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Hydrophilic interaction vs ion pair liquid chromatography for the determination of *streptomycin* and *dihydrostreptomycin* residues in milk based on mass spectrometric detection

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ABSTRACT

Streptomycin (STR) and *dihydrostreptomycin* (DHSTR) are two of the most common aminoglycoside antibiotics used in veterinary medicine. The physicochemical properties of both substances, make their determination challenging. In the present study the development of methods based on ion-pair chromatography (IPC) and on hydrophilic interaction chromatography (HILIC), for the determination of the above mentioned aminoglycosides in the range of 100–1000 $\mu g\,L^{-1}$ is described. The two methods were validated according to EU requirements for residues in food. The recoveries for the IPC method were 69.3% and 56.5% of STR and DHSTR, respectively, and for HILIC method 85.5% and 72.3%, respectively. The intra- and inter-day precision, studied at 100, 200 and 300 $\mu g\,kg^{-1}$ levels in milk samples, gave %RSD $\leq$ 13 for both methods. LOQs for the HILIC method were 14 $\mu g\,kg^{-1}$ for both analytes and for the IPC method were 109 and 31 $\mu g\,kg^{-1}$, for STR and DHSTR, respectively. The sensitivity of the HILIC method is 80 and 210 times greater than that of the ICP method, for STR and DHSTR, respectively.

1. Introduction

Since the isolation of STR from soil bacteria, in 1944, by Selman Waksman, aminoglycosides have been at the front of antibacterial drug treatment in humans. *Dihydrostreptomycin* (DHSTR) is a streptamine-based relative of STR that was discovered in 1947 [1]. In veterinary medicine *streptomycin* (STR) and DHSTR are used in the treatment of leptospirosis in cattle, sheep, goats and swine, and also for the treatment of various infections caused by Gram-negative bacteria. However, their relatively high toxicity reduces their use, as the consumption of food from animal origins, such as milk, that contains high quantities of aminoglycoside residues can be potential hazard for the humans' health. The European Union community has established maximum residue limits (MRL) for STR and DHSTR, which for the milk are set to 200 $\mu g\,kg^{-1}$ [2].

The lack of chromophore groups and the three ionisable groups in their chemical structure (Fig. 1), reveal the analytical challenge of the determination of STR and DHSTR. With the development of mass spectrometric detection, the disadvantage of the lack of chromophore groups was confronted, without the use of derivatisation. In the past few years a number of methods have been published for the determination of STR, DHSTR and other aminoglycosides in various matrices of food origins, such as milk, animal tissues, kidneys and honey based on mass spectrometric detection [3–12].

STR and DHSTR are high polar compounds. The strongest ionisable group has pK_a 13.70 (±0.50). Due to this characteristic, the retention of the aminoglycosides on the common reversed phase columns is impossible. The use of an ion-pair additive, in the mobile phase is the most common solution for the retention problem. It is well known that the addition of such agents causes a severe contamination of the column [13] and serious ion suppression, with electrospray ionization detection [14,15]. In order to avoid the use of ion-pair reagents Turnipseed et al. [3], proposed a derivatisation process to retain and separate gentamicin, tobramycin and neomycin B, on a reversed-phase column. However, the addition of a derivatisation step in the sample pretreatment leads to longer analysis time and matrix ion suppression.

Another approach to achieve of retention is the use of hydrophilic interaction chromatography (HILIC), in which a hydrophilic stationary and an aqueous–organic solvent mobile phase with high organic–solvent content are used [16]. Nguyen and Schug have reviewed the advantages of electrospray ionization in combination with HILIC mode separations [17]. This technique was successfully applied to the determination of six aminoglycosides in serum by Oertel et al. [18]. Peru et al. [19] have also developed a method for the determination of spectinomycin and lincomycin by the use of HILIC separation mode in combination with mass spectrometry. HILIC has been recently applied for the determination of impurities in STR and DHSTR in raw materials [20].

