Preliminary evidence that hydroxyurea is associated with attenuated peripheral sensitization in adults with sickle cell disease

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Abstract

Introduction: Hydroxyurea (HU) is a drug that targets the underlying pathophysiology of sickle cell disease (SCD); however, it continues to be an underutilized treatment for adults. Previous research suggests that HU treatment can result in fewer hospital contacts for acute vaso-occlusive pain crises (VOC). Hydroxyurea’s impact on non-VOC pain, however, is not well established.

Objectives: This study examined whether HU moderated patterns of static and dynamic pain processing and clinical pain in SCD individuals.

Methods: Fifty-eight patients with SCD (N taking HU = 17) underwent quantitative sensory testing (QST) and completed twice daily symptom diaries for 12 weeks. Quantitative sensory testing established thermal threshold and tolerance, mechanical thresholds, and thermal and mechanical temporal summation of pain.

Results: Groups did not differ in age, sex, or opioid use. After controlling for morphine use, QST results showed that participants taking HU had higher heat and mechanical pain thresholds (static QST measures) but not thermal and mechanical temporal summation (dynamic QST measures). Participants taking HU also reported lower VOC pain compared with SCD participants not taking HU; however, HU did not moderate non-VOC clinical pain ratings.

Conclusion: Findings cautiously suggest that HU acts on pain hypersensitivity and VOC pain, rather than inhibiting pain facilitation and non-VOC pain. These differences may reflect HU’s influence on peripheral rather than central sensitization. Future research is warranted to replicate these findings in a larger sample and determine whether early HU administration can prevent peripheral sensitization in SCD individuals.

Keywords: Sickle cell disease, Hydroxyurea, Quantitative sensory testing, VOC pain, Chronic pain

1. Introduction

Sickle cell disease (SCD) is a collection of congenital hemoglobinopathies that causes erythrocyte malformations. These “sickled” cells promote hemolysis, vaso-occlusion, and tissue hypoxia, leading to a myriad of health complications. Episodes of severe, acute pain, classically attributed to vaso-occlusive crises (VOC), are a hallmark of the disease. Up to 65% of adult patients also experience chronic pain. Whereas VOC pain is acute, episodic, and associated with ischemic events, the etiology of noncrisis chronic pain (ie, non-VOC pain) is complex and poorly understood.

Central sensitization (CS), or plasticity of neurons in response to inflammation or injury, is one possible cause of non-VOC pain. Contemporary theory suggests peripheral neuron hyperexcitability—likely triggered by VOCs, organ complications, and opioid use—might have compounding effects on central pain modulation systems, contributing to the amplification and facilitation of nociception. In patients with SCD, previous work suggests that CS is associated with lower fetal hemoglobin levels, which is a modulator of SCD severity. Treatments targeting fetal hemoglobin might reduce CS and non-VOC pain, however, no study has previously examined such effects.

Hydroxyurea (HU), an FDA-approved treatment, decreases the proportion of sickled cells by increasing fetal hemoglobin, which reduces hemoglobin polymerization and inhibits noxious...
2. Methods

This secondary analysis uses data from a case-control protocol examining pain processing and daily function in SCD adults. Previous reported sample sizes vary based on data collection stage and aim. This study uniquely focuses on HU’s association with evoked and clinical pain to address the knowledge gap about HU and non-VOC pain. Based on data collection stage and aim. This study uniquely focuses on HU’s association with evoked and clinical pain to address the knowledge gap about HU and non-VOC pain.9

2.1. Data collection procedures

Inclusion and exclusion criteria are listed in our previous work. Phone-screened eligible individuals attended an in-person visit on a day of typical pain (but <5/10 intensity) with no VOCs in the previous 3 weeks. Consented participants completed questionnaires, QST, and daily electronic diaries for 12 weeks.

