg/kg grass to 1244 mg/kg grass, depending upon soil type. Uptake by the grass was greater in the soil where NTO was more strongly retained; uptake was lowest when NTO rapidly percolated through the soil.

**NTO – Ecotoxicity**

Few data could be found on the ecotoxicology of NTO. TOPKAT modelling was unable to predict a toxicity value in the fathead minnow test, but estimates an EC\textsubscript{50} of 7.7 mg/L in Daphnia with low confidence. Hawari found that the EC\textsubscript{20} of NTO in the Microtox assay was 2405 mg/L, only slightly toxic (DRDC 2011).
The 48-hour survival of *Pimephales promelas* was examined in moderately hard water containing NTO at concentrations ranging from 0 to 5.0%. The LC$_{50}$ was 1.14 g/L calculated using the Trimmed-Spearman Karber method (BAE Systems 2007).

*Ceriodaphnia dubia* was used in a 7-day survival and reproduction study and the unicellular green algae *Selenastrum capricornutum* in a 96-hour growth inhibition study. Addition of NTO to aqueous systems was found to cause a concentration-related decrease in the pH of the system. This is apparently due to the ability of the ring-bonded hydrogen adjacent to the ring nitrogen to dissociate, producing a hydrogen ion. In 24- and 48-hour range finding studies, the pH of the added NTO solution was found to have a significant impact on the results, as indicated in Table 1. Because of the impact of pH, all subsequent testing was done with NTO that had been adjusted for pH. In the definitive 7-day exposure study, the IC$_{50}$ value was found to be 57 mg/L. The NOEC and LOEC values were found to be 34 mg/L and 66 mg/L, respectively. While no mortality was observed at concentrations less than 523 mg/L, no eggs were produced at 262 mg/L, and at 133 mg/L, eggs were produced but failed to develop (Haley et al. 2009).

Table 1. Results of NTO Ecotoxicity Range-Finding Experiment

<table>
<thead>
<tr>
<th>LC$_{50}$ in <em>Ceriodaphnia</em> (mg/L)</th>
<th>24-hr</th>
<th>48-hr</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH-adjusted</td>
<td>830</td>
<td>460</td>
</tr>
<tr>
<td>unadjusted</td>
<td>66</td>
<td>62</td>
</tr>
</tbody>
</table>

In a 96-hour growth inhibition study using pH-adjusted NTO in *Selenastrum capricornutum*, the IC$_{50}$ was estimated to be 3465 mg/L, based upon a slight extrapolation of the IC$_{20}$ value of 2195 mg/L (Haley et al. 2009). In a 96-hour freshwater green algae (*P. subcapitata*) inhibition test, NTO had an EC$_{20}$ of 1587 mg/L (DRDC 2011).

EPA’s ECOSAR program models NTO in the hydrazine class, with a 96-hour EC$_{50}$ in green algae predicted to be 10.34 mg/L, the 48-hour LC$_{50}$ in *Daphnia* to be 878.98 mg/L, and a 96-hour LC$_{50}$ in fish of 61.52 mg/L.

**NTO – Degradation/Treatment**

Hawari (DRDC 2012) found that NTO was removed slowly (25% NTO removed after 28 days) in aerobic microcosms supplemented with glucose and succinate only. NTO persisted in unamended aerobic and anaerobic microcosms.

Biodegradation was also explored by Haley and coworkers (Haley et al. 2009) using neutralized NTO in *Ceriodaphnia* growth medium. Under these conditions, approximately 10 percent of the NTO was lost over the course of a 7-day experiment. Assuming first order kinetics, this gives a rate constant of 0.015 d$^{-1}$, and a half-life of 46 days.

NTO has been found to be degraded by light by means of a solar simulator (DRDC 2013).

NTO is reported to readily decomposed by photodegradation in the presence of titanium dioxide. When exposed to light of wavelength >290 nm, NTO solutions of concentration 150
mg/L in the presence of 0.4 g/L TiO$_2$ were completely degraded within 3 hours (LeCampion, Giannoti, et al. 1999). NTO is also readily degraded by a strain of the bacterium Bacillus licheniformis (LeCampion, Vandais, et al. 1999). This degradation is reported to proceed through an oxygen-insensitive nitroreduction leading to production of the primary amine, ATO (5-amino-1,2,4-triazol-3-one), followed by cleavage of the triazone ring (LeCampion et al. 1998).