A chromatographic study using HILIC–APCI–MS for several aminoglycosides has been presented by McGrane et al. [21]. A multiresidue semiquantitative screening method for animal tissue has

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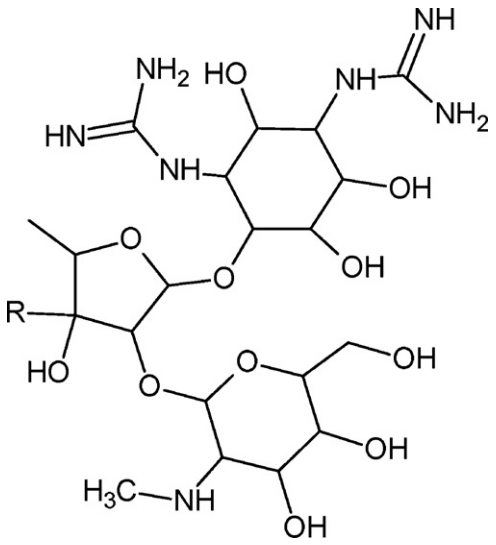

Fig. 1. Chemical structure of STR (R: CHO) and DHSTR (R: CH_2OH).

also been proposed [22] based on ZIC–HILIC column and LC–ESI-MS/MS.

To our knowledge, there is no any validated HILIC–ESI-MS method for the determination of STR and DHSTR in milk. In the present work two different methods, based on ion-pair chromatography (IPC) and HILIC, for the determination of the two aminoglycosides in milk, have been developed, validated and compared.

2. Experimental

2.1. Chemicals and reagents

Streptomycin sulfate (potency $\geq$771 IU mg^{-1}) and dihydrostreptomycin sesquisulphate (purity $\geq$98%) were purchased from Sigma–Aldrich (Steinheim, Germany). All solvents, used were HPLC-grade and obtained from Fisher Scientific (Loughbovough, UK). Water of HPLC-grade (>18 MΩ) was used. The heptafluorobutyric acid (HFBA, purity 99%) was purchased from Alfa Aesar (Lancaster, UK). The ammonium formate, formic acid and acetic acid were of LC/MS purity (Fluka, Steinheim, Germany). Trichloroacetic acid (TCA), KH_2PO_4, KOH and H_3PO_4 that used in sample pretreatment were purchased from Sigma–Aldrich (Steinheim, Germany).

2.2. Standards solutions

The stock solutions of both aminoglycosides (1000 μg mL^{-1}) were prepared in water and stored at 4 °C, for a period of less than two weeks. Standard solutions (0.1, 0.2, 0.3, 0.4 and 1.0 μg mL^{-1}) used for the preparation of the standard calibration curve and working solutions (10.1, 20.2, 30.3, 40.4 and 101 μg mL^{-1}) used for the preparation of the matrix (milk) matched standards were prepared daily, by appropriate dilutions of the stock solutions in water.

2.3. Deproteinization procedure

The deproteinization procedure of the milk samples was the same for both methods. 4 mL of milk was transferred to a 15 mL centrifuge plastic tube and 500 μL of a 30% (w/v) TCA solution was added. The mixture was vortexed for 1 min and centrifuged at 4000 rpm for 10 min. The supernatant was transferred to a new tube and the deproteinization process was repeated. After that the supernatant was quantitatively transferred to a new tube and 1 mL of 1 M KOH solution was added to adjust the pH of the extract to become approximately 7–8. After alkalization, the mixture was centrifuged at 4000 rpm for 3 min and the supernatant was quantitatively applied to SPE cartridges.

2.4. Solid phase clean-up

2.4.1. SPE for IPC method

A DSC-WCX cartridge (500 mg/3 mL, Supelco, USA) was conditioned with 2 mL of methanol followed by 2 mL of water and equilibrated with 3 mL of 20 mM KH_2PO_4 solution of pH 6.8 (adjusted with 1 M H_3PO_4). The sample supernatant was loaded on the cartridges and a washing step was performed with the addition of 2 mL of acetonitrile. The elution of the analytes was achieved by the addition of 4 mL of 300 mM HFBA solution in acetonitrile. Elute was evaporated to dryness at 50 °C under a gentle steam of nitrogen gas. The dried residue was dissolved in 4 mL of a 1% (v/v) acetic acid solution and injected on the chromatograph.

