Hair Color Bias
Literature Review

James A. Bourland, Ph.D., D-ABFT
Drugs bind to melanin
Melanin Content greater in Darker Hair
Incorporation and binding of drugs into hair greater in Pigmented versus Non-pigmented Hair
Higher drug concentrations demonstrated with same dose in dark hair versus light hair
Bias introduced could mean the difference between a positive or negative drug test outcome
Implies potential “Racial” Bias to Hair Testing
Hair Color Bias Literature Review:

- Description of Hair Morphology, Chemical make up, Racial/ Ethnic Differences
- Animal Studies
- Human \textit{In Vitro} Studies
- Human Studies General
- Human Controlled Dosing Studies
- Retrospective Statistical Studies
Hair Morphology
Hair Morphology

Spearman *et al.* (1960) - *AmJPhysicalAnthro*
- African American Hair
- Marked Asymmetry of pigmentation
- Concentration of Melanin Higher African A v. Asian
- No Asymmetry in Asian Hair

Lindelof *et al.* (1988) – *ArchDermatol*
- African American Hair – Helical Factor
- Asian Hair – Completely Straight
- Caucasian Hair - Variation
Hair Morphology

Hrdy et al. (1973) - Am J Physical Anthro

- Studied Hair from 7 populations
- Used 8 Factors
  - Ave Diameter
  - Medullation
  - Scale
  - Kinky
  - Ave Curvature
  - Ratio of Max to Min Curvature
  - Crimp
  - Ratio of “natural” to “straight” length

Racial Differences

African to Malaysian
Hair Morphology

Hopps et al. (1977) - SciTotalEnviron

- Diameter of Hairs range from 0.05-0.125 mm, Mean = 0.09 mm
- Sebaceous and Sweat Glands assoc with hair
- Growth Rest Cycle of Scalp Hair -1000 days
  - Anagen=Growth Phase: ~900 days,
  - Catagen=Degeneration transitional: several days
  - Telogen=Resting Phase ~100 days
Hair Color Bias

Animal Studies
Animal Studies – Hair Color Bias

Green et al. (1996) – JAT
- Incorporation of Methadone (MtD) in Rat Hair
- Male, Hooded, Lister Rats (n=36)
- Black pigmented and White non-pigmented hair
- Mean MtD ratio 21:1 in pigmented : non-pigmented hair
- Mean Melanin Content 3.5:1

Gygi et al. (1996) - Drug Metab Dispos
- Male Sprague Dawley Rats
- Distribution of Codeine and Metabolites
- Codeine IP injection daily for 5 days different doses
- Conclusion Dose dependent concentrations in hair
Animal Studies – Hair Color Bias

Gygi et al. (1996) - DrugMetabDispos
Incorporation of Codeine: Role of Pigmentation
Sprague-Dawley (SD): White non-pigmented
Dark Agouti (DA): Brown pigmented
Hooded Long Evans (LE): Black Pigmented and White Non-Pigmented
Measured Codeine, Morphine and Mor-GLuc
Codeine 40 mg/kg IP injection daily for 5 days
<table>
<thead>
<tr>
<th>Drug</th>
<th>SD</th>
<th>DA</th>
<th>P-LE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Codeine</td>
<td>0.98</td>
<td>5.99</td>
<td>111.93</td>
</tr>
<tr>
<td>Morphine</td>
<td>0.34</td>
<td>0.51</td>
<td>14.46</td>
</tr>
<tr>
<td>MOR-gluc</td>
<td>0.67</td>
<td>1.04</td>
<td>13.80</td>
</tr>
</tbody>
</table>

*Gygi et al. (1996) - Drug Metabolism and Disposition*
Wilkins et al. (1998)-*JPharmSci*

Incorporation of Drug in Pigmented v. Non-Pigmented Hair

L-\(\alpha\)-Acetylmethadol, Buprenorphine or Methadone

Male Long Evans (LE): Black Pigmented and White Non-Pigmented Hair

Conclusion: Concentration of each drug at either high or low dose was considerably less in non-pigmented v. pigmented hair
Borges et al. (2001)-JAnalToxicol

Incorporation of Amphetamine and N-Acetylamphetamine in Hair
Role of Basicity in Hair Color Bias
Male Long Evans (LE): Black Pigmented and White Non-Pigmented Hair
Rats dosed with Amphetamine >3X more amphetamine in black versus white hair pigmented hair
No significant difference in concentrations of N-Acetylamphetamine among black or white hair
Borges et al. (2001)-JAnalToxicol
Incorporation of Amphetamine and N-Acetylamphetamine in Black and White LE Rat Hair

Figure 4. Concentrations of amphetamine found in white and black rat hair (A) and N-AcAP found in white and black rat hair (B). Error bars represent standard deviation. n = 8 for each drug. *Significant at p < 0.001.
Hair Color Bias