Treatment of a NTO waste stream was found to be unaffected by alkaline hydrolysis; however, treatment with bimetallic Fe/Cu particles rapidly degraded the compound (SIT 2011).

Singh and coworkers recently prepared a short review on the chemistry and decomposition products of NTO (Singh et al. 2001). As is expected for a compound containing carbon and nitrogen, CO$_2$, NO$_2$, and N$_2$O are prominent combustion products, as well as water, carbon monoxide, diatomic nitrogen, hydrogen gas, and a product believed to be a polymeric form of 1,2,4-triazine-5-one (TO) with empirical formula C$_2$H$_3$N$_3$O. NO$_2$ free radicals are also believed to be produced by the cleavage of the nitrate group during the combustion process, but these would be too short-lived to have a biological effect.

**NTO – Metabolism**

A study using $^{14}$C-NTO found that metabolic degradation of this compound in rats appears to involve two separate enzymatic pathways. In the presence of oxygen, NTO is metabolized to two separate products: 5-amino-1,2,4-triazol-3-one (ATO) and 5-hydroxy-1,2,4-triazol-3-one (urazole). The presence of oxygen did not affect the overall conversion of NTO, but did alter the proportion of the metabolites. Under anaerobic conditions, the ATO is the primary product while urazole comprised only 5 percent of the product. Under aerobic conditions, urazole represented 40 percent of the product with a decrease in nitroreduction decreased by 75%. Two separate pathways are represented here, since incubation of ATO with activated microsomes did not result in production of urazole, indicating that ATO does not represent an intermediate in this pathway and urazole is formed directly from NTO in mammalian systems (LeCampion et al. 1997).

Adult male rhesus monkeys received oral doses of NTO from 5 to 50, mg/kg followed by serial blood and urine sampling up to 48 hours post exposure. Results showed that NTO was absorbed quickly and eliminated unchanged by 8 hours, with urinary concentrations at least 100-fold higher than those of blood or serum. (Hoyt et al. 2013).

**NQ – Oral Toxicity**

The acute LD$_{50}$ is 3850 mg/kg in mice and 3120 mg/kg in guinea pig. Mortality is the result of respiratory cyanosis. The acute LD$_{50}$ in rats is >5000 mg/kg (Brown et al. 1988, Hiat et al. 1988, Lewis 2004).

Subacute and subchronic oral toxicity of NQ was evaluated in male and female rats Sprague-Dawley rats (Morgan, Brown, et al. 1988; Morgan, Pierce, et al. 1988). Nitroguanidine was administered in the diet at dose levels of 0, 100, 316, and 1000 mg/kg-day for 90 days. There were 15 animals/sex/dose. The addition of nitroguanidine to the diet consistently reduced food
consumption and caused significant (p ≤ 0.05) increases in water consumption. Blood samples taken at necropsy for hematology and serum chemistry analysis exhibited that no significant abnormalities could be attributed to nitroguanidine exposure. Microscopic examination of tissues from the control and 1000 mg/kg-day dose group animals revealed no lesions attributable to the administration of nitroguanidine. Thus, NQ exposure did not result in any observed adverse effects to rats when exposed to NQ in feed at equivalent doses as high as 1000 mg/kg-day for 14 days. Based on a decrease in the rate of growth of female rats at 1000 mg/kg-day on weeks 5, 6, 8, 9 and 12, the NOAEL was reported as 316 mg/kg-d (Morgan, Brown, et al. 1988; Morgan, Pierce, et al. 1988).

The 90-day subchronic oral toxicity of nitroguanidine was evaluated in male and female ICR mice (Frost et al. 1988). There were 15 animals/sex/dose. Nitroguanidine was administered in the diet at dose levels of 0, 100, 316 and 1000 mg/kg-d for 90 days. The addition of nitroguanidine to the diet had no effect on food consumption or weight gains, but there was a dose-response increase in water consumption. Several serum chemistry parameters did exhibit statistically significant (p ≤ 0.05) differences compared to control values, but these changes were isolated occurrences with no consistent dose-related trends reported. With the exception of the brain-to-body weight ratio in the high-dose males at interim sacrifice, organ weights and their respective ratios were not affected by dosing. Microscopic examination of tissues from the control and 1000 mg/kg-day dose group animals revealed no lesions attributable to the administration of nitroguanidine. The findings of increased water consumption suggest that nitroguanidine, which is excreted unchanged in mouse urine, may be acting as an osmotic diuretic. The finding of increased brain-to-body weight ratios in male mice at 1000 mg/kg-day at interim sacrifice is supportive of a 316 mg/kg-day NOAEL (Frost et al. 1988).