2.4.2. SPE for HILIC method

A different SPE sorbent than that used in the IPC method was used in the HILIC method. An Oasis WCX cartridge (150 mg, 6 cm^3, Waters, USA) was conditioned and equilibrated according to the manufacturer guidelines, with 3 mL of methanol followed by 3 mL of water, without any pH adjustment. The sample supernatant was loaded and allowed to pass through the column. The cartridges were washed using 2 mL of methanol and the elution was performed using 5 mL of aqueous 2% (v/v) acetic acid solution. The obtained elute was injected on the chomatograph.

2.5. LC system and conditions

A Prominence series system (Shimadzu, Germany) consisting of a LC-20 AB pump, an SIL-20AC autosampler, a DGU-20 A_3 vacuum degasser and a CTO-20 AC column oven was used for all chromatographic separations.

IPC separations were carried out using an XTerra MS C_{18} column (50 mm × 2.1 mm, 5 μm, Waters) maintained at 30 °C. The flow rate of the mobile phase was 0.3 mL min^{-1}. Mobile phase component A was 10 mM HFBA in acetonitrile and component B was 10 mM HFBA in water. A gradient elution program was started with 10% of A increased to 12% in 8 min, changed to 40% at 8.01 min, kept for 3 min at 40% returned to 10% at 11.1 min and maintained for 4 min for the column equilibration. The increase of acetonitrile after the elution of the analytes (8.01 min) was performed for column wash of matrix components that were diluted in higher content of organic solvent.

HILIC separations were performed at silica based HILIC Fortis (100 mm × 3.0 mm, 3 μm, Fortis, UK) column maintained at 40 °C. This column is a straight silica stationary phase corresponding to L3 USP column. The mobile phase was a mixture of a 150 mM $HCOONH_4$ solution in water of pH 4.5 adjusted with formic acid and acetonitrile (35%: 65%), with the flow rate being 0.4 mL min^{-1}.

The injection volume for both methods was 20 μL.

2.6. MS apparatus and parameters

A LCMS-2010 EV single quadrupole mass spectrometer (Shimadzu, Germany) equipped with an electrospray ionization (ESI) interface was used for detection and quantification of the aminoglycosides. For the IPC method the MS parameters were: ESI interface voltage 3.0 kV, curved dissolvation line (CDL) temperature 250 °C and the block heater temperature 250 °C. Nitrogen was the nebulizing gas with a flow rate of 1.5 L min^{-1}. Drying gas pressure was set at 0.1 MPa.

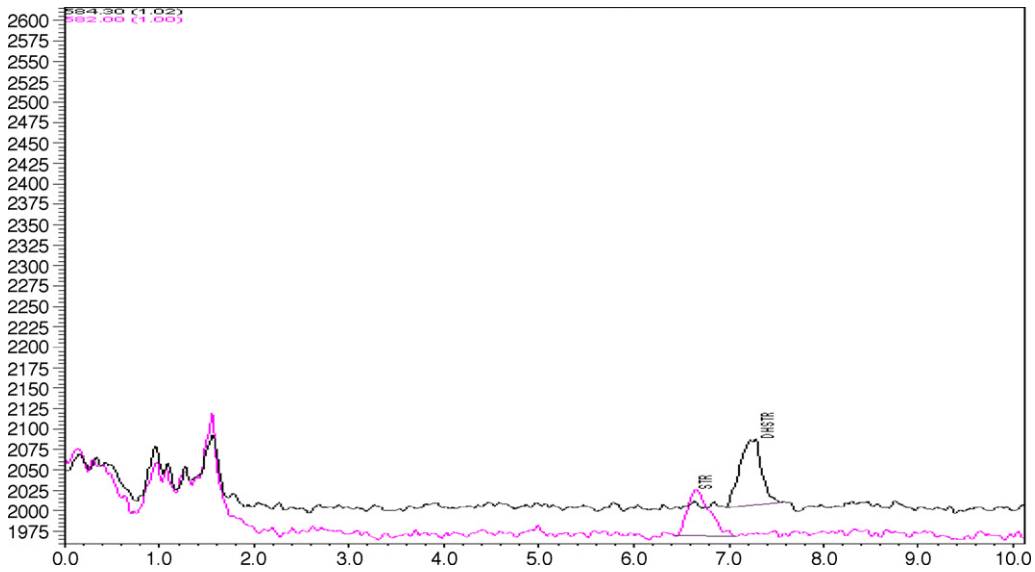

Fig. 2. Chromatogram of a spiked milk sample fortified with STR and DHSTR at half MRL (100 μg kg^{-1}) and analyzed by IPC method.

