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INVESTIGATION OF URINARY EXCRETION OF HYDROXYETHYL STARCH AND DEXTRAN BY UHPLC-HRMS IN DIFFERENT ACQUISITION MODES

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ABSTRACT: Plasma volume expanders (PVEs) such as hydroxyethyl starch (HES) and dextran are misused in sports because they can prevent dehydration and reduce haematocrit values to mask erythropoietin abuse. Endogenous hydrolysis generates multiple HES and dextran oligosaccharides which are excreted in urine. Composition of the urinary metabolic profiles of PVEs varies depending on post-administration time and can have an impact on their detectability. In this work, different mass spectrometry data acquisition modes (full scan with and without in-source collision-induced dissociation) were used to study urinary excretion profiles of HES and dextran, particularly by investigating time-dependent detectability of HES and dextran urinary oligosaccharide metabolites in post-administration samples. In-source fragmentation yielded the best results in terms of limit of detection (LOD) and detection times, whereas detection of HES and dextran metabolites in full scan mode with no in-source fragmentation is related to recent administration (< 24 hours). Urinary excretion studies showed detection windows for HES and dextran respectively of 72 and 48 hours after administration. Dextran concentrations were above the previously proposed threshold of 500 $\mu g \cdot mL^{-1}$ for 12 hours. A "dilute-and-shoot" method for the detection of HES and dextran in human urine by ultra-high-pressure liquid chromatography-electrospray ionization-high resolution Orbitrap™ mass spectrometry was developed for this study. Validation of the method showed an LOD in the range of 10-500 $\mu g \cdot mL^{-1}$ for the most significant HES and dextran metabolites in the different modes. The method allows retrospective data analysis and can be implemented in existing high-resolution mass spectrometry-based doping control screening analysis.

KEY WORDS: LC-HRMS, hydroxyethyl starch, Dextran, plasma volume expanders, doping, Orbitrap

INTRODUCTION

Hydroxyethyl starch (HES) and dextran are polysaccharide-based plasma volume expanders (PVE) which consist respectively of high molecular weight hydroxyethylglucose and glucose polymers with heterogeneous structure and properties. They are used for treatment of hypovolaemia and haemorrhage as plasma substitutes since they can significant increase plasmatic volume [14,27]. Nowadays, therapeutic use of dextran is less common compared to HES due to the increased risk for anaphylactic shock [11].

The use of PVEs is prohibited in sports because they can be used to prevent dehydration and because they can mask the use of erythropoietins or blood doping, since they can influence the blood passport by decreasing haemoglobin and haematocrit values [20]. For these reasons, PVEs are included by the World Anti-Doping Agency (WADA) in the Prohibited List of substances and methods [26].

Aside from colorimetric microtitre plate methods [2,7,18], used for screening due to the low cost and limited sample work up, detection of HES and dextran in doping analysis can be accomplished by mass spectrometric techniques including gas chromatography–mass spectrometry (GC-MS) [15,16, 21,23], liquid chromatography–mass spectrometry (LC-MS) [4,9,10,13] and matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF) [17]. Due to the polar nature and high molecular weight of HES and dextran, analysis by GC-MS requires an acidic hydrolysis step and a time-consuming derivatization step [15].

During the last decade, LC-MS has proven to be the ideal technique for doping control analysis of polar and high molecular weight compounds [25]. Among the LC-MS methods described for the detection of PVEs in urine, those from Guddat [9] and Kolmonen [13] were

based on in-source fragmentation (isCID) to generate low mass fragment ions from the sugar backbone and can be analyzed with a "dilute-and-shoot" strategy.

HES and dextran show a complex urinary excretion profile due to their macromolecular nature. HES is available as a mixture of chemically modified glucose polymers with an average molecular weight (MW) of 130 kDa, whereas dextran products have typically an average molecular weight of 40 (DEX 40) or 70 kDa (DEX 70) [14,19,22].

Since the nephron tubules only allow excretion of compounds with an MW less than 50 kDa, fractionation during urinary elimination can be observed as low MW components rapidly excreted in urine whereas high MW components are not filtered [1,12]. Once the high MW polysaccharides are cleaved by plasma amylase into smaller oligosaccharides (≈ 200-40 000 Da), they can be excreted in urine, with a speed inversely correlated to their size [13,17].

In this work, the urinary excretion profiles in time of HES and dextran as a matter of doping control were investigated using the conventional isCID approach. Additionally, since detection of HES and dextran oligosaccharides in full scan mode without in source CID (FSMS) is not described yet, the latter was also investigated.

Experiments were carried out by means of ultra-high pressure-liquid chromatography coupled to Orbitrap™ high resolution mass spectrometry (UHPLC-HRMS). This technology has not yet been described for the detection of polysaccharide-based PVEs.

MATERIALS AND METHODS

Chemicals. The commercial solution Voluven® (average molecular weight: 130 kDa; molar substitution: 0.4; concentration: 6% w/v) was used for HES. Dextran (average molecular weight: 40 kDa) was purchased from Fluka Chemie AG (Buchs, Switzerland).

Ammonium formate (NH_4COOH) and HPLC-grade water (H_2O) and acetonitrile (ACN) were from Biosolve (Valkenswaard, The Netherlands). Formic acid (FA) and sodium azide (extra pure) were from Merck (Darmstadt, Germany). TFA was from Fluka (Bornem, Belgium). The internal standard (2-hydroxypropyl)-β-cyclodextrin (HPCD) was obtained from Sigma–Aldrich (Bornem, Belgium). Internal standard solution containing 100 $\mu g \cdot mL^{-1}$ of HPCD was prepared in water. Gas used for the mass spectrometer was nitrogen (Alphagaz2-grade) from Air Liquide (Desteldonk, Belgium).