Korolev and coworkers (1980), conducted a chronic toxicity study in animals given 5, 50 or 500 µg/kg. The species of animals used, the method of administration and duration of study were not reported. However, a NOAEL was reported to be 5 µg/kg-day, the lowest dose tested. TOPKAT modelling of nitroguanidine predicts a chronic LOAEL of 620.9 µg/kg-day with high confidence.

**NQ – Inhalation Toxicity**

No experimental data were found. TOPKAT modeling predicts an LC50 of 557.5 mg/m³-h with high confidence.

**NQ – Dermal Toxicity**

Nitroguanidine was found to not be irritating to rabbit skin (Morgan et al. 1986; Hiatt, Wheeler, et al. 1988). McCain et al., (USAPHC 2012a) studied in vitro dermal penetration of NTO in static Franz diffusion cells. Human epidermal membranes were prepared from frozen cadaver skin according to OECD guidelines and were mounted on static diffusion receptor cells so that the visceral side was in contact with the receptor fluid. Test chemicals at infinite dose (100 mg) powder were carefully placed on the skin in the donor chamber, and at different times (1, 2, 4, 6 and 8 hours) about 0.1mL of receptor fluid (buffer) was collected and quantified for
component content by HPLC. The dermal penetration rate showed that the steady state flux of neat NQ was 31.25 μg/cm²-hour.

**NQ – Ocular Toxicity**

Nitroguanidine was found to not be irritating to rabbit eyes using the standard Draize technique (Morgan et al. 1986, Hiatt et al. 1988).

**NQ – Developmental/Reproductive Toxicity**

Nitroguanidine has been tested and found to not be teratogenic in both the rat and rabbit (Korte et al. 1990; Schardein 1993). However, in rats nitroguanidine produced maternal and fetal toxicity at the 1000 mg/kg-day dose level; the NOAEL was 316 mg/kg-day (Coppes et al. 1988a). In rabbits, although there were no dose-related malformations, fetuses in the 1000 mg/kg-day group were lighter in weight and had an increased incidence of retarded ossification of the sternebrae, olecranon, patellae, and phalanges (Coppes et al. 1988b).

The potential of nitroguanidine to produce reproductive toxicity was evaluated in Sprague-Dawley rats. Nitroguanidine was mixed into the diet at 0, 1.3, 4.0 and 12.7 parts per thousand (ppt). In young adult rats these dose levels in ppt approximated the 100, 316, and 1000 mg/kg-day nitroguanidine dose levels in developmental toxicity studies in rats and rabbits. The diet was fed to parental males and females starting at 56 to 58 days of age and continued throughout their lives and to the F1 and F2 generation animals. Parental males and females were paired for mating. All matings were within the same dose group. Nitroguanidine caused a decrease in some of the weekly body weights in the high dose animals, but the decrease was not consistent throughout the study.

**NQ – Mutagenesis**

Nitroguanidine was not mutagenic for *Salmonella typhimurium* strains nor was it mutagenic for mouse lymphoma cells in the presence or absence of rat hepatic homogenates (Ishidata and Odashima 1977; McGregor et al. 1980). Nitroguanidine-associated recombinant activity was not observed in *Saccharomyces cerevisiae* (McGregor et al. 1980), and it was negative in dominant lethal assays with rats and mice (Brusick and Matheson 1978).

There was no evidence of DNA damage in nitroguanidine-treated WI-38 cells (USDOD 1978); however, nitroguanidine did show evidence of clastogenicity in a screening test in Chinese hamster lung cells. Chromosomal aberrations observed included chromatid gaps, chromatid or chromosomal breaks and translocation homogenates (Ishidata and Odashima 1977).

The SOS Chromotest is a biological assay to assess the genotoxic potential of chemical compounds. The test is a colorimetric assay which measures the expression of genes induced by genotoxic agents in Escherichia coli, by means of a fusion with the structural gene for β-galactosidase. NQ had positive genotoxic effect on E. coli in the SOS chromotest but at a high concentration (DRDC 2011).
NQ – Carcinogenesis

Applying the criteria described by the U.S. EPA for the assessment of carcinogenic risk (USEPA, 1986), NQ is classified in group D: not classifiable as to human carcinogenicity (USEPA 1992, USEPA 2011, USEPA 2009).