For the HILIC separations the MS parameters were: ESI interface voltage 3.5 kV, CDL temperature 300 °C and the block heater temperature 300 °C. The values of nebulizing gas flow and the drying gas pressure were the same as those in IPC method.

The quantification in both methods was carried out at single ion monitoring (SIM) in *m*/*z* 582.2 and 584.2 for STR and DHSTR, which correspond to the molecular ions, $[M-H]^+$ respectively, in positive ionization mode. For qualitative confirmation, fragmentation of the protonated ion of each analyte was obtained by in-source collision-induced dissociation (CID). The basic fragments for STR were 582.2 > 320.1, 263.1, 175.9 and for DHSTR were 584.2 > 542.4, 263.1, 175.

2.7. Validation protocol

The two methods were validated by examining the same analytical characteristics. A calibration curve of standard aqueous solutions and a matrix matched calibration curve at five fortification levels 0.5 MRL, 1.0 MRL, 1.5 MRL, 2.0 MRL and 5.0 MRL for both analytes were constructed. The spiked milk samples were prepared by adding 4.0 mL of milk to 40 μL of the appropriate standard aqueous solution. The spiked samples were treated and analyzed according to the procedure for each method.

The intra-day precision (repeatability) was determined at three fortification levels (0.5 MRL, 1.0 MRL and 1.5 MRL) by the analysis of six independent spiked samples. The inter-day precision (reproducibility) was determined for the same fortification levels at three different days.

The recovery was determined by the ratio [slope of matrix matched calibration curve]/[slope of standards calibration curve] × 100, for both IPC and HILIC method. LOQs were calculated using the standard deviation of the intercept (S_a) of the calibration curve of matrix matched standards and correspond to a signal greater than the signal of blank ten times the S_a.

3. Results and discussion

3.1. IPC method development

The first step of the sample pretreatment is the protein precipitation. Several mixtures containing an organic solvent and an acid solution or only an acid solution, usually a TCA solution, have been reported [4,7,11]. Different mixtures of acetonitrile and 30% (w/v) TCA solution were examined (addition of 10, 12, 14 and 16 mL of acetonitrile containing 250 μL or 500 μL of a 30% TCA solution to 4.0 mL of milk). The recoveries of both aminoglycosides ranged from 12% to 32%. The low recoveries are due to the low solubility of the analytes in acetonitrile and to the increased proteins solubility. Therefore, only TCA solution was used without the addition of any organic solvent, achieving sufficient recoveries (Table 2).

For further sample clean-up a solid phase extraction procedure was used. Due to the high polar nature of STR and DHSTR, different SPE sorbents of weak or strong cation exchange mechanism were tried. The Waters Oasis MCX (mixed mode-cation exchange) gave low recoveries, probably due to the strong retention of the analytes on the sorbents sulphonic groups, while the weak cation exchange mode (WCX), where the retention of the analytes is achieved by the sorbent's carboxyl groups, was found successful. Two different commercial sorbents, i.e. Supelco DSC-WCX and Waters Oasis WCX were evaluated. For the DSC-WCX it was found that the equilibrating step with 3 mL of 20 mM KH_2PO_4 (pH 6.8) solution was crucial for the high retention of the analytes. The acidic elution (2% or 5% of acetic acid in acetonitrile) gave poor chromatographic performance, with broad peak shape for both analytes. In order to eliminate this problem and have the elution solvent similar with the IP mobile phase additive, HFBA was examined at the concentrations 50, 100, 200 and 300 mM in acetonitrile. Improved peak shape was achieved, with the higher elution recovery achieved with 4 mL of 300 mM of the HFBA solution.

