Rapid Salbutamol Screening from Micro-volume Blood Samples using GC-MS – Applications to Paediatric Asthma

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Salbutamol Screening

Salbutamol is a medication used to relieve the symptoms of asthma, bronchospasm and chronic obstructive pulmonary disease (COPD).

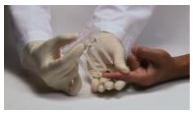
- First line therapy: inhalation.
- Next step: intravenous administration.

At present there are insufficient data to recommend a dosage regime for children and extrapolation of data from one age group to another is recognized to be unsafe[1]. Excessive administration can lead to lactic acidosis, which precipitates respiratory failure[2].

- Research gap: dose finding studies to determine optimal doses for paediatric asthma applications.
- Challenge: measuring blood concentrations of a drug in children.
- Research aim: development of a novel assay to detect and quantify salbutamol from micro-volumes of blood.

Blood Samples

- Dried Blood Spots (DBS): spotting and drying of micro-volumes blood onto filter paper[3].
- Sampling issues using filter paper: haematocrit variation, sample inhomogeneity and unknown sample volumes.



Other blood collection devices have been introduced. One of them is used alongside filter paper in this study; Volumetric Absorptive MicroSampling (VAMS)*, also named Mitra[4].

*kindly donated by James B. Rudge, Phenemonex

- Research samples: 10 µl blood samples spiked with salbutamol directly after spotting and with d3-salbutamol as internal standard after drying and extraction. The samples were evaporated to dryness and derivatised with BSTFA plus 1% TMCS prior to analysis with a developed GC-MS method.

Assay Performance

- Lower limit of quantification (LLOQ): 5 ng/ml - below the therapeutic range.
- Accuracy and precision: <20% for LOD samples and <15% for other samples, meeting the guidelines for bioanalytical method validation of the United States Food and Drug Administration (US FDA)[5].
- Recovery: salbutamol recovered from DBS shows a higher recovery (86%) compared to blood sampled with the Mitra device (68%). This applies, however, only to samples left to dry for 2 hours and processed immediately upon drying.
- Stability: DBS samples show a decreasing recovery with increasing drying time compared to Mitra tip samples (Fig 1).
- Application test: three healthy volunteers were administrated 1 mg of salbutamol via a 100 µg per dose inhaler and spacer (Fig 2).

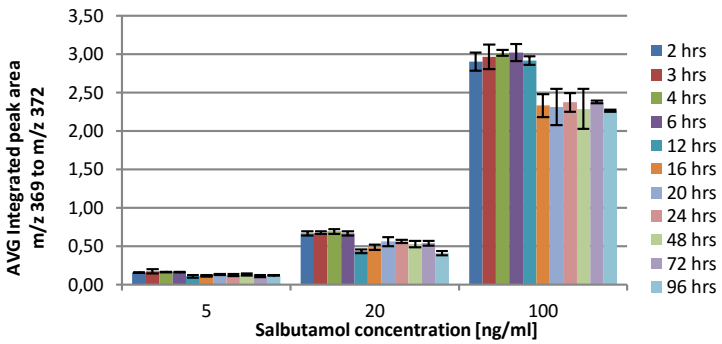


Figure 1: Effect on salbutamol recovery from DBS samples of increasing drying/storage times.

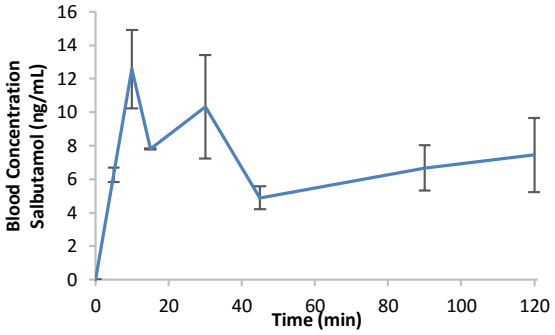


Figure 2: Screening of salbutamol over time from 10 µl blood sampled with Mitra tips.

Conclusion

- An assay to detect and quantify salbutamol from micro-volumes of blood has been developed, optimised and validated according to US FDA guidelines, especially valuable for paediatric asthma applications.
- Two blood collection devices were tested and compared for this study; the novel developed VAMS devices have demonstrated sampling and stability advantages compared to the conventional filter cards used for DBS research and applications.
- As a final method evaluation, salbutamol could be detected in samples from three healthy volunteers dosed with salbutamol.

References

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