2.2. Measures

2.2.1. Quantitative sensory testing

Detailed QST procedures were previously described (see Refs. 12 and 13). Briefly, we assessed pain threshold and tolerance (ie, static QST measures) as well as temporal summation of pain (ie, dynamic QST measures). Heat pain threshold and tolerance stimuli were delivered using a Peltier element-based stimulator (Medoc, Israel; Pathway, Advanced Thermal Stimulator thermode) applied on the ventral forearm with an ascending method of limits paradigm that had a 0.5/sec rise rate. Mechanical pain threshold stimuli were delivered twice each using an algometer (SBMedic) with a 1-cm² hard rubber probe applied bilaterally on the trapezius muscle, interphalangeal joint of the thumb, the proximal third of the forearm, and the middle of the quadriceps. Heat pain threshold and tolerance (ie, static QST measures) as well as temporal summation of pain (ie, dynamic QST measures) were included.

2.2.2. Daily electronic symptom monitoring

Participants completed daily diaries for 12 weeks after the visit, providing daily average pain intensity ratings on a 0 to 100 scale. Pain ratings were categorized as “VOC” or “non-VOC” based on separately averaged ratings for crisis and noncrisis days.

3. Results

3.1. Participants

Of the 58 participants, 17 endorsed regular HU use (SS genotype = 16). Table 1 reports demographic characteristics. All individuals were African American/black, and one individual identified as Hispanic (HU group). Chi-squared tests showed no group differences in sex (P = 0.984), ethnicity (P = 0.519), and education (P = 0.241). There were no differences in age (P = 0.14) or daily morphine equivalents (P = 0.431). HU+ participants had significantly lower white blood cell counts than HU− participants (t15.0 = −3.1, P = 0.004), demonstrating HU’s therapeutic efficacy.

3.2. Quantitative sensory testing

Figure 1 shows results from analyses of covariance comparing groups on measures of static (Fig. 1A, B) and dynamic (Fig. 1C) pain processing. Total daily morphine equivalents were positively associated with mechanical pain threshold in the thumb (P = 0.03), quadriceps (P = 0.02), and forearm (P = 0.08, trend) but not heat pain threshold/tolerance (P = 0.98, 0.63, respectively), trapezius pain threshold (P = 0.23), or temporal summation (Ps > 0.12). On static QST measures, HU+ individuals demonstrated significantly higher thermal pain threshold (F1,62 = 9.2, P = 0.004, n2 = 0.15) but not tolerance (F1,61 = 1.3, P = 0.25, n2 = 0.03) compared with HU− participants. HU+ individuals also had significantly higher mechanical pain thresholds in the trapezius (F1,62 = 14.3, P < 0.001, n2 = 0.22), forearm (F1,62 = 9.7, P = 0.003, n2 = 0.16), thumb (F1,61 = 7.0, P = 0.01, n2 = 0.12), and quadriceps (F1,62 = 9.1, P = 0.004, n2 = 0.15). However, there were no group differences in thermal or mechanical temporal summation (thermal: F1,62 = 0.75, P = 0.4, n2 = 0.02; mechanical: F1,48 = 0.03, P = 0.86, n2 = 0.0).
3.3. Clinical pain ratings

Forty-seven percent of HU+ and 63% of HU− participants reported non-VOC pain (Fisher’s Z = 1.7, P = 0.1). Twenty-four percent of HU+ and 46% of HU− participants reported VOC pain (Fisher’s Z = 1.6, P = 0.2). Total daily morphine equivalents were positively associated with non-VOC (P = 0.003) and VOC pain (P = 0.01). After controlling for this factor, non-VOC pain did not significantly differ between HU+ and HU− individuals (F 1,49 = 1.4, P = 0.25, h²p = 0.03), but HU− participants reported significantly lower VOC pain ratings (F 1,31 = 10.4, P = 0.003, h²p = 0.2). Table 2 provides QST and clinical pain descriptive statistics, and Figure 2 depicts differences in diary pain ratings averaged over a 12-week period.

4. Discussion

Findings from this study did not support our hypotheses. HU+ participants did not report significantly lower non-VOC pain on daily diaries, nor did they show evidence of reduced pain facilitation on dynamic QST measures. Instead, HU+ participants reported significantly lower VOC pain and had lower pain thresholds on static QST measures (ie, thermal pain and mechanical pain across the 4 testing sites) compared with HU− individuals. This effect remained after controlling for total daily morphine equivalents, suggesting that opioid use did not confound the present results.