The performance of the Waters Oasis WCX was evaluated, at the same conditions, but without the pH control step for the sorbent. Using acetic acid in acetonitrile, the same unsymmetrical peak shape was observed, while the use of the HFBA solutions, gave irreproducible recovery results. Therefore the DSC-WCX was finally chosen.

The chromatographic behavior of the two aminoglycosides with a mobile phase of 15% of acetonitrile and 85% of an aqueous solution containing different volatile ion-pair reagents compatible with LC–MS was tested. The following aqueous solution components of the mobile phase were tested: 5% formic acid, 5% acetic acid, 10 mM HFBA, 10 mM pentafluoropropionic acid (PFPA) and 0.2% trifluoroacetic acid (TFA). No significant retention of both analytes on the C_{18} column was observed with the use of the formic acid, acetic acid and TFA. The mobile phase containing 10 mM HFBA showed longer retention time and better separation for both analytes than the mobile phase with the same concentration of PFPA.

Table 1
Comparison of the calibration curves for different concentrations of HFBA for the IPC method.

HFBA (mM)	Slope	Intercept	Relative reduction[a] (%)
(I) Streptomycin			
10	459.9	3635	–
12	189.1	2127	58.9
15	173.9	1160	62.2
HFBA (mM)	**Slope**	**Intercept**	**Relative recovery[a] (%)**
(II) Dihydrostreptomycin			
10	425.9	888	–
12	382.7	1950	10.1
15	316.3	3391	25.7

[a] In comparison with the 10 mM slope.

In order to investigate the ion suppression due to the concentration of HFBA (10, 12 and 15 mM) in the mobile phase, calibration curves of standard solutions, as described in the experimental section, were constructed. Comparing the slopes of the calibration curves (Table 1), a reduction of STR signal up to 62.2% and up to 25.7% for DHSTR, relatively to the low 10 mM concentration was observed. This phenomenon is well known and is due to the ion suppression from high concentration of HFBA and the formation of the neutral-pair. Fig. 2 shows the chromatogram of a spiked milk sample using the developed IPC method.

3.2. HILIC method development

The same deproteinization process as in the IPC method was used for the sample pretreatment. The two different weak cation exchange sorbents were also examined. As it was revealed in the IPC method, the most important step for the clean-up procedure was the elution step. Eight different solvents (2% and 5% of acetic or formic acid in methanol or in acetonitrile) were examined and DSC-WCX for all the elution solvents was found worse than the Oasis WCX, with STR and DHSTR recoveries being lower than 36% and 43%, respectively. For the Oasis WCX the best results were found with the solvent of 2% of acetic solution in acetonitrile (Table 3).

In the development of a HILIC method, the ion strength and composition of the mobile phase is of great importance. Two different volatile salts were investigated ($HCOONH_4$ and CH_3COONH_4) at concentrations ranging from 100 to 300 mM. The influence of the concentration of the $HCOONH_4$ on the retention time of the two analytes is shown in Fig. 3 and Fig. 4 shows the influence of the acetonitrile content in the mobile phase. A $HCOONH_4$ concentration of 150 mM and an acetonitrile content of 65% were selected as optimum for a sufficient retention. The results concerning the effect of mobile phase components are comparable with those obtained by the study of McGrane [21].

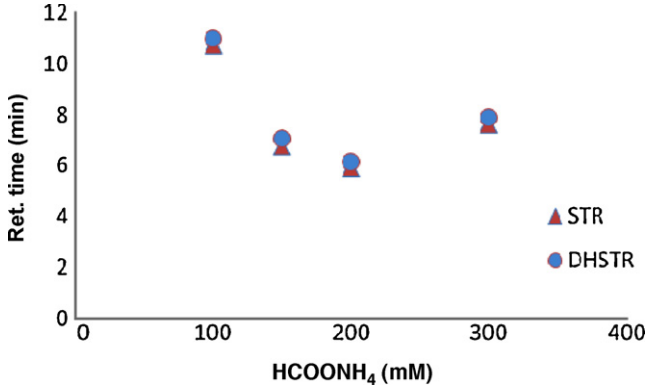

Fig. 3. Influence of $HCOONH_4$ concentration on the retention time of STR and DHSTR. Aqueous composition pH 4.5 and content 35%.

