

# Physiological Analysis of Human Dorsal Root Ganglia Provides a Promising Approach to Accelerating Translational Pain Research

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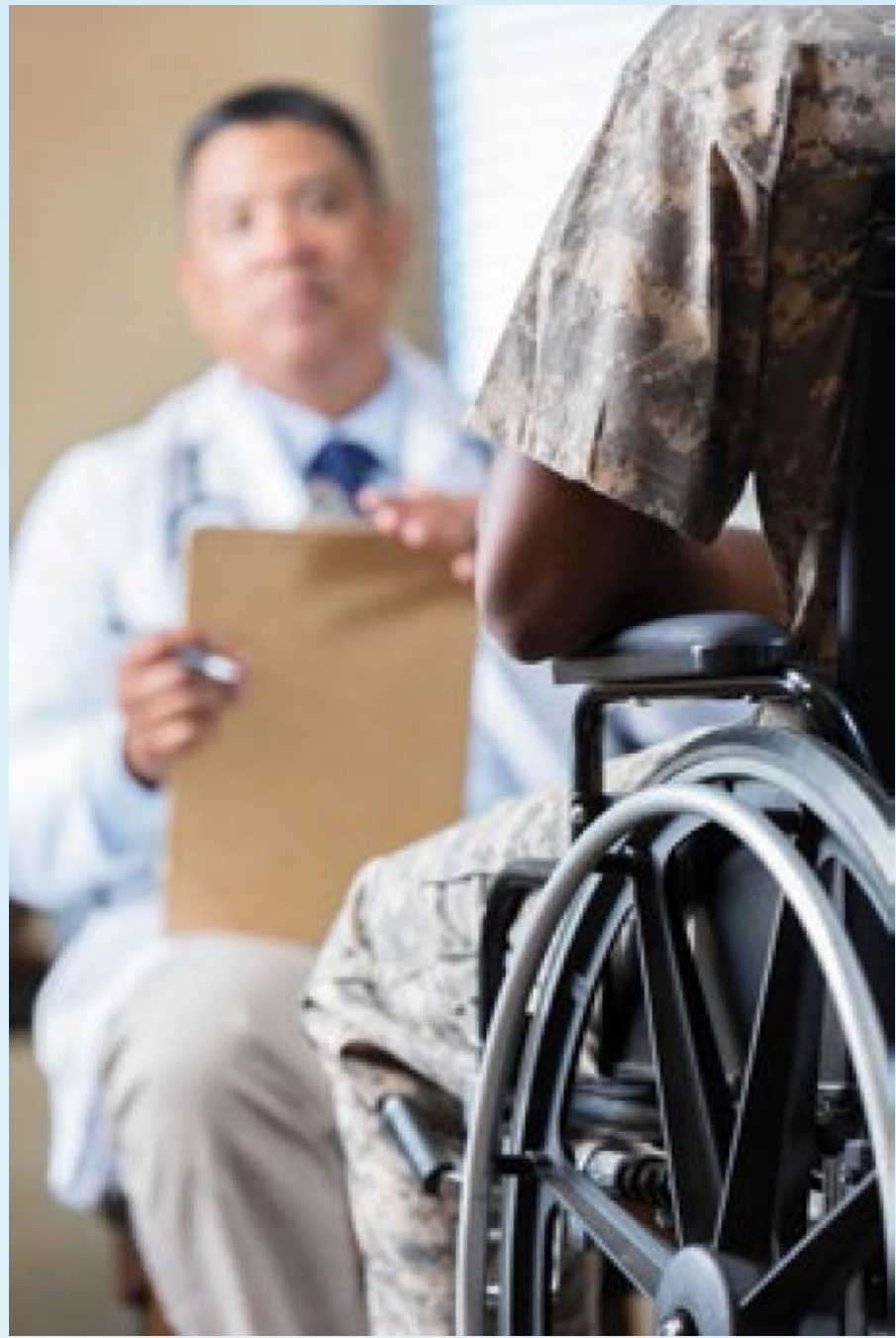
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## Background