Sample preparation

HES and dextran oligomers were obtained by hydrolysis with trifluoroacetic acid (TFA) 3M, adapting a procedure previously described by Gallego and Segura [6]. Briefly, hydrolyzed HES (10 $mg \cdot mL^{-1}$) was prepared by pipetting 150 μL of TFA in a tube containing 500 μL of the commercial solution Voluven®. For dextran, 10 μL of 10 $mg \cdot mL^{-1}$ solution were dissolved in 500 μL of 3M TFA. Both solutions were incubated for 30 minutes at 80°C. The hydrolysed material was dried under a stream of nitrogen at 60°C and redisolved in water to the desired concentration.

An aliquot of 100 μL of urine was transferred to an Eppendorf type vial containing 870 μL of 95:5 H_2O:ACN, 1 mM NH_4COOH, 0.01% FA and 30 μL of HPCD. After centrifugation for 5 minutes at 10 000 rpm, 20 μL were injected into the chromatographic system.

Liquid chromatography

The liquid chromatographic system was an Accela LC (Thermo Scientific, Bremen, Germany) equipped with degasser, Accela 1250 pump, autosampler thermostated at 10°C and a heated column compartment. The column employed was a Zorbax SB-C8, 2.1 x 50 mm and 1.8 μm particle size from Agilent Technologies (Böblingen, *Germany*) thermostated at 25°C. The mobile phases were 1 mM NH_4OAc/0.01% FA in water (A) and 0.01% FA in acetonitrile (B). The gradient program was as follows: 100% A for 0.5 min, then decreased linearly to 20% in 7.0 min, and finally decreased linearly to 0% in 0.5 min and held at 0% for 1.5 min followed by an increase to the initial concentration of 100% A in 0.1 min. Equilibrium time was 2.4 min resulting in a total run time of 12 min. The flow rate was set constant at 250 $\mu L \cdot min^{-1}$ and the injection volume was 20 μL.

High-resolution mass spectrometry

The LC effluent was pumped to an Exactive benchtop, Orbitrap™-based mass spectrometer (Thermo Scientific, Bremen, Germany) operated in the positive-negative polarity switching mode and equipped with a heated electrospray ion source (HESI). Nitrogen sheath gas flow rate and auxiliary gas were set at 60 and 30 (arbitrary units), respectively. The capillary temperature was 350°C, the spray voltage 3 kV or -3 kV and the capillary voltage 30 V or -25 V, in positive or negative ion modes, respectively. The instrument operated from m/z 150–3000 at 50,000 resolving power in FSMS and isCID at 80 eV. The automatic gain control (AGC) was set to 10e6. The data acquisition rate was 2 Hz. The Orbitrap™ performance in both positive and negative ionization modes was evaluated daily and when it failed, external calibration was performed with Exactive Calibration Kit solutions (Sigma-Aldrich, St. Louis, USA and ABCR GmbH & Co. KG, Karlsruhe, Germany). A mass extraction window of 5 ppm was used.

Method validation

The validation was carried out following Eurachem validation guidelines [5]. Ten human urine samples, declared negative after routine doping analysis, were spiked with HES and dextran at a measurement range from 10 to 1000 $\mu g \cdot mL^{-1}$. The limit of detection (LOD) was defined as the lowest level at which a diagnostic ion could be identified in all 10 urine samples with a signal-to-noise (S/N) ratio greater than 3 and a retention time difference of less than 0.3 min to the reference.

For dextran semi-quantitation, calibration curves from 10 to 1000 $\mu g \cdot mL^{-1}$ (10, 50, 100, 500, 1000) were used. 30 μL of in-

ternal standard (100 $\mu g \cdot mL^{-1}$) were added. The precision of the method was checked by repeatability (intraday, n=6), intermediate precision (interday, n= 2), and reproducibility (interoperator, n= 2). Intraday repeatability was studied by calculating the percent relative standard deviation (% relative standard deviation, RSD) for 6 determinations each of peak area of dextran (500$\mu g \cdot mL^{-1}$) performed on the same day. The matrix effect was calculated by comparing the peak areas in reference samples, containing only water, with the peak areas of the 10 urine samples (spiked at 500 $\mu g \cdot mL^{-1}$), after analysis in triplicate.

Selectivity was tested during the validation procedure. The 10 blank urines used for determining detection capability were analyzed as described above.

Selectivity was further tested by analyzing urine samples containing common doping agents including diuretics, stimulants, β-blockers, β-agonists, narcotics, corticosteroids, anabolic steroids and non-steroidal anti-inflammatory drugs. Similarly to Kolmonen et al. [13], also urine samples from 20 diabetics were analyzed to evaluate abnormal excretion of endogenous oligosaccharides.

Excretion samples

Excretion urine samples of HES and dextran were collected at the Ghent University Hospital during a controlled study approved by the ethical committee (EC UZG 2006/178). Two patients received 500 mL of Volulyte® (average molecular weight: 130 kDa; molar substitution: 0.4; concentration: 6% w/v), and two patients received 500 mL of Rheomacrodex® (average molecular weight: 40 kDa; concentration: 10% w/v), both in a 2-hour period. Samples were collected before and until 5 days after administration.

