

**Painful Procedures Correlate With Markers of Hypoxia, Oxidative Stress and Intestinal Injury in Premature Neonates**

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**Purpose:**

We previously published that common NICU tissue-damaging procedures (TDPs) such as tape removal or heel lance increase plasma markers of hypoxia and oxidative stress [1]. To determine if these changes are detectable in urine, we measured the concentration of these markers from urine samples prospectively collected on day of life (DOL) 1-4 from 62 premature neonates. Sample size was 8 for DOL1, 37 for DOL 2, 52 from DOL 3 and 17 from DOL4. Sample sizes were variable due to reduced urine output on DOL 1 and presence of stool on many of the samples from DOL 4. The frequency of TDPs were collected from the medical record

**Methods:**

Uric acid (urine marker of hypoxia) was measured using high performance liquid chromatography as we previously described [2]. Allantoin (urine marker of oxidative stress) was measured using gas chromatography mass spectrometry [2]. Intestinal fatty acid binding protein (early marker of intestinal injury) was measured using enzyme linked immunosorbent assay [3]. The number of TDPs was obtained from the medical records. Data was analyzed using Spearman's rho (SPSS version 24), and a P value of less 0.05 is considered statistically significant.

**Results:**

On DOL1-4, we found significant positive correlations between the number of common tissue-damaging procedures and uric acid, which suggest that an increase in TDP can increase ATP breakdown or decrease ATP synthesis. We also found significant positive correlations between the number of common tissue-damaging procedures and allantoin, an in vivo free radical marker. This data suggests a relationship between TDPs and oxidative stress, as reactive oxygen species are produced during ATP breakdown [4]. More importantly, we found a significant positive relationship between allantoin and IFABP, suggesting a possible link between oxidative stress and early intestinal injury.

**Conclusion:**

Significant positive correlations were detected between the number of tissue-damaging procedures and markers of hypoxia, oxidative stress and early intestinal injury. These data suggest that NICU nursing care delivery must be modified to prevent or reduce the number of times premature neonates are exposed to painful and tissue-damaging procedures. Future studies are required to determine if markers of oxidative stress and early intestinal injury are associated with the incidence of necrotizing enterocolitis.

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**Keywords:**

Biochemical markers, NICU and Pain

## References:

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## Abstract Summary:

The correlation between common tissue-damaging procedures and urine markers of hypoxia (uric acid), oxidative stress (allantoin) and early intestinal injury (intestinal fatty acid binding protein) in premature neonates will be discussed.

## Content Outline:

**INTRODUCTION:** We previously published that common NICU tissue-damaging procedures (TDPs) such as tape removal or heel lance increase plasma markers of hypoxia and oxidative stress [1]. To determine if these changes are detectable in urine, we measured the concentration of these markers from urine samples prospectively collected on day of life (DOL) 1-4 from 62 premature neonates. Sample size was 8 for DOL1, 37 for DOL 2, 52 from DOL 3 and 17 from DOL4. Sample sizes were variable due to reduced urine output on DOL 1 and presence of stool on many of the samples from DOL 4. The frequency of TDPs were collected from the medical record.

**METHOD:** Uric acid (urine marker of hypoxia) was measured using high performance liquid chromatography as we previously described [2]. Allantoin (urine marker of oxidative stress) was measured using gas chromatography mass spectrometry [2]. Intestinal fatty acid binding protein (early marker of intestinal injury) was measured using enzyme linked immunosorbent assay [3]. The number of TDPs was obtained from the medical records. Data was analyzed using Spearman's rho (SPSS version 24), and a P value of less 0.05 is considered statistically significant.

**RESULTS:** On DOL1-4, we found significant positive correlations between the number of common tissue-damaging procedures and uric acid, which suggest that an increase in TDP can increase ATP breakdown or decrease ATP synthesis. We also found significant positive correlations between the number of common tissue-damaging procedures and allantoin, an in vivo free radical marker. This data suggests a relationship between TDPs and oxidative stress, as reactive oxygen species are produced during ATP breakdown [4]. More importantly, we found a significant positive relationship between allantoin and IFABP, suggesting a possible link between oxidative stress and early intestinal injury.

**CONCLUSION:** Significant positive correlations were detected between the number of tissue-damaging procedures and markers of hypoxia, oxidative stress and early intestinal injury. These data suggest that NICU nursing care delivery must be modified to prevent or reduce the number of times premature neonates are exposed to painful and tissue-damaging procedures. Future studies are required to determine if markers of oxidative stress and early intestinal injury are associated with the incidence of necrotizing enterocolitis.

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**Professional Experience:** I spent 20 years as an NICU nurse (3 years as the clinical director, 5 years as the nurse manager, 7 years educator, 5 years transport manager and bedside nurse). I am currently an NIH-NINR funded researcher examining the relationship between procedural pain and biochemical markers of hypoxia (hypoxanthine, xanthine, uric acid) and oxidative stress (malondialdehyde and allantoin).

**Author Summary:** I am currently a Professor of Physiology at Loma Linda University School of Medicine with an NIH-funded grant examining the mechanisms that link procedural pain to cellular injury.