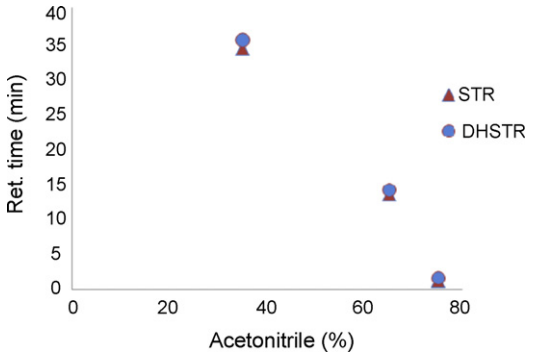

Fig. 4. Influence of the acetonitrile content on the retention time of STR and DHSTR. Aqueous component $HCOONH_4$ 150 mM, pH 4.5.

Fig. 5 shows the chromatogram of a blank milk sample and Fig. 6 the chromatogram of a spiked milk sample. As shown no interference peaks exist at the elution times of STR and DHSTR. Rather increased peak widths (~2 min) were obtained due to the strong interactions of the basic analytes with the silica column. However that does not affect the detectability and sensitivity of the method.

3.3. Validation results

The calibration curves equations of standard aqueous solutions and matrix (milk) matched standards at five fortification levels (0.5, 1.0, 1.5, 2.0 and 5.0 MRL) are presented in Tables 2 and 3, for IPC and HILIC method, respectively. As shown from the increased negative (in the case of STR) and positive (in the case of DHSTR) intercept of the calibration curves, there is no zero-crossing and a multi-point calibration should be used every day.

Comparing the slopes of the calibration curves of the aqueous standards for the two methods, it is observed that the sensitivity of the HILIC method is 80 and 210 times greater than in the ICP method (Tables 2 and 3), for STR and DHSTR, respectively, due to the

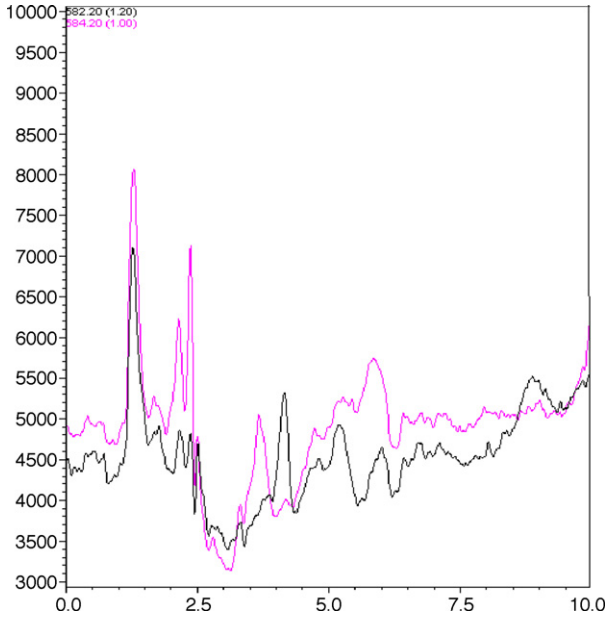

Fig. 5. Chromatogram of a blank milk sample analyzed by HILIC method.

