

Article

Insulin Solution Stability and Biocompatibility with Materials Used for an Implantable Insulin Delivery Device Using Reverse Phase HPLC Methods

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Abstract: Insulin (Humulin[®] R IU500) has been delivered from an implantable artificial pancreas in diabetic rats and pigs. The artificial pancreas which was implanted in the peritoneum was fabricated from several biocompatible materials such as polycarbonate, stainless steel, polyurethane, titanium and a polyurethane resin. The device also contains a glucose responsive smart gel which controls the diffusion of insulin dependent on the surrounding glucose environment. As the insulin reservoir is refillable and in contact with the device materials, assessing its biocompatibility with these various device component materials was conducted. Insulin can undergo chemical degradation mainly via a deamidation reaction on glutamine and asparagine residues rendering its biological hormone functionality. Two Reverse Phase High Performance Liquid Chromatography (RP-HPLC) methods were developed and validated for detection of insulin and degradant Asn A21 desamido insulin (method A) and insulin and degradant Asn B3 desamido insulin (method B). Material biocompatibility studies show that stainless steel and titanium are suitable for an implantable insulin delivery device design over a 31-day period. The use of polycarbonate and polyurethane could be considered if the insulin reservoir in the device was only to remain in the device for less than 11 days after which time there is a loss in cresol which acts in a protective capacity for insulin stability.

Keywords: reverse phase HPLC; recombinant human insulin; validation; desamido insulin; insulin stability; Humulin[®] R

1. Introduction

The stability of an insulin solution can be significantly reduced by a number of chemical changes to its primary structure which result in insulin derivatives with modified secondary and quaternary structures resulting in denaturation, aggregation, and precipitation. This leads to drastic reductions in the hormone's biological functionality posing serious problems for its use in drug delivery systems [1–6].

During storage and use, insulin deteriorates via two main chemical reactions: deamidation from hydrolysis and polymerisation from the formation of intermolecular covalent bonds with other insulin molecules to form higher molecular weight transformation products [6,7]. Deamidation is a reaction in which the side chain group in glutamyl or asparaginyl residues is hydrolysed to form a free carboxylic acid. The six residues in insulin which pose as potential deamidation sites are GlnA5, GlnA15, AsnA18, AsnA21, AsnB3 and GlnB4. The asparagine (Asn) residues are more prone to deamidation than glutamine (Gln) residues. Deamidation products essentially retain native activity and are not associated with adverse immunogenicity [6]. In acidic solutions (pH <2) direct hydrolysis of the side chain amide leads to deamidation at AsnA21 and forms A21 desamido insulin [8]. This

alters the charge and hydrophilic/hydrophobic properties of insulin which are the key forces controlling its tertiary structure and responsible for its biological functionality. However, in neutral solutions, deamidation primarily occurs at residue AsnB3 but at a reduced rate. Deamidation of AsnB3 and hydrolysis of the peptide bond between residues A8 and A9 contribute to formation of a mixture of iso-aspartate (isoAsp) and aspartate (Asp) derivatives (B3 desamido insulin) [9,10].

Reversed phase high performance liquid chromatography (RP-HPLC) has been widely used to develop and validate simple specific methods with short run times for the determination of insulin in biological environments [11–14]. The British Pharmacopoeia official monographs [15] highlight a HPLC method for the assay of human insulin and A21 desamido insulin utilising a gradient method with a run time of 50 min, whilst the United States Pharmacopoeia [16] describes a method for similar analysis at a longer run time of 90 min. A RP-HPLC method described by Oliva et al. [17] for the analysis and characterisation of a rapid acting insulin (Novo Nordisk®) and its degradation products in pharmaceutical preparations was able to detect A21 desamido insulin when formulations were stored at 60 °C with shaking. However, B3 degradants were detected for samples stored at 50 °C without shaking. The validation studies carried in this study were also performed using pure crystalline insulin and not the pharmaceutical preparations where A21 and B3 degradants were detected. However, as both degradation products are formed as a function of the pH environment a single HPLC method which has been able to capture both degradation products remains elusive. An isocratic HPLC method reported by Moslemi et al. [18] detected A21 desamido insulin prepared by storing human insulin in 0.01 M HCl at 40 °C for 48 h with the peak appearing close to the human insulin peak but not B3 degradants.