Previous work supports our finding of reduced VOC pain ratings in SCD individuals treated with HU.2,7,10,12,21,33 Hydroxyurea’s ability to increase levels of fetal hemoglobin,15,19 change erythrocyte properties,8,26 and decrease cell adhesion results in reduction of inflammatory mediators in the periphery.23 It is possible, then, that our finding of lower static QST ratings among HU+ participants reflects treatment-induced changes in peripheral pain mechanisms.

Alternatively, we did not observe differences in non-VOC pain ratings, nor dynamic QST measures. Non-VOC pain in SCD has been described as a potential interaction of peripheral and CS mechanisms.20 Furthermore, the selected dynamic QST measures are used to probe sensitization of the central nervous system.28 Combined, these findings suggest that CS is not directly influenced through HU treatment. Future, large-scale studies yielding normative QST values for individuals with SCD will help track changes in pain processing over the course of HU treatment.

5. Limitations

Our study had limitations for future work to expand upon. First, this study is a small, secondary analysis with cross-

### Table 2

|                  | No HU, mean (SD) | Taking HU, mean (SD) |
|------------------|------------------|----------------------|
| **Thermal pain (°C)** |                  |                      |
| Threshold        | 39.9 (2.7)*      | 42.1                 |
| Tolerance        | 43.7 (1.9)       | 44.4 (1.6)           |
| **Mechanical pain (kilopascals)** | | |
| Trapezius threshold | 203.8 (70.3)*   | 308.9 (129.7)        |
| Forearm threshold  | 203.8 (70.4)*    | 291.4 (137.4)        |
| Thumb threshold   | 278.8 (93.8)*    | 339.8 (112.8)        |
| Quadriceps threshold | 443.8 (186)*   | 599.4 (227)          |
| **Temporal summation (difference score)** | | |
| Thermal          | 3.3 (7.5)        | 3.4 (7.9)            |
| Mechanical       | 15.7 (18.9)      | 11.1 (15.8)          |
| **Clinical pain (0–100 scale)** | | |
| VOC pain         | 56.5 (14.1)*     | 36.8 (19.1)          |
| Non-VOC pain     | 19.5 (18)        | 11.6 (14.5)          |

* Significant between-group difference at P < 0.05.

VOC, vaso-occlusive crises.
sectional reporting of HU use. Sickle cell disease is a rare disorder in the United States, which limits sample sizes. Second, we did not collect information about HU treatment duration or dosing over time. It is possible that long-term or early-initiated treatment impacts CS progression. Given the study’s cross-sectional nature, we cannot speak to whether early HU initiation might prevent the development of non-VOC pain. Third, we did not collect information about adherence to HU, which might confound outcomes. Finally, future research is encouraged to examine a larger cohort of individuals taking HU to address this study’s limitation of unequal sample sizes. Although these limitations support cautious interpretation of these results, findings suggest that HU more robustly impacts VOC than non-VOC pain mechanisms.

6. Conclusion

Non-VOC pain remains understudied and, critically, powerfully impacts quality of life in individuals with SCD. The present results suggest that HU is not associated with lower non-VOC pain but is robustly related to better VOC outcomes. Future research is encouraged to establish alternative treatments acting on central pain mechanisms in SCD to address non-VOC pain.

Disclosures

The authors have no conflict of interest to declare.