NQ – Environmental Fate and Transport

If released to air, an estimated vapor pressure of 0.03 mmHg at 25ºC indicates NQ will exist solely in the vapor phase in the ambient atmosphere. Vapor phase NQ will be degraded by reaction with photochemically-produced hydroxyl radicals, with an estimated half-life of 18-20 hours. NQ has poor sorption in soil and should be mobile in soil with an estimated KOC of 25 (DRDC 2011). If released into water, NQ is not expected to adsorb to suspended solids or sediments in the water; NQ is expected to migrate through subsurface soil and reach the water table causing groundwater contamination (DRDC 2011). Volatilization from water surfaces is not expected to be an important environmental fate based upon the low Henry’s Law constant. An estimated bioconcentration factor of 3.2 suggests the potential for bioconcentration is low (HSDB 2003; van der Schalie 1985).

NQ – Ecotoxicity

The acute toxicity of NQ to ten species of freshwater aquatic organisms was determined by van der Schalie (1985). Fish exposed to NQ for 96 hours included fathead minnows (Pimephales promelas), bluegill (Lepomis macrochirus), Channel catfish (Ictalurus punctatus), and rainbow trout (Salmo gairdneri). Invertebrates were exposed for 48-hours and included Daphnia magna, amphipods (Hyallela azteca and Gammarus minus), midge larvae (Paratanytarsus dissimilis), and aquatic worms (Lumbriculus variegatus). The acute toxicity of NQ was very low; fewer than 50% of the exposed organisms experienced mortality at concentrations up to the solubility limit of NQ in water (1700 mg/mL at 12ºC for trout to about 3000 mg/L at 22 ºC for most other species). The alga (Selenastrum capricornutum) was slightly more sensitive, with 120-hour EC50’s of about 2000 mg/L. Complete photolysis of NQ with ultraviolet light greatly increased toxicity, with LC50/EC50 values decreasing to 20-35 mg/L (nominal concentration estimates).

Burrows et al.,(1988), investigated this further and reported NQ is readily degraded in water by ultraviolet and natural sunlight. The principal end products of photolysis from unbuffered nitroguanidine solutions are guanidine, urea, and nitrite ion, with lesser quantities of cyanoguanidine, nitrate ion and ammonia, accounting for 80% of the carbon and virtually all of the nitrogen. Nitrosoguanidine is an early intermediate which is even more readily photolyzed, to guanidine. Photolysis of nitroguanidine at pH 10 proceeds at nearly the same rate as the unbuffered reaction, but the product mix is different; less than 25 percent of nitroguanidine carbon is accounted for as urea, guanidine and cyanoguanidine. Elemental nitrogen is a significant product. All the identified photolysis products of NQ (except urea) are more toxic to aquatic organisms than the parent compound. However, only nitrite ion is present at a level high enough to account for the greatly enhanced toxicity of photo-NQ. But given the photolytic half-life of 1-2 days for NQ in natural waters, and considering the dilution that would take place in
that timeframe, it is highly unlikely that wastewaters discharged to a body of moving water could present a hazard to aquatic life unless the NQ levels substantially exceeded the present National Pollutant Discharge Elimination System (NPDES) daily average limit of 25 mg/L for Sunflower Army Ammunition Plant.

In a 96-hr freshwater green algae (P. subcapitata) inhibition test, NQ had an EC$_{20}$ of 760 mg/L (DRDC 2011).

**NQ – Degradation/Treatment**

Nitroguanidine is not susceptible to aerobic biodegradation in activated sludge, and was stable under sterile reducing conditions. After acclimation, NQ co-metabolizes to form nitrosoguanidine. In an anaerobic continuous culture test, no NQ was present after 7 days (Kaplan et al. 1982).

Nitroguanidine is not susceptible to aerobic biodegradation in activated sludge, and was stable under sterile reducing conditions. After acclimation, NQ co-metabolizes to form nitrosoguanidine. In an anaerobic continuous culture test, no NQ was present after 7 days (Kaplan et al. 1982).

Burrows et al.,(1988), reported that NQ and nitrosoguanidine are readily and rapidly photolyzed to guanidine and nitrite. It is highly unlikely that accumulation of nitrite in a body of water could present a hazard unless the NQ levels substantially exceeded the NPDES daily limits.