The aim of this work was to develop and validate two RP-HPLC methods at acidic and neutral pH for the quantification of insulin and A21 and B3 desamido insulin degradants in accordance with United States Food and Drug Administration (FDA) -approved methodology [19]. Both RP-HPLC methods were subjected to an assessment of selectivity, system suitability, linearity, accuracy, precision, sensitivity and stability for the quantification of insulin and A21 and B3 desamido insulin degradants, respectively, using Humulin® R (Hum R) and not a pure crystalline insulin as with Oliva et al. [17].

A further objective was to assess insulin stability and compatibility with materials that form part of the implantable closed-loop insulin delivery device (artificial pancreas), InSmart (Figure 1a).

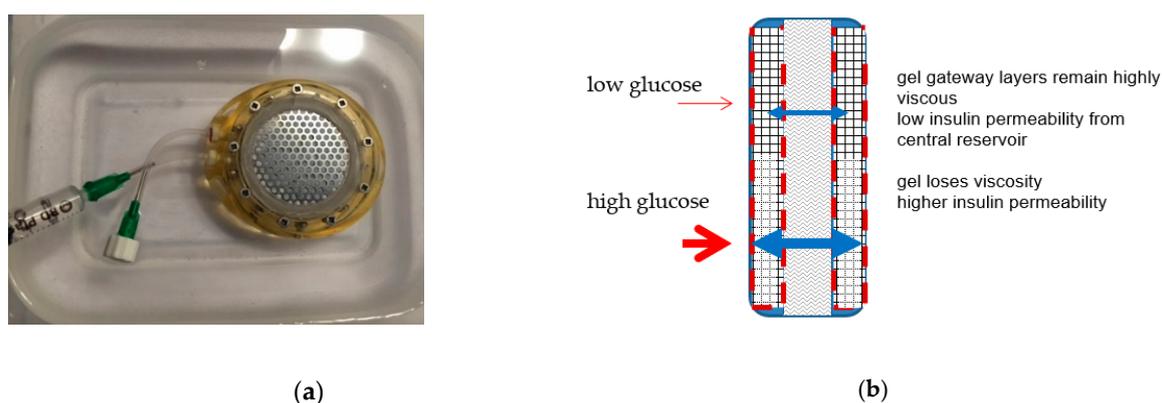


Figure 1. (a) Artificial pancreas (InSmart). (b) Mechanism for insulin delivery from artificial pancreas (InSmart).

Briefly, the InSmart device is a peritoneally implantable device under development for the delivery of insulin for type 1 insulin-dependent diabetes patients. The device is composed of a smart biopolymer which is able to undergo a viscoelastic response to glucose allowing insulin to be delivered at a rate allowing the patient to remain close to normoglycaemia (Figure 1b).

Currently the InSmart device containing smart biopolymer gel has been able to control the blood glucose of a diabetic pig for five weeks during which the diabetic pig was subjected to several high

concentration glucose challenges which allowed a faster output of Hum R to bring the blood glucose of the diabetic pig to near normoglycaemia [20]. Hum R is five times more potent than other insulin formulations which are typically 100 IU. Its use in this device allowed for a high concentration depot so that the device would not need refilling on a more frequent basis. The smart biopolymer and InSmart rigid device are composed of parts made from polycarbonate (a thermoplastic polymer containing monomer bisphenol A) and stainless steel (Grade 316), polyurethane (a long chain thermoplastic polymer made up of polyol and isocyanate mixed), titanium (99.99% purity) and resin (a polyurethane two-part thermosetting resin made of polyol and isocyanate mixed) which are all potential materials of interest in future device designs. The material compatibility study was therefore conducted with Hum R which it was reasoned would give valuable information with regard to selection of material for future design as it is the insulin which would be utilised in such a device.