RESULTS & DISCUSSION

Mass spectrometry. The high number of molecular species originating from the in vivo hydrolysis of HES and dextran yields complex spectra with plenty of information (Figure 1). Investigation of the total ion current chromatograms of post-administration samples showed for HES and dextran typical ion clusters with constant m/z shifts, as already presented in previous works [3,6,9]. The clusters were observed in positive and in negative mode with different ion types for both PVEs, including molecular or adduct ions, in positive and negative mode.

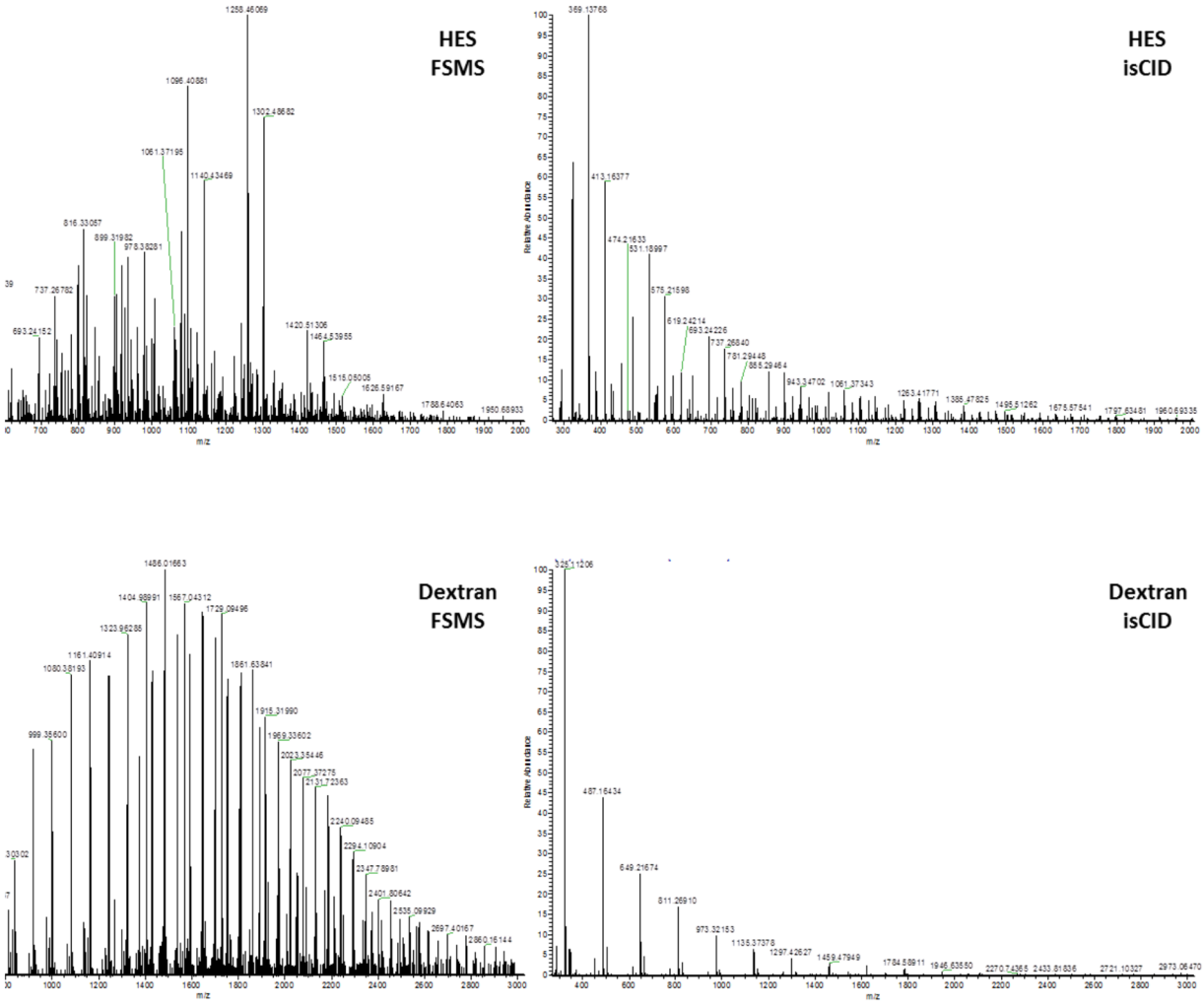

FIG. 1. MASS SPECTRA OF HES AND DEXTRAN IN FSMS AND ISCID MODE

An in silico approach was applied to identify the components of the clusters observed. In the positive mode, predicted minimal m/z values were 181.0706 (single-charged ions) corresponding to a glucose (Glu) residue with no hydroxyethyl (HE) groups added. Δm/z values for the corresponding sodium (M+22.9892 Da), potassium (M+38.9631 Da) and ammonium (M+18.0338 Da) adducts were also calculated. In the negative mode, minimal m/z= 179.0561 (molecular ion) was predicted, and also formate adducts (M+44.9982 Da) were considered. Δm/z= 162.0528 was predicted for any additional Glu monomer. These calculations were valid for HES and dextran, since they both present a glucose backbone. Loss of water (Δm/z= -18.0106), a typical fragmentation product of polysaccharides, was also registered. For HES, additional Δm/z= of 44.0262 for an additional HE group was calculated.