Combustion products predicted by the Lawrence Livermore National Laboratory’s Cheetah code are consistent with the expected products of compounds containing carbon, nitrogen and hydrogen, e.g. the oxides of these elements. Combustion products that would potentially be of concern such as ozone, superoxide, and nitrous oxide, are predicted to be present at such low levels that there is no reason for a health concern as a result of a transient exposure.

**IMX Mixture – Oral Toxicity**

IMX-101 in a corn oil suspension had an LD$_{50}$ in male rats of 1237 mg/kg and in female rats of 924 mg/kg, with a combined (male-female average) value of 1100 mg/kg (USACHPPM 2012e).

In a 14-day IMX-101 repeated oral dose study the notable adverse events were: 1) lethality in the 500 mg/kg-day and 1000 mg/kg-day dosage groups; 2) splenomegaly (increased spleen weight) primarily in females; and 3) testicular atrophy, histopathologic moderate to severe tubular degeneration, and decreased sperm density and motility (USAPHC 2012e). The LOAEL from this study was 100 mg/kg-day based on testicular mass with a calculated BMDL$_{10}$ of 30.6 mg/kg-day.

The oral toxicity data for the IMX-101 mixture are suggestive of an enhanced potentiated or synergistic relationship. The lethality and splenomegaly observed in the oral testing are likely
due to the DNAN component of the IMX-101 mixture; the adverse events occur at concentrations similar to those of DNAN alone. The testicular toxicity of IMX-101 is consistent with that described for NTO (USAPHC 2010); however, the DNAN and NQ components of the mixture appear to reduce the level at which testicular effects (changes in testicular mass) occur, i.e., at an order of magnitude (10-fold) lower as a result of exposure to the mixture. NTO caused a reduction in testes mass at the 1000 mg/kg-day level following the 14-day administration IMX-101 caused reductions in testicular mass at 100 mg/kg-day of IMX-101; a 24 mg/kg-day NTO equivalent dose (USAPHC 2012e).

McCain et al., (USAPHC 2012a) studied in vitro dermal penetration of IMX-101 as both the mixture and its individual components in static Franz diffusion cells. Human epidermal membranes were prepared from frozen cadaver skin according to OECD guidelines and were mounted on static diffusion receptor cells so that the visceral side was in contact with the receptor fluid. Test chemicals at infinite dose (100 mg) powder were carefully placed on the skin in the donor chamber, and at different times (1, 2, 4, 6 and 8 hrs) about 0.1mL of receptor fluid (buffer) was collected and quantified for component content by HPLC. The dermal penetration rate (μg/cm²-hour) was calculated for each chemical. Analysis of absorbed chemical in the receptor fluid showed that the steady state fluxes of neat NTO, DNAN and NQ were 332, 1.10 and 31.25 μg/cm²-hour, respectively. When 100 mg of the IMX-101 mixture was applied to the cell, the steady fluxes for NTO, DNAN and NQ were 135.9, 1.80, and 236 μg/cm²-hour, respectively. NTO and NQ showed about 0.4 and 7 times greater rates of penetration in the mixture than as individual compounds.

SUMMARY AND CONCLUSIONS

This work presents a review of the literature and preliminary toxicology information for IMX-101. It represents an example of a data package that is needed to enable manufacturers, health professionals, and environmental managers to conduct mission-essential activities in a sustainable manner. It is recommended that all munition managers consider such a toxicological data requirement in the development and use of new compounds to conduct useful life cycle assessments.

Presently, the data suggest that primary human toxicity endpoints from DNAN exposure are to be expected to be similar to that of other nitroaromatic compounds, specifically anemia from red blood cell lysis from nitro groups. Comparatively, data suggest that DNAN toxicity is slightly less than that of TNT. The primary sensitive sub-lethal effects from oral NTO exposure are direct effects to sperm production, which occurs consistently in animal models at high repetitive daily exposures. Marked lethal or sublethal effects from NQ exposure are lacking, suggesting relatively low toxicity risks from exposure. However, there are reports of a photolytic breakdown product in water that suggests a slightly greater risk to aquatic organisms. No chronic data are available for DNAN or NTO and chronic data available for NQ are incomplete and equivocal.
FUTURE WORK

*In vitro* data suggesting skin absorption from NTO and IMX-101 mixture require further confirmatory tests, preferably in an animal model. An aquatic study is needed to confirm the presence and analytical concentrations of a more toxic breakdown intermediate from photolysis of NQ and confirm the greater toxicity previously associated with it. Chronic animal data are needed for NTO and DNAN to enable military operations using and manufacturing these compounds to operate in a sustainable manner.