2. Experimental

2.1. Chemicals and Materials

Humulin[®] R (Hum R) 500 IU/mL manufactured by Eli Lilly and Co was used. It has a composition of 500 IU/mL insulin, 2.5 mg/mL meta-cresol and 16 mg/mL glycerine, as well as zinc oxide to supplement the endogenous zinc to obtain a total zinc content of 0.017 mg/100 units, and water for injection. The pH (reported as between 7.0 and 7.8) was adjusted to 7.4. Diluting fluid manufactured by Novo Nordisk was used. HPLC grade acetonitrile, sodium dihydrogen phosphate, disodium hydrogen phosphate, ortho-phosphoric acid and ethanolamine were purchased from Fischer Chemicals (Loughborough, UK). Nylon membrane filters, 0.45 μm , 47 mm were purchased from Whatman International Ltd. (UK). Distilled water was used throughout.

Clear 1.8 mL vials (E-C sample Wheaton 33 low extractable borosilicate glass 12 \times 35 mm dia. \times ht (cap on) 8–425 screw cap size) with septum top, rubber lined black cap (Fisher Scientific, Loughborough, UK) was used. Machined material samples of polycarbonate (a thermoplastic polymer containing monomer bisphenol A) (PC), polyurethane (a long chain thermoplastic polymer made up of polyol and isocyanate mixed) (PU), resin (a polyurethane two part thermosetting resin made of polyol and isocyanate mixed) (R) and stainless steel (Grade 316) (SS) were acquired from Renfrew Group (Renfrew Group International, Leicester, UK). These were rounded rods with a diameter of 2.8 ± 0.2 mm and height of 44 ± 1 mm with polish and finish matching those on the device. Titanium wire (Ti) manufactured by Sigma Aldrich (Saint Louis, MO, USA) was 0.5 mm diameter, 99.99% and 50 cm wire was used. These were cut to size to fit the sample bottles.

2.2. Preparation of Humulin R Standards

Hum R standard solutions from 50 to 500 IU/mL (50, 100, 150, 200, 250, 300, 350, 400, 450, 500 IU/mL) were prepared by dilution of the standard stock solution with diluting fluid. A calibration graph was constructed in the range of 50 to 500 IU/mL Hum R ($n = 6$).

2.3. Material Compatibility Experimental Set-Up and Method

Rods of the differing materials were secured into sample bottles by inserting through the septum present in the lids. Then, 1 mL of Hum R was placed in 2 mL test sample bottles each with PC, PU, R, SS rod and Ti wire. In addition, Hum R was placed in sample bottles without test materials as control standards. The test sample bottles were stored at 20 $^{\circ}\text{C}$ and 37 $^{\circ}\text{C}$. Three sets of sample bottles with each material rod (3 \times 10 sample bottles) were set up to provide for the two RP-HPLC methods. Sampling was performed after 0.083 (2 h), 1, 3, 7, 11, 21 and 31 days and analysed by RP-HPLC. For the Hum R samples, RP-HPLC acidic analytical method A was used in order to determine the amount of insulin remaining in the sample and to assess the amount of A21 desamido insulin degradant. The RP-HPLC neutral analytical method B was used to determine the amount of insulin remaining in the sample and to assess the amount of B3 desamido insulin degradant. In addition, the recovery

of meta-cresol (m-cresol) in the Hum R samples was determined. Insulin and m-cresol recovery was determined by comparison with control samples stored at 20 °C without the test material rods. A calibration curve using the control samples was generated at each time point.

2.4. Methods RP-HPLC Chromatographic System

Chromatographic analyses were performed using a Shimadzu Prominence HPLC system consisting of an in-line DGU-20AS Prominence degasser, LC-20AD Prominence quaternary pump, SIL-20A Prominence auto sampler, CTO-20AC Prominence column oven and SPD-M20A Prominence diode array detector.