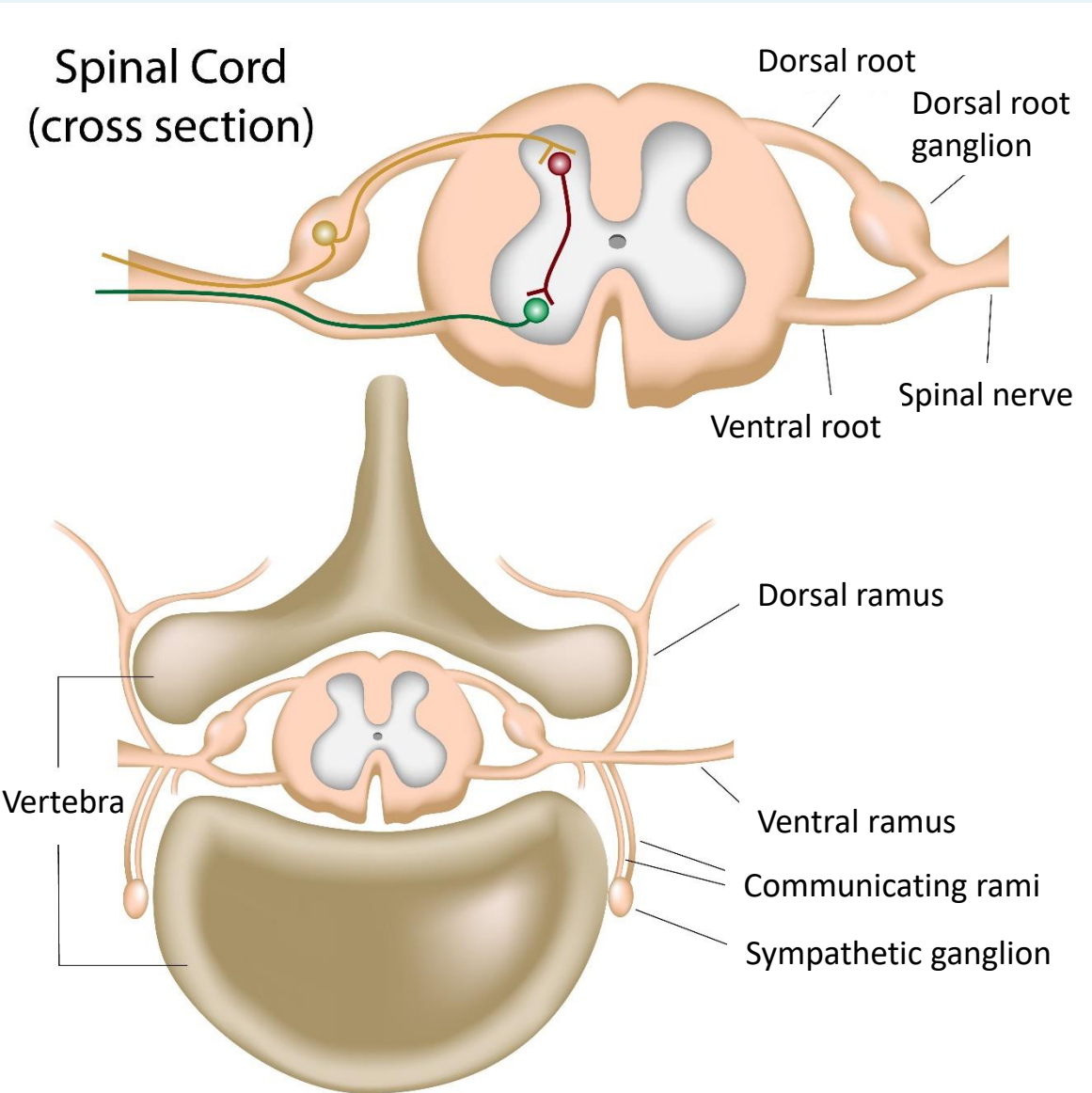
**Advancing Trauma and Combat Casualty Research:** Soldiers routinely endure injuries in both combat and training exercises that require effective pain management. Opioid-based therapies have been the primary pain treatment for over 100 years. Although opioid-based therapies mitigate acute pain, they can have side-effects, including potential for addiction, that make their chronic use challenging. More effective experimental approaches and model systems are needed to advance the field.