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References

[1] Adragna NC, Fonseca P, Lauf PK. Hydroxyurea affects cell morphology, cation transport, and red blood cell adhesion in cultured vascular endothelial cells. Blood 1994;83:553–60.
[2] Agrawal RK, Patel RK, Shah V, Nainiwal L, Trivedi B. Hydroxyurea in sickle cell disease: drug review. Indian J Hematol Blood Transfus 2014;30:91–6.
[3] Aich A, Beitz AJ, Gupta K. Mechanisms of pain in sickle cell disease, sickle cell disease—pain and common chronic complications. Baba Psalm Duniya Inusa, IntechOpen, 2016. doi: 10.5772/64647. Available at: https://www.intechopen.com/books/sickle-cell-disease-pain-and-common-chronic-complications/mechanisms-of-pain-in-sickle-cell-disease.
[4] Akhshaye I, Alsultan A, Solovieff N, Ngo D, Baldwin CT, Sebastiani P, Chui DHK, Steinberg MH. Fetal hemoglobin in sickle cell anemia. Blood 2011;118:19–27.
[5] Arendt-Nielsen L, Yamtsky D. Experimental and clinical applications of quantitative sensory testing applied to skin, muscles and viscera. J Pain 2009;10:566–72.
[6] Armstrong RA. When to use the Bonferroni correction. Ophthalmic Physiol Opt 2014;34:502–8.
[7] Badawy SM, Thompson AA, Penedo FJ, Lai JS, Rychnik K, Liem RJ. Barriers to hydroxyurea adherence and health-related quality of life in adolescents and young adults with sickle cell disease. Eur J Haematol 2017;98:609–14.
[8] Ballas SK, Dover GJ, Charache S. Effect of hydroxyurea on the rheological properties of sickle erythrocytes in vivo. Am J Hematol 1989;32:104–11.
[9] Brandow AM, Zappia KJ, Stucky CL. Sickle cell disease: a natural model of acute and chronic pain. PAIN 2017;158(suppl):S79–84.

[10] Brawley OW, Cornelius LJ, Edwards LR, Gamble VN, Green BL, Inturrisi C, James AH, Laraque D, Mendez M, Montoya CJ, Pollock BH, Robinson L, Scholnik AP, Schori M. National Institutes of Health Consensus Development Conference statement: hydroxyurea treatment for sickle cell disease. Ann Intern Med 2008;148:302–8.

[11] Brugnara C, Bunn HF, Tosetto DC. Regulation of erythrocyte cation and water content in sickle cell anemia. Science 1986;232:388–90.

[12] Campbell CM, Carroll CP, Kiley K, Han D, Haywood CJ, Lanzkron S, Swedberg L, Edwards RR, Page GG, Haythornthwaite JA. Quantitative sensory testing and pain-evoked cytokine reactivity: comparison of patients with sickle cell disease to healthy matched controls. PAIN 2016; 157:949–56.

[13] Campbell CM, Moscou-Jackson G, Carroll CP, Kiley K, Haywood C, Lanzkron S, Hand M, Edwards RR, Haythornthwaite JA. An evaluation of central sensitization in patients with sickle cell disease. J Pain 2016;17:617–27.

[14] Carroll CP, Lanzkron S, Haywood C, Kiley K, Pejsa M, Moscou-Jackson G, Haythornthwaite JA, Campbell CM. Chronic opioid therapy and central sensitization in sickle cell disease. Am J Prev Med 2016;51:S69–77.

[15] Charache S, Terrin ML, Moore RD, Dover GJ, Barton FB, Eckert SV, McMahon RP, Bohns DR. Effect of hydroxyurea on the frequency of painful crises in sickle cell anemia. Investigators of the Multicenter Study of Hydroxyurea in Sickle Cell Anemia. N Engl J Med 1995;332:1317–22.

[16] Dampier C, Palermo TM, Darbari DS, Hassell K, Smith W, Zempsky W. AAPT diagnostic criteria for chronic sickle cell disease pain. J Pain 2017; 18:490–8.

[17] Darbari DS, Vaughan KJ, Roskom K, Seamon C, Diaw L, Quinn M. Correia A, Schechter AN, Haythornthwaite JA, Waclawiw MA, Wallen GR, Belfer I, Taylor JG. Central sensitization associated with low fetal hemoglobin levels in adults with sickle cell anemia. Scand J Pain 2017; 17:279–86.

[18] Feise RJ. Do multiple outcome measures require p-value adjustment? BMC Med Res Methodol 2002;2:8.

[19] Goldberg MA, Brugnara C, Dover GJ, Schapira L, Charache S, Bunn HF. Treatment of sickle cell anemia with hydroxyurea and erythropoietin. N Engl J Med 1990;323:396–72.

[20] Gupta K, Jahagirdar O, Gupta K. APS sickle cell disease conference mini-review: targeting pain at its source in sickle cell disease. Am J Physiol Integr Comp Physiol 2018;315:R104–R112.