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23 May 2012

http://hdl.handle.net/2440/58234
Title
Digital image analysis of fingernail colour in cadavers comparing carbon monoxide poisoning to controls.

Short title
Carbon monoxide and fingernail colour

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Abstract
Carbon monoxide is present in motor vehicle exhaust fumes, provided a functional catalytic converter has not been fitted. This gas binds avidly to the haemoglobin molecule in red blood cells and prevents its oxygen transport function, effectively poisoning the body by starving it of oxygen. In binding to haemoglobin, carbon monoxide forms carboxyhaemoglobin, which has a characteristic bright pink colour. It has been remarked that the fingernails of victims of carbon monoxide tend to exhibit pink colour, otherwise fingernails of deceased bodies tend towards a dark red to blue colour. This study sought to objectively determine by using digital image analysis if there was a colour difference in the fingernails of a group of cadavers with carbon monoxide poisoning compared to a group of controls.

Key words:
Carboxyhaemoglobin, Fingernail, Computer-assisted image analysis
Introduction

Carbon monoxide is a colourless, odourless gaseous product of combustion that is toxic even at low concentrations (1, 2). It has a higher affinity for haemoglobin than oxygen, which it displaces thus rendering red blood cells unable to transport oxygen from the lungs to the tissues (3, 4). Toxicity may also be exerted by direct effects on enzymes of respiration (3, 4) and possibly via interruption of postulated potential neurotransmitter roles of carbon monoxide (5).

The commonest source for carbon monoxide poisoning is exhaust fumes of motor vehicles, providing a method for committing suicide. Although modern petrol vehicles are fitted with catalytic converters, which convert carbon monoxide to carbon dioxide (amongst other processes) (6), victims are still able to access older vehicles that do not have converters. In the absence of a functioning catalytic converter motor-vehicle exhaust fumes may be rapidly fatal if permitted to accumulate in a confined space (7).

After death blood becomes de-oxygenated (due to residual anaerobic metabolism of cells after cessation of the circulation). Thus, normal post-mortem hypostasis has a bluish to bluish-red hue due to the presence of deoxyhaemoglobin. However, the colour may become pink in cold situations due to resaturation of haemoglobin, resulting from a shift in the oxygen binding affinity occurring at low temperatures (8). Binding of carbon monoxide to haemoglobin not only displaces oxygen, but also causes a colour change. Cherry pink hypostasis characteristically develops, but not in all cases (9).

Through the fingernails of the hands it is possible to view the capillaries of the nail bed. It would be expected that these are shielded from passive re-oxygenation by the fingernail. However, if carboxyhaemoglobin is present to a significant degree it would be anticipated that the fingernail beds would exhibit a pink colour.

Determination of carboxyhaemoglobin levels can be performed using examination of absorption of light using a range of wavelengths (10), but complex mathematical modelling is required to perform reflective spectrophotometric measurement of carboxyhaemoglobin levels from skin (11). This study sought to determine if it would be possible to predict the presence of significant levels of carboxyhaemoglobin post-mortem by simple observation of the fingernails of cadavers. This was performed by obtaining a series of photographs of fingernails from cases of carbon monoxide poisoning and from controls. The photographs were subjected to digital image analysis to obtain the red and blue intensities from the digital RGB (Red Green Blue) values from the fingernails.

Methods

Cases of carbon monoxide poisoning were acquired from the Westmead Department of Forensic Medicine from 2001 to 2003. Bodies with changes of decomposition (for example marbling, skin slippage, bloating or worse) were excluded. The presence of
Carboxyhaemoglobin was confirmed by toxicological analysis of femoral blood samples. Cases with under 60% saturation were excluded. Controls were taken at random. Toxicology for carbon monoxide was not performed in the controls. Permission to photograph cadavers was obtained from the local Human Research Ethics Committee. The hands of control and carbon monoxide poisoning bodies were photographed using Fuji Superia 100 ASA colour film in a Nikon F70 camera fitted with a ring flash (to ensure even illumination). The negatives of the film were scanned in a Microtek 9800 scanner at 1200 dpi and saved as .tif files. All photographs included a Kodak colour scale. Measurement of the RGB blue and red values was performed using the eyedropper tool set to 5x5 using Adobe Photoshop Elements 6 for Macintosh. Ten measurements were averaged from the fingernails in the photograph and from the grey area of the Kodak colour scale. The red value for the fingernails was divided by the red value obtained from the grey area to correct for colour cast; a similar computation was performed for the blue values. The ratio of red to blue was also calculated for the fingernail RGB data. Comparison was performed using the Mann Whitney U test on Statview v4.5 for Macintosh.

Results

There were 30 cases of carbon monoxide poisoning. The carboxyhaemoglobin saturations ranged from 64 % – 80 % (mean 74 %). In all cases the source of the carbon monoxide was from motor-vehicle exhaust fumes. The mean age of the carbon monoxide deaths was 43.3 years, there were 28 males and the mean postmortem interval (time between death and autopsy) was 1.8 days. The corresponding figures for the 30 cases in the control group were 52 years, 17 males and 2.4 days. Causes of death in the control group were trauma 9, coronary artery disease or its complications 9, hanging 3, drug toxicity (not carbon monoxide) 2, pneumonia 2, pulmonary thromboembolus 1, metabolic 1, epilepsy 1, subarachnoid haemorrhage 1, drowning 1. The best differentiator was the ratio of red to blue RGB values for the fingernails between the two groups (p<0.02). The red value of the fingernails when corrected for colour cast using the red value obtained from measurement of the standard grey region of the colour card was also significantly different between the carbon monoxide and control groups, but to a lesser degree (p<0.03); the mean red value was higher in the carbon monoxide group. However, reference to the box plots of the data (fig 1) shows a large degree of overlap between the two groups (carbon monoxide poisoning and controls).

Discussion

A level of 60% carboxyhaemoglobin was taken as the cut-off for determining death to be due to carbon monoxide poisoning (4, 12-14). Levels of up to 5% or even 10% could be expected in the controls (13-16). However, controls were not tested due to restrictions imposed by Coroner’s and Human Tissue Acts that render it illegal to perform research (non-diagnostic testing) on human tissue (which includes blood). Nonetheless, examination of the circumstances of death rendered it unlikely that any of the control cases would have significant blood carboxyhaemoglobin levels.
During the study it was noted that the colouration varied between fingernails and that even within a fingernail the colour could vary. For this reason, all the fingernails in the photograph were sampled and varying regions within nails were sampled. Analysis of the colour data revealed that the fingernails of cadavers with significant carboxyhaemoglobin levels tend to be redder than in a control population, with a statistically significant difference between the two groups. However, due to the overlap between the two groups, observation of the fingernails cannot be recommended as a rapid screening test.

Acknowledgements
The author wishes to acknowledge the assistance of staff of the former Westmead Department of Forensic Medicine, Dr JHK Grieve for the inspiration for this project and the NSW Coroner for allowing the publication of the data.

Key Points:
1. Significant blood carboxyhaemoglobin levels cause pink colouration of the blood that may be observed through the fingernails of deceased victims
2. Although the fingernail regions of cadavers with significant carboxyhaemoglobin blood levels are shifted towards red when compared with control cases, there is overlap between the two groups.

Figure Legends
Figure 1: Box plots. Left showing results of dividing red by blue values obtained from fingernails. Right showing fingernail red values corrected by the red value measured from grey card included in photographs. Results are split for control and carboxyhaemoglobin poisoning.

References:

Fig 1