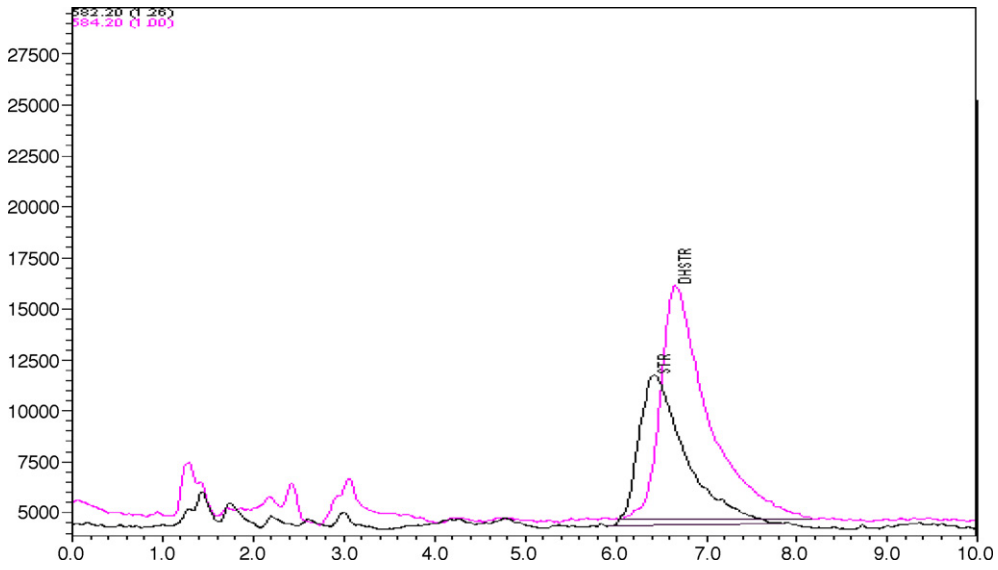

Fig. 6. Chromatogram of a spiked milk sample fortified with STR and DHSTR at half MRL (100 μg kg^{-1}) and analyzed by HILIC method.

Table 2
Calibration curves equations (100–1000 μg kg^{-1}) and relative recovery for IPC method.

	STR	DHSTR
Aqueous standards	$y = -26(\pm 86) + 265(\pm 15)C$ $r^2 = 0.998$ ($n = 5$)	$y = 9908(\pm 39) + 156(28)C$ $r^2 = 0.997$ ($n = 5$)
Matrix matched standards	$y = -20(\pm 27) + 183.91(0.48)C$ $r^2 = 0.993$ ($n = 5$)	$y = 3978(\pm 276) + 88.35(\pm 0.13)C$ $r^2 = 0.995$ ($n = 5$)
Recovery[a]	69.3%	56.5%

[a] Ratio of the slopes in matrix matched and aqueous standards.

Table 3
Calibration curves equations (100–1000 μg kg^{-1}) and relative recovery for HILIC method.

	STR	DHSTR
Aqueous standards	$y = -94.73(\pm 0.92) \times 10^4 + 21.22(\pm 0.18) \times 10^3 C$ $r^2 = 0.998 (n = 5)$	$y = -99.95(\pm 0.92) \times 10^4 + 32.84(\pm 0.19) \times 10^3 C$ $r^2 = 0.993$ ($n = 5$)
Matrix matched standards	$y = 4.12(\pm 0.25) \times 10^5 + 18.15(\pm 0.60) \times 10^3 C$ $r^2 = 0.996$ ($n = 5$)	$y = 5.6(\pm 0.33) \times 10^5 + 23.74(\pm 0.79) \times 10^3 C$ $r^2 = 0.99$ ($n = 5$)
Recovery[a]	85.5%	72.3%

[a] Ratio of the slopes in matrix matched and aqueous standards.

ion suppression phenomenon caused by the addition of the ion-pair reagent. Also the relative recovery, as determined by the ratio of the slopes of matrix matched standards and the aqueous standards, it is greater for the HILIC method, revealing that the extraction procedure for HILIC method was more efficient than that of the IPC method.

The intra- and the inter-day precision for STR and DHSTR for each method are presented in Tables 4 and 5, respectively. Both

Table 4
Precision (% RSD) for STR and DHSTR for IPC method.

Fortification level (μg kg^{-1})	STR		DHSTR	
	Intra-day ($n = 6$)	Inter-day ($n = 3 \times 6$)	Intra-day ($n = 3 \times 6$)	Inter-day ($n = 3 \times 6$)
100	6.3	12.1	5.3	13.4
200	6.7	10.2	4.8	9.7
300	5.1	8.9	5.6	10.3

Table 5
Precision (% RSD) for STR and DHSTR for HILIC method.