Human *In Vitro* Studies
Reid et al. (1994)-JToxicolClinToxicol

*In vitro* differential binding of BE to pigmented human hair samples

*In vitro* Incorporation of BE to Black, Brown and Blonde hair types

Incorporation of BE Black>Brown>Blond

**TABLE 1**

<table>
<thead>
<tr>
<th>Test No.</th>
<th>Black Hair</th>
<th>Brown Hair</th>
<th>Blond Hair</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1,938,775</td>
<td>483,552</td>
<td>319,905</td>
</tr>
<tr>
<td>2</td>
<td>1,790,565</td>
<td>517,746</td>
<td>298,899</td>
</tr>
<tr>
<td>3</td>
<td>951,620</td>
<td>665,557</td>
<td>299,899</td>
</tr>
<tr>
<td>Mean (X)</td>
<td>1,560,319</td>
<td>555,692</td>
<td>306,234</td>
</tr>
<tr>
<td>SD</td>
<td>532,332</td>
<td>96,649</td>
<td>11,849</td>
</tr>
</tbody>
</table>

ANOVA: F = 13.543; P = 0.0006
Joseph et al. (1994)-TIAFT Meeting

*In vitro* binding of $[^3\text{H}](\text{-})$ Cocaine to human hair samples

Subjects

- “Light Colored” Female Caucasian Hair (n=8)
- Female African American Hair (n=9)

Hair homogenized - 100uM $[^3\text{H}](\text{-})$ Cocaine

Amt binding defined amount of $[^3\text{H}](\text{-})$ Cocaine displaced by 10 uM of (\text{-}) Cocaine

Radioactivity measured

- 592 ±488 Female Caucasian Hair
- 11,080 ± 8,225 Female African American Hair
Joseph et al. –JPET:1997

In Vitro Characteristics of Cocaine Binding Sites in Human Hair

$^3$H-Cocaine

Radio Ligand Binding:
- Male Africoid > Female Africoid
- Africoid > Caucasoid

Melanin most likely Binding Site

5-43 fold greater binding capacity in Dark Hair versus Light Hair:
Hair Color Bias

Human Studies General
15 patients

Medical treatment with:
- Carbamazepine, Amitriptyline, Chlorprothixene, Diclofenac, Doxepine, Indomethacin, Maprotiline, Metaclopramide

Chronic Heroin and Cocaine Abuse

Hair separated into pigmented and white non-pigmented fractions
Rothe et al. (1997) – ForensicScilnt

Conclusions:

(1) Pigmentation strongly affects the drug concentration in hair. Therefore the natural colour or the melanin content of the sample should be registered and taken into account, if forensic or clinical conclusions are drawn from the data.

(2) Drugs can also be determined in weakly or non-pigmented hair. White- haired and very fair-haired people are not excluded from hair analysis.
Hair Color Bias

Human Controlled Dosing Studies
Henderson et al. (1998) – JAnalToxicol
Incorporation of Isotopically Labeled Cocaine into Human Hair: Race as a Factor

- Cocaine-D5
- *In Vivo* Study
  - 2 mg/kg COC-d5
  - Intranasal Administration
- Subjects
  - 8 Non Caucasians
  - 6 Caucasians from previous study
- 2.7 X more COC-d5 in Non-Caucasian Group
- Not Conclusive - Small Sample Size
Kronstrand et al. (1999) - ClinChem

- Codeine Controlled Dose Study
- 9 Subjects – Single Oral Dose
- Plasma samples collected -24 hours
- Hair samples – Weekly – one month
- Total Melanin, Eumelanin and Codeine Assayed
- Codeine vs. Total Melanin ($r^2=0.86$)
- Codeine vs. Eumelanin ($r^2=0.90$)
- Suggests Normalizing for Melanin Content
Study Group

- Caucasian: black hair (n=6), brown (n=12), blond (n=8), red (n=6)
- Non-Caucasian: black hair (n=12)

Oral Codeine Syrup – 30 mg, 3X/day -5 days

Plasma samples collected -24 hours

Hair samples – Week 4, 5, 6, & 7

Codeine vs. Total Melanin (r²=0.73)
## Rollins *et al.* (2003) – JAnalToxicol

**RESULTS**

<table>
<thead>
<tr>
<th>Hair Type</th>
<th>[Codeine ] (pg/mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Black (n=18)</td>
<td>1429 ± 249</td>
</tr>
<tr>
<td>Brown (n=12)</td>
<td>208 ± 17</td>
</tr>
<tr>
<td>Blond (n=8)</td>
<td>99 ± 10</td>
</tr>
<tr>
<td>Red (n=6)</td>
<td>69 ± 11</td>
</tr>
<tr>
<td>Asian Black (n=12)</td>
<td>2564 ± 170</td>
</tr>
<tr>
<td>Caucasian Black (n=6)</td>
<td>865 ± 162</td>
</tr>
</tbody>
</table>
Hair Color Bias