HES and dextran showed very similar ionization properties. In fact, when detecting HES and dextran, multiple ions were recorded both in positive ($[M+H]^+$, $[M+Na]^+$, $[M-K]^+$, $[M-H_2O+Na]^+$, $[M+NH_4]^+$) and in negative mode ($[M-H]^-$, $[M+FA]^-$ and $[M-H_2O-H]^-$). Relative abundances of the ion types of interest varied significantly. Overall, positive mode yielded the best results. In particular, $[M+H]^+$, $[M+Na]^+$, $[M+K]^+$ and $[M-H_2O+H]^+$ gave the best MS response and allowed for the longest detection window. For both PVEs, abundances of Na^+ and K^+ adducts were generally comparable for the two different acquisition modes, whereas peaks of protonated ions were generally higher when isCID was used (Figure 2). Also negative ionization was observed, as previously described by Cmelik et al. [3], but showed less sensitivity than the positive mode.

Detection of metabolites in excreted urine in FSMS and isCID modes

As expected, a large number of metabolites were excreted in urine for both PVEs. The diagnostic ions for the metabolites covered the whole scan range. Therefore, three high-abundant ions were selected as representative for low, medium and high m/z ions and

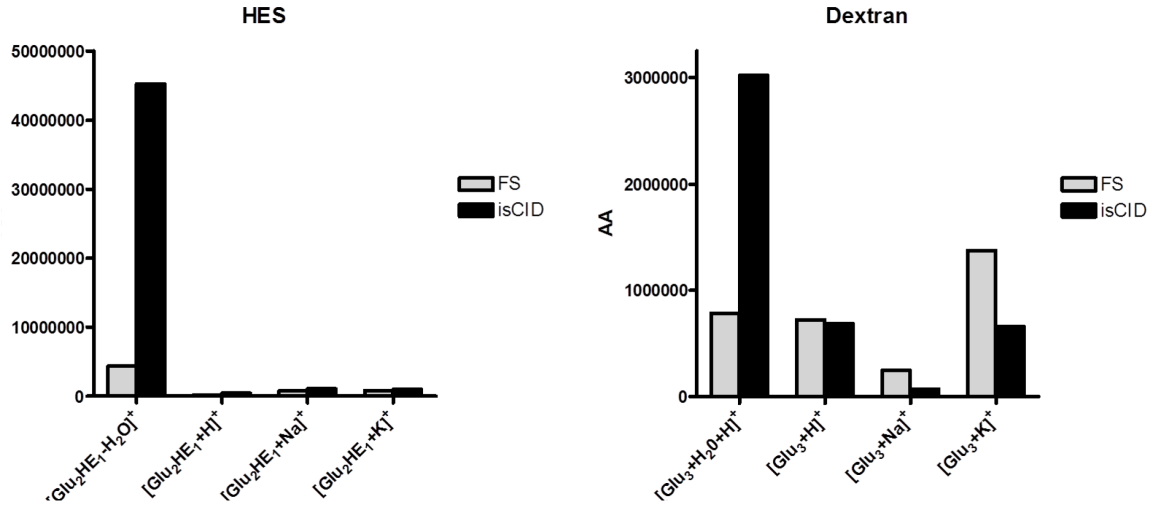

FIG. 2. COMPARISON BETWEEN MS RESPONSE OF DIFFERENT ION TYPES FOR THE REPRESENTATIVE HES METABOLITE Glu_2HE_1 (LEFT) AND DEXTRAN METABOLITE GLU_3 (RIGHT) IN REFERENCE STANDARD SOLUTIONS

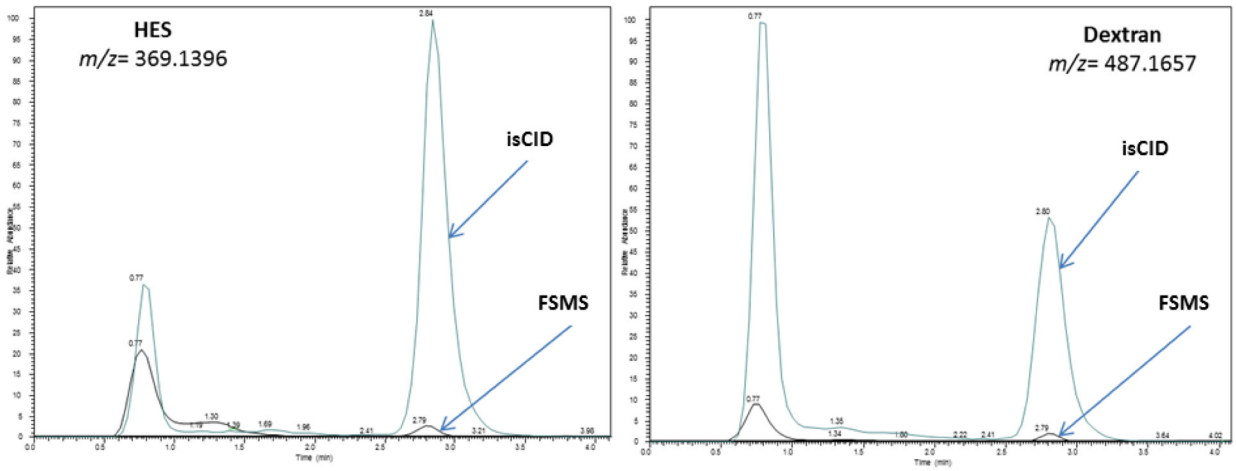

FIG. 3. EXTRACTED ION CHROMATOGRAMS FOR HES AND DEXTRAN REPRESENTATIVE IONS SHOWING THE PRESENCE OF MULTIPLE PEAKS

detectable both in FSMS and in isCID mode (respectively $[Glu_2HE_1\text{-}H_2O+H]^+$ with m/z= 369.1396, $[Glu_5HE_3\text{-}H_2O+H]^+$ with m/z= 943.3500 and $[Glu_9HE_4\text{-}H_2O+H]^+$ with m/z= 1635.5875 for HES, and $[Glu_3\text{-}H_2O+H]^+$ with m/z= 487.1657, $[Glu_6+Na]^+$ with m/z= 1013.3157 and $[Glu_{10}+Na]^+$ with m/z= 1661.5270 for dextran).