Fortification level (μg kg^{-1})	STR		DHSTR	
	Intra-day ($n = 6$)	Inter-day ($n = 3 \times 6$)	Intra-day ($n = 6$)	Inter-day ($n = 3 \times 6$)
100	4.6	6.7	7.5	9.8
200	5.2	8.1	9.4	11.2
300	3.8	7.4	8.1	10.9

methods showed good repeatability and reproducibility. The LOQs for the HILIC method were 13.9 and 14.0 $\mu g\,kg^{-1}$ and for the IPC method were 109 and 31 $\mu g\,kg^{-1}$, for STR and DHSTR respectively. The accuracy of the HILIC method was evaluated by analyzing spiked milk samples and using the milk matched calibration curve. Relative errors were less than 7.8% and 10.6% for STR and DHSTR respectively.

4. Conclusion

A novel HILIC method in conjunction with mass spectrometric detection for the determination of streptomycin and dihydrostreptomycin has been developed, validated and successfully applied to milk samples. The comparison of the analytical characteristics (sensitivity, precision, recovery) of the HILIC method to the ion-pair chromatographic method revealed great superiority due to severe ion suppression phenomenon when ion-pairing reagents are used.

References

[1] D.P. Arya (Ed.), Aminoglycoside Antibiotics: From Chemical Biology to Drug Discovery, John Wiley and Sons Publications, USA, 2007.
[2] EC Decision 2002/657, Off. J. Eur. Commun. L 221 (2002) 8.
[3] S.B. Turnipseed, S.B. Clark, C.M. Karbiwnyk, W.C. Andersen, K.E. Miller, M.R. Madson, J. Chromatogr. B 877 (2009) 1487.
[4] W. Zhu, J. Yang, W. Wei, Y. Liu, S. Zhang, J. Chromatogr. A 1207 (2009) 29.
[5] M. Cherlet, S. Baere, P. Backer, J. Mass Spectrom. 42 (2007) 647.
[6] S. Bogialli, R. Curini, A. Corcia, A. Laganà, M. Mele, M. Nazzari, J. Chromatogr. A 1067 (2005) 93.
[7] M. Bruijnsvoort, S.J.M. Ottink, K.M. Jonker, E. Boer, J. Chromatogr. A 1058 (2004) 137.
[8] D.N. Heller, S.B. Clark, H.F. Righter, J. Mass Spectrom. 35 (2000) 39.
[9] A. Kaufmann, K. Maden, J. AOAC Int. 88 (2005) 1118.
[10] D.N. Heller, J.O. Peggins, C.B. Nochetto, M.L. Smith, O.A. Chiesa, K. Moulton, J. Chromatogr. B 821 (2005) 22.
[11] F.L. Holthoon, M.L. Essers, P.J. Mulder, S.L. Stead, M. Caldow, H.M. Ashwin, M. Sharman, Anal. Chim. Acta 637 (2009) 135.
[12] R.H.M.M. Granja, A.M.M. Nino, R.A.M. Zucchetti, R.E.M. Nino, R. Patel, A.G. Salerno, Anal. Chim. Acta 637 (2009) 64.
[13] J.W. Dolan, LC GC Eur. 20 (2008) 258.
[14] J. Keever, R.D. Voyksner, K.L. Tyczkowska, J. Chromatogr. A 794 (1998) 57.
[15] A. Apffel, S. Fischer, G. Goldberg, P.C. Goodley, F.E. Kuhlmann, J. Chromatogr. A 712 (1995) 177.
[16] B. Dejaegher, D. Mangelings, Y.V. Heyden, J. Sep. Sci. 31 (2008) 1438.
[17] H.P. Nguyen, K.A. Schug, J. Sep. Sci. 31 (2008) 1465.
[18] R. Oertel, V. Neumeister, W. Kirch, J. Chromatogr. A 1058 (2004) 197.
[19] K.M. Peru, S.L. Kuchta, J.V. Headley, A.J. Cessna, J. Chromatogr. A 1107 (2006) 152.
[20] S. Kawano, Rapid Commun. Mass Spectrom. 23 (2009) 907.
[21] http://www.euroreside.nl/ER_IV/Contributions%20I-Z/McGrane%20765-770.pdf.
[22] P.A. Martos, F. Jayasundara, J. Dolbeer, W. Jin, L. Spilsbury, M. Mitchell, C. Varilla, B. Shurmer, J. Agric. Food Chem., publication date (web) February 3, 2010, doi:10.1021/jf903838F.