Retrospective Statistical Studies
<table>
<thead>
<tr>
<th></th>
<th>Cocaine Mean Concentration</th>
<th>Standard Deviation</th>
<th>#of Cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>All Arrestees</td>
<td>15.2</td>
<td>35.1</td>
<td>382</td>
</tr>
<tr>
<td>Black Arrestees</td>
<td>24.9</td>
<td>52.7</td>
<td>116</td>
</tr>
<tr>
<td>White Arrestees</td>
<td>11.1</td>
<td>22.9</td>
<td>258</td>
</tr>
</tbody>
</table>
## Mieczkowski-1995
### Ratios of Cocaine(+) Urinalyses, Hair Assays, & Self-Reports

<table>
<thead>
<tr>
<th></th>
<th>Black Arreestees</th>
<th>White Arreestees</th>
<th>Ratio: Black/White</th>
</tr>
</thead>
<tbody>
<tr>
<td>%Urine(+)</td>
<td>36.7</td>
<td>16.04</td>
<td>2.29</td>
</tr>
<tr>
<td>%(+)S/R 48 Hrs.</td>
<td>13.3</td>
<td>6.3</td>
<td>2.11</td>
</tr>
<tr>
<td>%Hair(+)</td>
<td>63.4</td>
<td>32.5</td>
<td>1.95</td>
</tr>
<tr>
<td>% (+)S/R 60 Days</td>
<td>17</td>
<td>10.98</td>
<td>1.56</td>
</tr>
</tbody>
</table>
Kidwell et al. (2000) — *Forensic Scilnt*
Evidence for Bias in Hair Testing...

“Cultural Bias” v. “Hair Color Bias”

<< Cocaine in black hair of Asian-Caucasian males than African American females

Cosmetic Treatments, Hygiene

Cut-off could determine whether bias is observed

Heavy Use population - little no bias

Light Use population – bias more likely
### Hoffman- 1999: Analysis of Race Effect on Drug Test Results

<table>
<thead>
<tr>
<th>Sex/Result</th>
<th>Urine</th>
<th></th>
<th>Hair</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Black</td>
<td>White</td>
<td>OR</td>
<td>Black</td>
</tr>
<tr>
<td>Males</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Positive</td>
<td>11</td>
<td>5</td>
<td>3.99</td>
<td>41</td>
</tr>
<tr>
<td>Negative</td>
<td>473</td>
<td>857</td>
<td>3.99</td>
<td>443</td>
</tr>
<tr>
<td>Females</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Positive</td>
<td>1</td>
<td>1</td>
<td>3.99</td>
<td>10</td>
</tr>
<tr>
<td>Negative</td>
<td>278</td>
<td>226</td>
<td>0.81</td>
<td>269</td>
</tr>
</tbody>
</table>
Kelly et. al. (2000)- ForensicSciInt

Hair Analysis for drugs of abuse. Hair color and race differentials or systematic differences in drug preferences?

- Large Retrospective Population
- 3 Groups (2000 each) (n=6,000)
- Hair color: 7 Categories
- Only COC, MAMP/AMP, THCA Positives and No Drug Detected
- SP1: Urine Drug Testing (UDT) Results v. Hair Color (Recorded at Collection Site)
- SP2: UDT Results v. Ethnicity Determination via Photographs and Surnames
- SP3: Hair Drug Testing Results v. Hair Color Determination
Cocaine Positives: Pattern in Urine and Hair

Kelly et al. (2000)- ForensicSciInt
Amphetamine Positives: Pattern in Urine and Hair

Kelly et. al. (2000) - ForensicSciInt
Our summary impression is that the observed outcome patterns were largely consistent with differences in drug preferences among the various societal groups. There was little evidence of a pattern attributable to hair color bias alone or selective binding of drugs to hair of a particular color. Likewise, there was no discernible pattern associated with race or ethnicity that would lend support to a "race effect" in drug analysis.
HAIR COLOR BIAS OBSERVATIONS

Drugs /metabolites: bind to melanin
  - Authors suggest normalizing
  - Others suggest Lab Procedures to compensate

Equal doses of many basic drugs are detected in significantly greater concentrations in pigmented than non-pigmented hair in animals and humans.

Differences in binding and ultimate concentrations based on:
  - Hair Color
  - Hair Type
Hair Color Bias Results may be dependent on
  - Cut-off Concentrations
  - Type of User: Recreational –Heavy

Statistical Findings: Bias in Urine similar to Bias in Hair among different ethnic groups.

Retrospective Studies Hair Color Bias Model:
  - Cocaine –Fits Model
  - Amphetamine- Contrary to Model
  - THC-COOH-Not Supportive of Model