### Key Objectives:

1. Identify challenges with animal versus human DRG models
2. Demonstrate key physiological differences between human and rat DRG model systems
3. Recommend an experimental approach suitable to develop and validate new pain therapies for wounded soldiers and civilians

## DRGs Are a Model System for Pain Research



- ✓ **Experimental Model**  
Primary cultures of DRG sensory neurons
- ✓ **Advantages**  
DRG neurons maintain functionality in vitro – respond to stimuli and display sensitization
- ✓ **Utility**  
Cellular and molecular mechanisms of sensory neuron function and drug screening assays

**DRG Basics:** DRGs contain the cell bodies of sensory neurons that transmit the sensory messages of pain and touch from the periphery to the CNS. This critical function makes them the target of pain research. Primary cultures of DRG neurons maintain functionality in vitro allowing for the evaluation of response to thermal, mechanical, and chemical stimuli, and for testing new drugs.

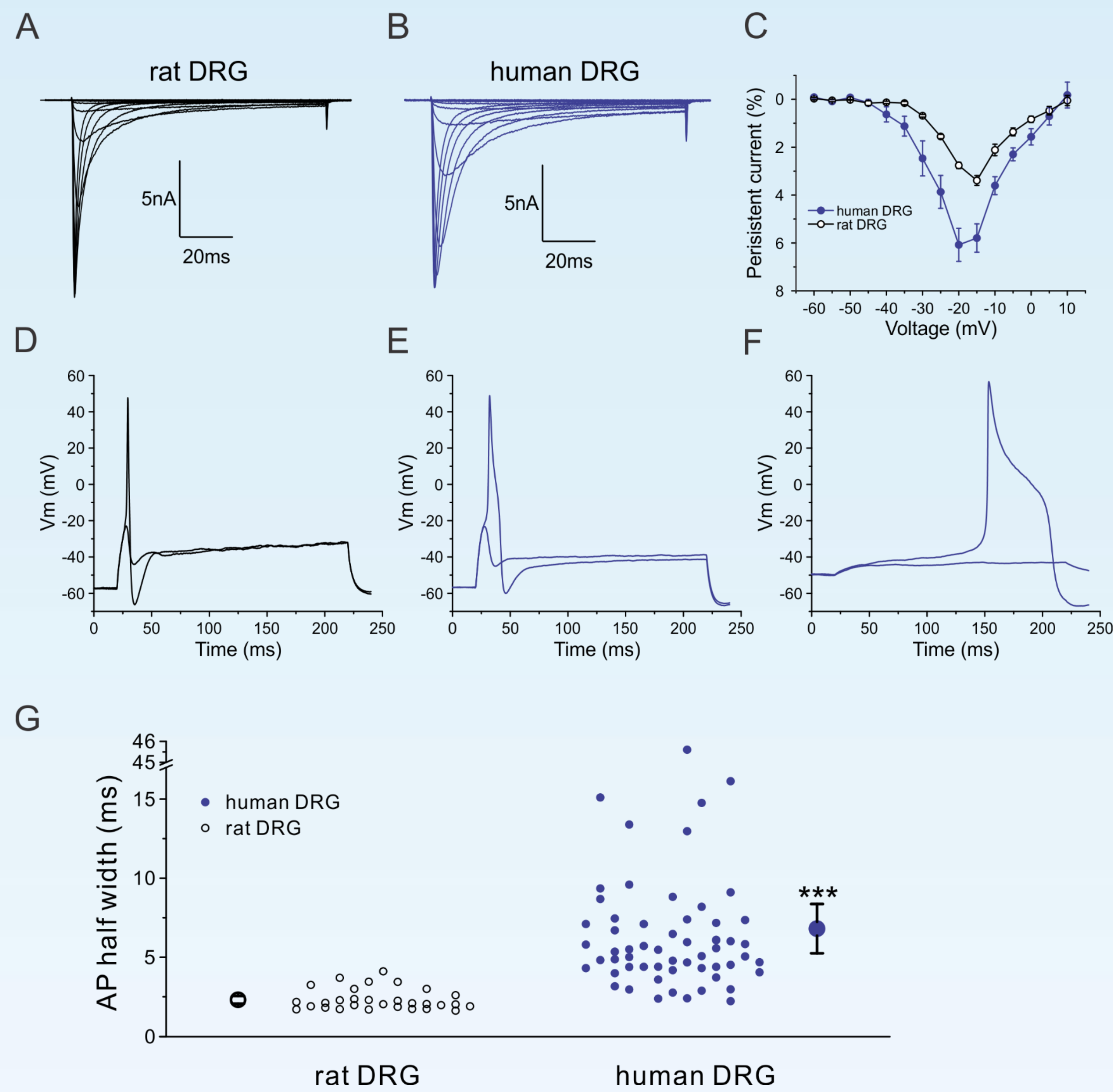
## Challenges Associated with DRG Model System



**Evaluation of Experimental Models for Pain Research:** The evaluation of effective pain therapies requires validation in human cells/tissues. While animal models provide an easily accessible model system for investigators, species-specific functional properties of animal versus human DRGs can negatively impact advances in pain management.