Full scan analysis without isCID allows for detection of intact oligosaccharides, produced by in vivo hydrolysis. Isobaric species can also be generated by isCID from bigger polysaccharides (< 2-3 kDa).

Total ion current, but also extracted chromatograms for dextran (Figure 3), showed distorted peaks with a 2-minute peak width. This is most likely due to the broad molecular weight distribution of dextran metabolites including molecules, including multiple isobaric isomers, with an MW of 1 kDa up to 50 kDa, eluting at different retention times. Additionally, the presence of a certain species is also dependent on the excretion stage.

The presence of multiple metabolite populations for both HES and dextran has been previously described [13].

Figures 4 and 5 show chromatograms for HES and dextran in samples from excretion studies at different post-administration times. In the excretion samples, HES' most abundant population eluted at 2.8 min, whereas for dextran the most abundant peaks were recorded at 0.8 min (Figure 3).

Use of isCID yielded a higher MS response for all 6 investigated ions. This can be explained by the fact that, in isCID mode, the same fragment can be generated by a high number of metabolites with different masses, whereas for FS acquisition only a few isomeric oligosaccharides, with identical chemical formula, contribute to the signal.

Concerning HES, most of the detected species show similar n_{HE}/n_{Glu} ratios, reflecting the fact that the Voluven® degree of substitution is 0.4. $[Glu_2HE_1\text{-}H_2O+H]^+$ (molecular formula: $C_{14}H_{24}O_{11}$, which reveals loss of a H_2O molecule from monohydroxyethylated maltose) represents a very good marker for HES administration and was also described by previous research groups. $[Glu_2HE_1\text{-}H_2O+H]^+$ was also used in the work from Kolmonen et al. [13] and Guddat et al. [9], though in these papers it was referred to as $[Glu_2HE_1+H]^+$, which has a theoretical m/z= 387.1502 and has been used by Mazzarino et al. [16]. The high abundance of this ion in FSMS mode demonstrates that monohydroxyethylated maltoses are the main metabolites. Contemporarily, sensitive detection in isCID shows that fragment ions with the same m/z values are common as well. $[Glu_2HE_1\text{-}H_2O+H]^+$ was still detectable after 72 hours both with and without isCID, whereas $[Glu_5HE_3\text{-}H_2O+H]^+$ could be detected only in isCID mode after 24 hours. Larger metabolites and fragments such as $[Glu_9HE_4\text{-}H_2O+H]^+$ were only detected in the excretion samples until 6 hours after administration (data not shown).

Dextran ions were all clearly detected after 3 and 12 hours, both with isCID and without isCID (Figure 5). They were not detected in FS mode after 24 hours, whereas the use of isCID prolonged the detection window up to 48 hours. This is in agreement with previous work, which showed that in the 0-24 h fraction up to 5 kDa is excreted [13] and can be detected without isCID.

It can be concluded that contemporary detection in FS and isCID mode is particularly interesting since it can provide important information on the administration time. Taking into account that the duration of volume expansion is 8-12 hours for HES and 6-12 hours for dextran [22], detection of multiple metabolites both with and without isCID can therefore discriminate whether the PVE has been administered on the same day of the doping control.

Method validation

HES

For the validation, 130 kDa HES was partially hydrolyzed to yield lower molecular weight HES. This was necessary because the 130 kDa HES could not be detected without isCID because the high molecular weight species could not enter the mass spectrometer. Validation results for HES are presented in Table 1. Due to the presence of multiple peaks, it is very important to select the retention time window for the most abundant of them to increase sensitivity. In this case, the peak at 2.8 min was selected.

HES showed the lowest LOD of 10 $\mu g \cdot mL^{-1}$ for the ion $[Glu_2HE_1\text{-}H_2O+H]^+$ in isCID mode; this LOD is comparable to those from previous works [9,13,16]. A great difference was observed with FSMS where for the same ion an LOD of 500 $\mu g \cdot mL^{-1}$ was calculated. This difference in sensitivity has already been explained by the fact that, in isCID mode, the same fragment can be generated by

TABLE 1. VALIDATION DATA

	Ion	m/z	LOD (µg/mL)				Ion suppression (%)			
			FS	CID	HCD	HCD + CID	FS	CID	HCD	HCD + CID
HES	$[Glu_2HE_1+H]^+$	369.1396	50	10	10	50	51.8	35.8	57.4	44.5
	$[Glu_5HE_3+Na]^+$	983.3420	100	50	50	50	68.7	-12.3	72.3	21.7
	$[Glu_{10}HE_4+Na]^+$	1837.6318	ND	500	ND	500	-	28.3	-	27.2
Dextran	$[Glu_3+H]^+$	487.1657	500	50	50	100	78.6	70.2	88.6	79.8
	$[Glu_6+Na]^+$	1013.316	500	50	100	50	83.3	24.7	82.0	35.8
	$[Glu_9+Na]^+$	1499.474	ND	500	ND	500	-	32.4	-	23.9

