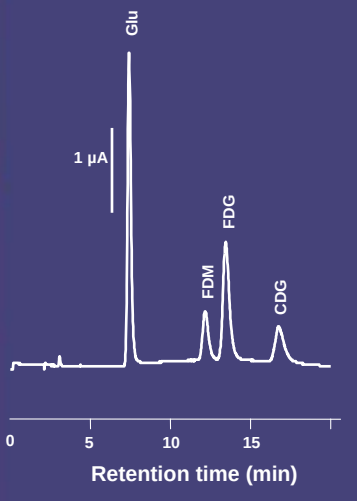
FLUORODEOXYGLUCOSE

THE MOST RELIABLE LC-EC APPLICATIONS FOR PHARMACEUTICAL & BIOTECH ANALYSIS EVER FORMULATED

- Aminoglycosides
  - *Amikacin*
  - *Framycetin Sulphate*
  - *Gentamicin Sulphate*
  - *Kanamycin Sulphate*
  - *Lincomycin*
  - *Neomycin*
  - *Spectinomycin*
  - *Tobramycin*
- PET imaging tracer
  - *FDG*
- Macrolide antibiotics
  - *Azithromycin*
  - *Azaerythromycin*
  - *Clarithromycin*
  - *Erythromycin*
  - *Roxithromycin*
- Bioanalysis of pharmaceutics
  - *Artemisinin*
  - *Dihydro-artemisinin*
  - *Artemether*
  - *Etoposide*
  - *8-OH-DPAT*
  - *mesna BNP7787*
  - *Vincristine*



INTRODUCTION

In PET imaging, 2-[^{18}F]fluoro-2-deoxy-D-glucose (FDG) can be used for the assessment of glucose metabolism in the heart, lungs, and the brain. It is also used for imaging tumors in oncology, where usually dynamic images are analyzed in terms of Standardized Uptake Values. The 109.8 minute half-life of ^{18}F makes rapid and automated chemistry necessary; therefore the FDG is produced in a cyclotron in vicinity of the PET facility.

One of the tests that needs to be performed on the solution before it can be injection into a patient, is to check for the actual concentration of FDG, and the presence of the by-products 2-fluoro-2-deoxy-D-mannose (FDM) and 2-chloro-2-deoxy-D-glucose (CDG).

For the analysis of FDG, the EP [1] requires a *'detector suitable for carbohydrates'*. A detector that is sensitive enough and easy to use is pulsed amperometric detection (PAD) [2].

- Glucose, FDG, FDM and CDG
- PET imaging tracer
- European Pharmacopoeia compliant
- Reproducible & Robust

Summary

A method is described for the analysis of FDG using the ALEXYS FDG Analyzer. The method shows excellent detection limits and linearity. It meets the EP requirements for selectivity as also the metabolites FDM and CDG are measured.
For pulsed electrochemical detection a FLEXCELL has been used which has an exchangeable working electrode, and a maintenance free reference electrode.

Fig. 1. ALEXYS FDG Analyzer.

Method

Because the number of samples per day is limited, the ALEXYS FDG Analyzer has been configured with a manual valve. The main elements of this analyzer are a almost pulse free HPLC pump, a polymer based column (resistant to the mobile phase with pH 13), and a DECADE II electrochemical detector with a gold working electrode in the flow cell.

Table 1

Conditions	
HPLC	ALEXYS FDG analyzer
Flow rate	1 mL/min
Mobile phase	4 g/L NaOH in water
Column	ALC-525 (250x4.6mm, 7um)
Flow cell	Flexcell™ with Au WE and HyREF™
Temperature	35 °C for separation and detection
Range	50 µA/V
I-cell	0.5 - 1 µA

Table 2

EP system suitability requirement		
Parameter	EP criteria	Result
Resolution FDG-FDM	>1.5	1.75
s/n of FDG	>10	373

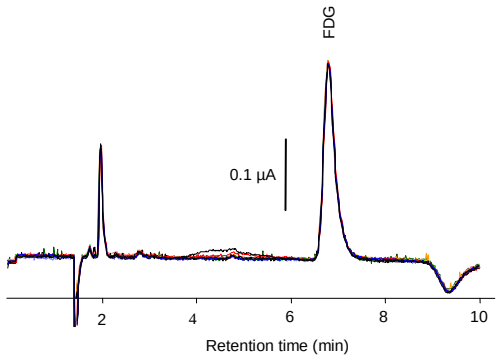


Fig. 2. FDG sample (20 µM, 20 µl injected). Overlay of 10 chromatograms. Flow rate: 1.5 mL/min.