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12.0 Discussion: Future Directions in Insensitive Munitions (IM) Environmental, Safety, and Occupational Health (ESOH): Data Needs to Support Mission Requirements

New developments in insensitive munition compounds and formulations have realized immediate benefits in saving lives through improvements in safe handling and storage requirements. However, it is essential that these benefits be also balanced with any potential adverse environmental and/or occupational outcomes associated with manufacturing and use.

Representatives from research and acquisition communities provided recommendations from lessons-learned through the process of IM development. Mr. Ruppert suggested the need for OSD-level support to oversee the collection and interpretation of environmental, safety, and occupational health-related data. He pointed to the success of the Army Environmental Quality Program, Pollution Prevention Pillar (Army Research, Development, Engineering Command, Environmental Acquisition Sustainment and Logistics Program) in their work to develop ASTM E-2552 (Standard Guide for Assessing the Environmental and Human Health Impacts of New Energetic Compounds); their relationship with the energetics research, development, testing and evaluation (RDT&E) communities with the U.S. Army Public Health Command (USAPHC), Toxicology Portfolio to ensure data are collected that is consistent and proportional with the level of effort associated with the research and development for each constituent. Results for this program suggest < 10% of the costs associated with the R&D effort can be used to develop toxicity data and evaluation for new substances.

Mr. Chang from Program Manager-Combat Ammunition Systems provided recommendations to other developers to reach out early with the ESOH communities to ensure the proper ESOH data are collected, programmed for, and elucidated. He expressed the need for clarity regarding these data requirements; to include what specifically is needed, when it is needed, how long will the collection take and how much will it cost to be provided in a clear and concise manner. He also expressed the need for a clear understanding and guidance regarding what entity is responsible for the delineation of specific ESOH issues and the process to resolve them. Examples included occupational medical surveillance, industrial hygiene requirements, data collection, and how data are interpreted.

Principals provided results and interpretation of the issues addressed through the Strategic Environmental Research Program’s (SERDP) Statement of Need ER 12-02 regarding the need for fate, transport, and effects data for IM constituents, particularly for IMX formulations. The data and results presented in this workshop addressed many questions presented previously regarding the potential for environmental loads, environmental fate, and toxicity. IM formulations tend to be safer than their counterparts in regards to handling, storage, and use. However, the probability for greater environmental deposition from use associated with high and low order detonation, partial detonation, and unexploded ordnance appears high. The panel suggested that given the insensitive nature of the formulation, new methods are needed for unexploded ordnance disposal that diverge from blow-in-place methods that are largely ineffective in the removal of constituents.
The high water solubilities and low affinities to organic carbon suggest environmental transport to groundwater for two constituents, NTO and NQ is likely. Recent data suggests that the mixture consisting of the formulation does not impede this process of dissolution. Data suggested the possibility for dimer and trimer formulation of soil DNAN residues that may result in enhanced toxicities to bacteria in laboratory tests, though further work is needed to confirm the presence of these constituents in the environment and the relevance of the bacterial toxicity to higher level organisms and soil microbial communities.

Earlier work reported that photolysis of NQ may result in more toxic nitroso breakdown products in water, that have not yet been quantified; however, the environmental half-lives for these intermediates may be short lived. Aquatic toxicity of NTO has been found to be largely due to the acidic nature of the compound in water. The aquatic toxicity of DNAN appears to be lower than that associated with TNT and other compounds of that class (nitroaromatics). Overall, aquatic toxicity associated with IMX constituents are lower than found for constituents of conventional formulations. Issues exist in the area of wastewater treatment, where the yellow coloration characteristic of DNAN-based formulations is persistent. Research is underway to help address these concerns.

Mammalian toxicity of DNAN appears to be slightly lower than that of TNT which is consistent with the aquatic data. Toxicity of NTO and NQ are low; however, there are no data or active plans to conduct chronic testing. Data are needed to reduce the uncertainty associated with the development of drinking water criteria, where the need is eminent. The target of toxicity for DNAN is the blood (consistent with other nitroaromatic compounds). The primary target for NTO exposure appears to be sperm production in mammalian models. Given the non-toxic nature of NQ in mammalian bioassays, no targets of toxicity of mammalian models for NQ exposure have been identified.

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