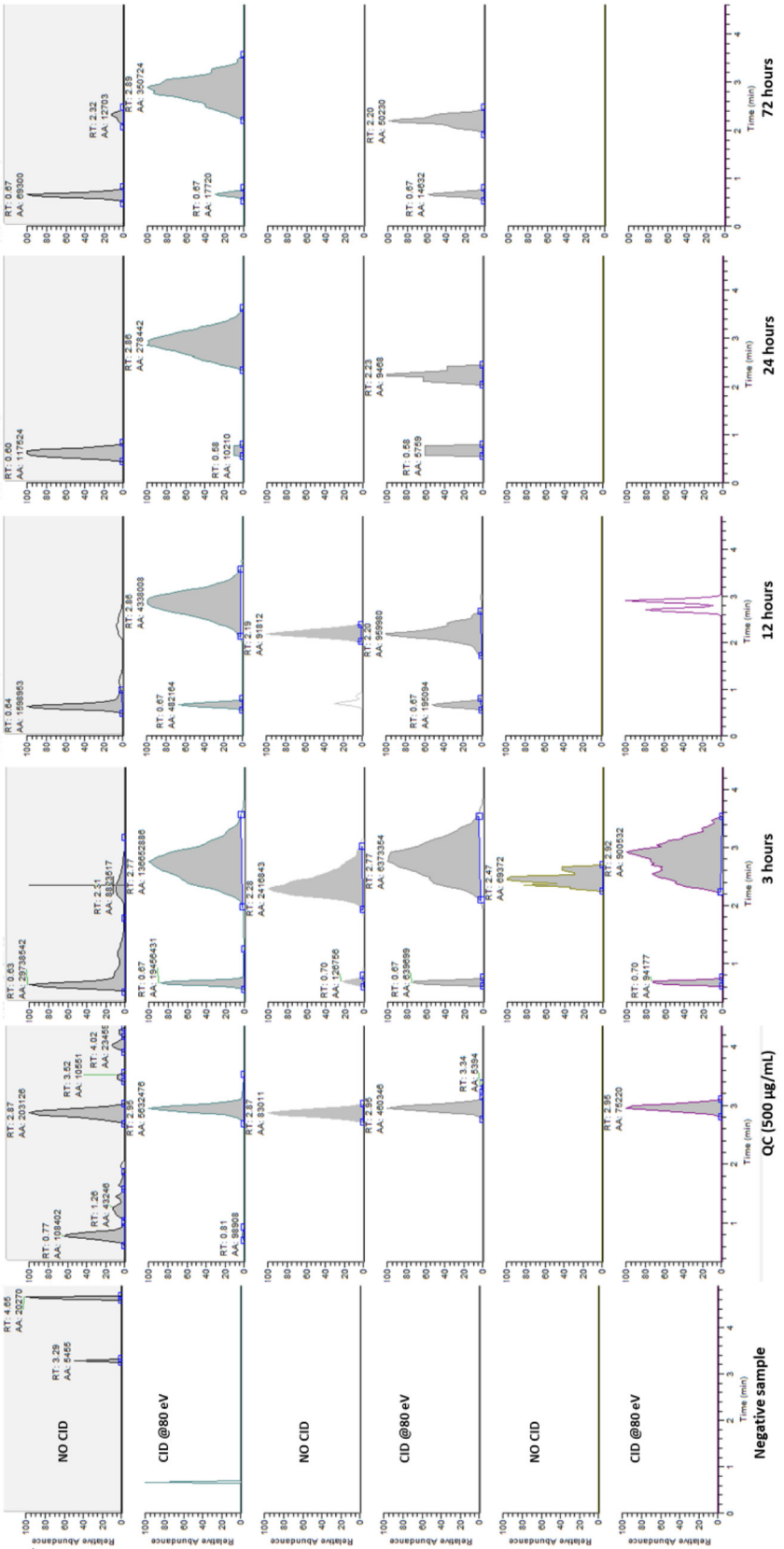

FIG. 4. DETECTION OF THREE REPRESENTATIVE HES IONS AT DIFFERENT POST-ADMINISTRATION TIMES, WITH AND WITHOUT CID. NEGATIVE URINE AND URINE SPIKED WITH HYDROLYSED HES (500 µg · mL^{-1}) ARE ALSO SHOWN