Linearity, Repeatability and Detection Limit

Linearity of FDG was investigated in the concentration range of 5 – 1000 µM. The correlation coefficient was better than 0.999 for peak areas and peak heights in this range, as well as sub-ranges.
The relative standard deviation (RSD) in peak area for 10 replicate injections of FDG was 1.4%. The RSD for the retention times was better then 0.2%. The detection limit of FDG is 1 µM.

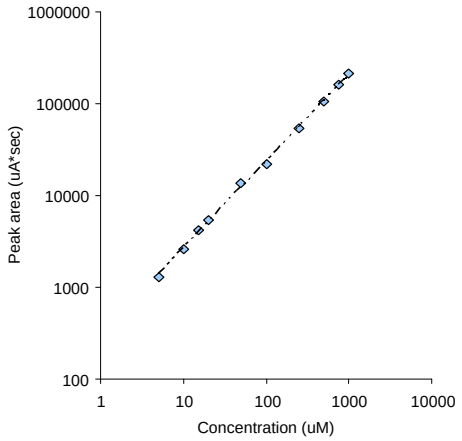


Fig. 3. Response plot of FDG.

EP requirements

In the EP monograph for FDG [1], a system suitability requirement is specified for *resolution* between FDG and FDM, and *signal-to-noise ratio* of FDG. The 'reference c solution' was prepared according to the guidelines given in the EP, taking into account a 'maximum recommended dose, in millilitres' of 20 mL. In this case, reference solution c has a final concentration of 250 mg/L FDM and 12.5 mg/L FDG (=1.3 mM FDM and 70 µM FDG).
In Table 2 the EP requirement is compared with the typical results obtained with ALEXYS Carbohydrates analyzer, on the basis of the analysis of 'reference solution c'.
It is evident from Fig. 4 that the detection limit and separation requirement as summarized in Table 2 are easily met by the ALEXYS Carbohydrates analyzer.

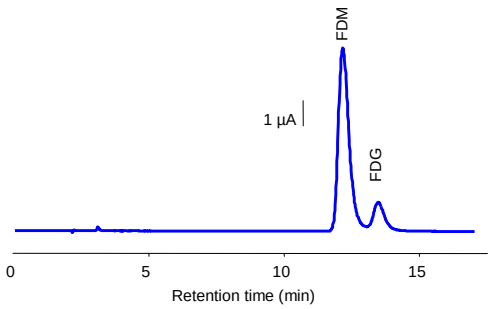


Fig. 4. 'Reference solution c', with a final concentration of 1.3 mM FDM and 70 µM FDG, 20 µl injected.

By-products of FDG production

According to the European Pharmacopoeia [1], the FDG solution needs to be tested among others for impurity 'A', which is CDG. Fig. 5 shows the chromatogram with the signals of Glucose, FDG, FDM and CDG.

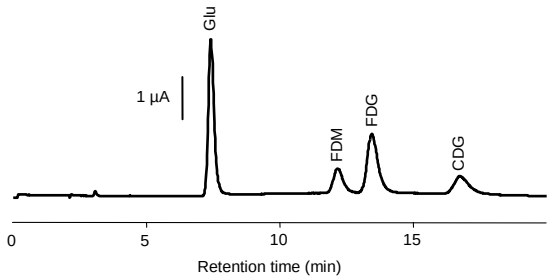


Fig. 5. Mix of 50 µM Glucose, 100 µM FDM, 100 µM FDG and 100 µM CDG in water (20 µl injected).

CONCLUSION

The ALEXYS® FDG analyzer provides a reliable solution for the analysis of FDG and its by-products. It meets the EP requirements for separation and sensitivity.

References

1. Fludeoxyglucose (^{18}F) Injection, European Pharmacopoeia 6.2, (2008) 3678-3680

2. W.R. Lacourse, "Pulsed Electrochemical Detection In High Performance Liquid Chromatography", John Wiley & Sons, New York, 1ed,1997.

PART NUMBERS	
180.0053A	ALEXYS FDG Analyzer with manual injector
Reordering information for application specific consumables	
250.1080	ALC-525 column, 250 x 4.6mm, particle size 7um
250.1084	ALC guard column replacement cartridges (5/pk)
250.1700	In-line filter (aqueous)