## What Are Key Differences Between Rat and Human DRG Properties In Vitro?

### Human DRGs Have Distinct Electrophysiological Properties



**Human DRG neurons demonstrate larger Nav1.8 persistent currents, and generate broader action potentials than rat DRG neurons.** **A&B.** Representative Nav1.8 current traces from a rat (A) or a human (B) DRG neuron. Cells were held at -60 mV and stepped to a range of potentials for 100 ms. **C.** An IV plot showing rat and human Nav1.8 persistent currents. **D&E.** Representative action potential traces from rat (D) or human (E) DRG neurons. The RMP for human DRG neurons was  $-55.0 \pm 0.8$  mV ( $n=59$ ), which was depolarized by 6 mV, compared with the RMP of rat DRG neurons ( $-61.0 \pm 1.9$  mV,  $n=30$ ,  $p < 0.01$ ). **F.** Action potential traces recorded from a human DRG neuron with prolonged duration. **G.** Comparison of the half-width of action potential between rat and human DRG neurons. Two larger symbols indicate means  $\pm$  SEM. For human DRG neurons, the mean half-width of the action potential was  $6.83 \pm 0.78$  ms ( $n=59$ ), which is 3-fold larger than that of rat DRG neurons where it was  $2.33 \pm 0.12$  ms ( $n=30$ ,  $*** p < 0.001$ ). Data from Han C et al *J Neurophysiol* 2015;113: 3172-3185.

### Methods

- **DRG Recovery**
  - Human – Organ donors within 6 hrs of cross-clamp
  - Rat – Dawley rats (2-8 wks of age)
- **Primary Cell Culture**  
DRG neurons isolated and plated at low density on glass coverslips
- **Electrophysiology**  
Within 24 hours of plating, cells recorded using standard voltage- and current-clamp protocols

## Advancing Pain Research Related to Wounded Soldiers and Civilians Requires Human Biospecimens

### Summary of Species-Specific Differences

- **Nav1.8 Channel – compared to rat, human:**
  - produce ~2x larger persistent currents
  - have slower kinetics of inactivation
- **DRG Neuron Properties – compared to rat, human:**
  - have more depolarized resting potential
  - have lower input resistance/higher threshold for action potential
  - have wider action potential

**Conclusion:** The properties of human DRG neurons, compared to rodent, show clear species-specific differences, which should be taken into account when extrapolating from rodent studies of pain to humans and testing novel blockers for treatment of pain.

## NDRI Provides Superior Access to Human DRGs

- **Low Post Mortem to Preservation Intervals (PMI)**  
< 6 hours
- **Recovery Customized to Researcher Need**
  - Researcher Specified Quantities and Locations
  - Samples Preserved Fresh in Media, Frozen, or Fixed
- **Experience Serving the Research Community**
  - >300 DRGs provided to 23 investigators in the last 3 years



**NDRI's DRG Program:** NDRI implemented the DRG project in 2011 to recover high quality, viable DRGs with a low-PMI to provide researchers with tissue suitable for rigorous experimental analysis, including cell culture and electrophysiological experiments.

## Other Highlighted DRG Publications

- Chen G et al *Nature Neuroscience* 2017; 20(7):917-926. PMID: 28530662.
- Han Q et al *Neuron* 2016; 92(6):1279-1293. PMCID: PMC5182147.
- Xu ZZ et al *Nature Medicine* 2015; 21(11):1326-31. PMCID: PMC4752254.

## NDRI: A Valued Partner for Military Medical Research

- ✓ Extensive nationwide network of recovery partners
- ✓ Standardized collection and preservation of biospecimens according to project-driven requirements
- ✓ Scientific and technical expertise and guidance on best practices