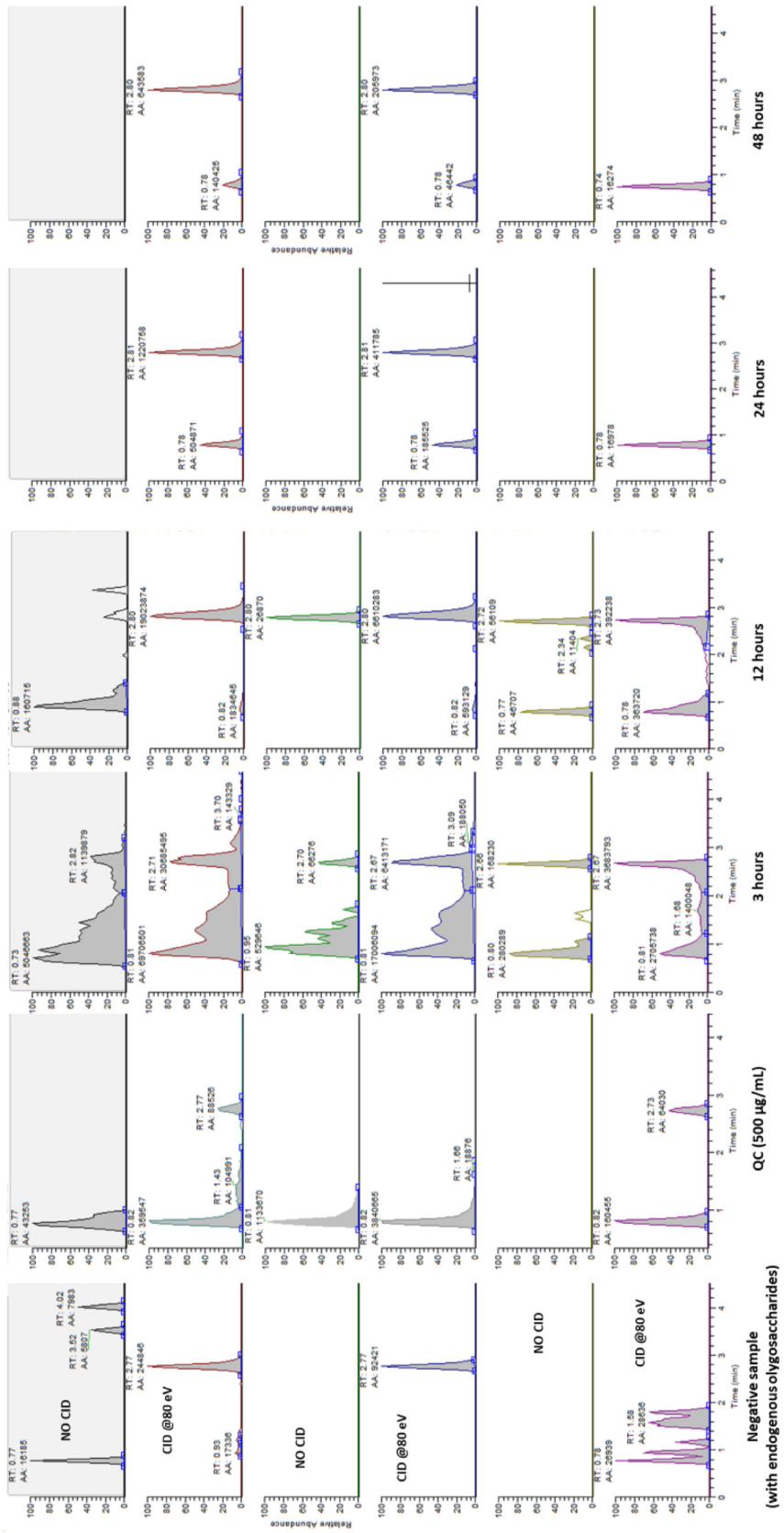

FIG. 5. DETECTION OF THREE REPRESENTATIVE DEXTRAN IONS AT DIFFERENT POST-ADMINISTRATION TIMES, WITH AND WITHOUT ISCID. A REPRESENTATIVE NEGATIVE URINE (CONTAINING ENDOGENOUS OLIGOSACCHARIDES) AND A NEGATIVE URINE (NO ENDOGENOUS SACCHARIDES DETECTED) SPIKED WITH HYDROLYZED DEXTRAN (500 $\mu g \cdot mL^{-1}$) ARE ALSO SHOWN

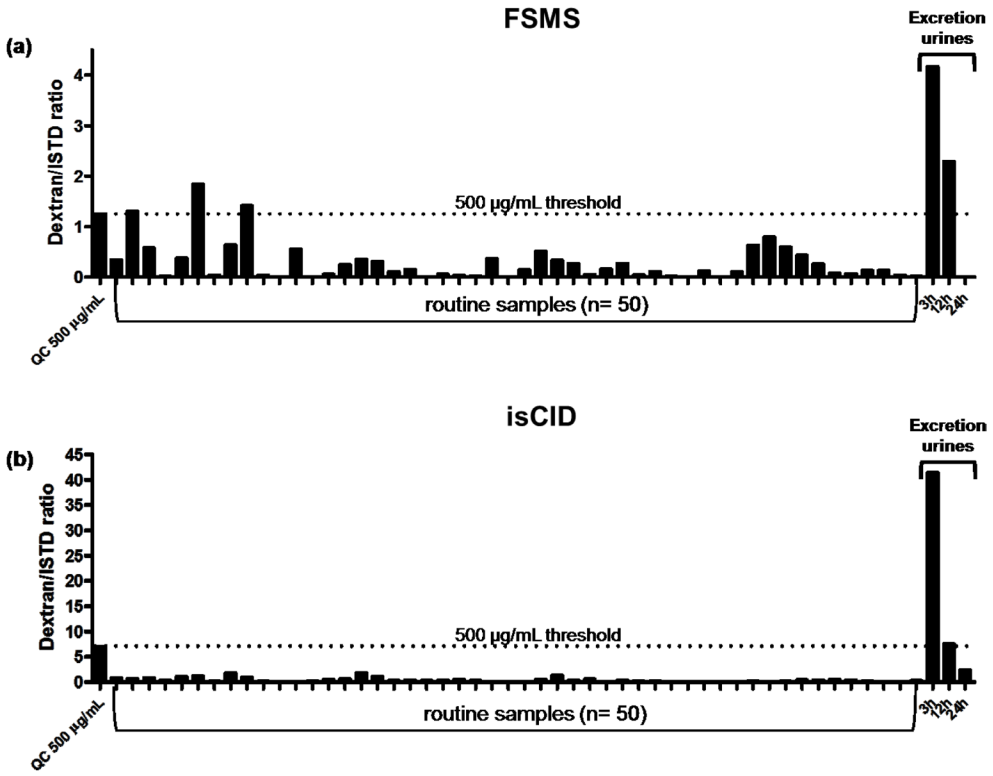

FIG. 6. SEMI-QUANTITATIVE DETERMINATION OF $[Glu_3\text{-}H_2O+H]+$ CONCENTRATION IN A REPRESENTATIVE BATCH OF ROUTINE SAMPLES (N= 50), COMPARED TO A QUALITY CONTROL SAMPLE SPIKED AT 500 $\mu g \cdot mL^{-1}$ IN FSMS (A) AND ISCID (B) MODE AND 3, 12 AND 24 HOURS EXCRETION SAMPLES.

several metabolites with higher masses, whereas for FSMS only a few isomeric oligosaccharides, with identical chemical formula, contribute to the signal. The matrix effect for HES was not significant, with even a slight trend to ion enhancement for all 3 ions (Table 1). No interferences were detected in blank samples, diabetics' urine or urine containing other doping agents, showing excellent selectivity of the method.

Dextran

Similar to the validation of HES, dextran with an average molecular weight of 40 kDa yielded a poor signal when analyzed without isCID. Therefore, partial hydrolysis of dextran 40 kDa was also performed.

Compared to HES, selectivity is a bigger issue for dextran because physiopathological excretion of hexose-containing oligosaccharides can theoretically generate false positive results. To solve this problem, a threshold of 500 $\mu g \cdot mL^{-1}$ has been proposed for screening purposes [9]. Indeed, several negative samples, analyzed with this method, showed an isCID signal from endogenous oligosaccharides with 10-100 fold lower signals than the suggested threshold (Figure 6b).

In FSMS mode, the molecular ion of quadruply hydroxypropylated cyclodextrin at m/z= 1384.5685 was used for semi-quantitation, whereas the fragment ion corresponding to the dihydroxypropyl isomaltose at m/z= 441.1966 was selected.

$[Glu_3\text{-}H_2O+H]^+$ was selected among the numerous diagnostic ions for dextran semi-quantitation both in FSMS and isCID mode, providing the best compromise between sensitivity and linearity. The peak at 2.7 minutes was selected for semi-quantitation since the calculated dextran concentration allowed to confirm recent administration (>500 $\mu g \cdot mL^{-1}$ after 12 hours), although early-eluting species are more abundant after 3 hours (Figure 5). However, once a suspicious peak at 2.8 is detected, it is recommended to the analyst to check the whole elution window (0.6-2.7 min) to have further evidence before confirmation analysis.

Intraday repeatability, interday precision, and reproducibility were respectively 6.1%, 20.9% and 10.9% in the FMSM mode, and 7.1%, 9.9% and 8.3% in the isCID mode. The excretion study presented in this work confirmed that the threshold proposed by Guddat is reliable to discriminate between natural excretion of oligosaccharides and recent dextran administration (up to 12 hours). Despite the elevated concentration measured in FSMS, it was not possible to define a threshold for this acquisition mode since concentrations in negative samples were comparable to the quality control samples (Figure 6a).

It should be stated that semi-quantitative results can be affected by the choice of the reference standard that is being used. In any

case, the reference can represent only partially the composition of dextran metabolites in urine, especially since this evolves over time.

Observing the data from excretion results, several low and medium m/z diagnostic ions showed abundant MS signals and the best detection windows and can be used to estimate dextran concentration in the sample. Since the method is for screening purposes, one of these ions can be monitored in the first instance to reveal suspicious samples, similarly as described in other high-throughput screening analyses [10]. In case a potential positive sample is detected, retrospective "post-targeted" data evaluation using multiple diagnostic ions is suggested before the necessary confirmatory analysis by partially methylated alditol acetates (PMAA) [24] or enzymatically derived isomaltose [8].

LOD in CID was well below the suggested threshold (50 $\mu g \cdot mL^{-1}$). Regression curves always showed $R^2 > 0.90$. Ion suppression in isCID was approximately 40%.

Another important issue was represented by samples from diabetic patients, which can lead to false positive results. In fact, as also described by Kolmonen et al. [13], high amounts of oligosaccharides (> 1 mg$\cdot mL^{-1}$) were detected in all samples from diabetics, both in FSMS and isCID mode. Also in this case, the above-mentioned confirmation methods can be used to discriminate dextran administration. No interferences were detected in the samples containing common doping agents.

CONCLUSIONS

Urinary excretion profiles of HES and dextran have been investigated with a "dilute-and-shoot" screening method by LC-HRMS by using different acquisition modes, investigating time-dependent detectability of HES and dextran urinary oligosaccharide metabolites in post-administration. In isCID mode, HES was detected after 72 hours, whereas dextran was present above the 500 $\mu g \cdot mL^{-1}$ threshold up to 12 hours. Detection of multiple metabolites without isCID is related to recent administration of the PVE (24 hours).

$[Glu_2HE_1\text{-}H_20+H]^+$ and $[Glu_5HE_3\text{-}H_20+H]^+$ are suggested for HES to have optimal detectability. $[Glu_3+H\text{-}H_2O]^+$can be used for semi-quantitative estimation of dextran. For the latter, thresholds of 500 $\mu g \cdot mL^{-1}$ are suggested in isCID mode only.

Since the instrument acquires full scan spectra, the detection of HES or dextran in a suspicious sample can be pre-confirmed by post-screening monitoring of other metabolites or diagnostic ions, providing even more robust analytical results.

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