2.5. Analytical Method A for Determination of Insulin and A21 Desamido Insulin Degradant

A Jupiter 5 μ C18 300 Å, 250 \times 4.6 mm column (Phenomenex, Cheshire, UK) was used for separation preceded by a 0.5 mm in-line filter and a widepore C18 4 \times 3 mm guard column. For detection of Hum R and Asn A21 desamido insulin degradant an acidic mobile phase was used, consisting of 74:26 (v/v) aqueous sodium sulphate pH 2.3 (adjusted with ethanolamine):acetonitrile. The buffer was filtered through a nylon membrane of 0.45 μ m pore size. Elution was isocratic with a flow rate of 1.5 mL/min, a column temperature of 40 °C and sample injection of 20 μ L. The detector was set to scan from 190 to 400 nm and had a channel set at 215 nm to detect Hum R and its degradation products.

2.6. Analytical Method B for the Determination of Insulin and B3 Desamido Insulin Degradant

All conditions used for method A were replicated except the mobile phase which consisted of 73:27 (v/v) 0.1 M sodium phosphate buffer pH 7.4:acetonitrile.

2.7. Validation Procedures

Table 1 shows the qualification parameters assessed for methods A and B.

Table 1. Validation procedures.

Parameter	Sample	Criteria
Selectivity/specificity	Samples of diluting fluid containing the same excipients as Humulin® R (Hum R), except human insulin	There should be no peaks present in diluting fluid interfering with the peak area response from Hum R and desamido insulin degradants
System suitability	$n = 6$ samples analysed at a selected concentration	An acceptance criterion of $\pm 2\%$ for percent relative standard deviation (%RSD) for the peak areas and retention times (Rt) for Hum R
Linearity	Minimum 5-point curve generated over range (50–500 IU/mL). Six replicate experiments run	$R^2 \geq 0.990$
Accuracy	$n = 4$ samples ($\times 3$) analysed at different concentrations	Determined concentration at each level must be 97.0%–103.0% of theoretical concentration
Precision/repeatability intra-day	$n = 4$ samples ($\times 3$) analysed at different concentrations	%RSD at each level $\leq 3.0\%$
Inter-day	$n = 4$ samples ($\times 3$) analysed at different concentrations	%RSD at each level $\leq 3.0\%$
LOD and LOQ	Regression data from linearity studies used LOD = $3.3 \frac{\mu\text{g}}{\text{mL}}$; LOQ = $10 \frac{\mu\text{g}}{\text{mL}}$	Report LOD and LOQ results
Sample stability	Sample stability to be assessed over a 31-day period	Sample preparations must be within 98.0%–102.0% of the initial concentration upon reanalysis

3. Results and Discussion

3.1. Selectivity/Specificity

The developed RP-HPLC analytical method A under chromatographic conditions achieved a run time of 30 min with elution of Hum R insulin at retention time (Rt) 13.3 min, m-cresol at Rt 5.6 min and degradation product A21 desamido insulin at Rt 24.1 min.

For RP-HPLC analytical method B a similar run time of 30 min was used and elution of Hum R insulin had a Rt of 16.6 min, m-cresol at Rt 5.4 min and degradation product B3 desamido insulin at Rt 12.6 min and 13.4 min. Comparison of chromatograms of placebo and Hum R for both methods revealed no additional peaks co-eluting with the peaks of Hum R and its degradation products.

3.2. System Suitability

System suitability for method A with 200 IU/mL Hum R showed a Rt of 13.3 min for A21 with a %RSD = 0.41 ($n = 6$). For method B with 250 IU/mL Hum R a Rt of 16.6 min with a %RSD of 1.49 ($n = 6$) was observed. %RSD for peak areas were found to be 0.41 and 0.45, respectively. Both methods were within the $\pm 2\%$ acceptance criteria indicating suitability of the corresponding system. The capacity factor and resolution were both >2 and the tailing factor <2 which was within accepted the criteria.

3.3. Linearity

A linear relationship between AUP and insulin concentration over the concentration range 50–500 IU/mL for method A (slope $387,831 \pm 8986$; intercept $2,089,296 \pm 1,506,488$; $R^2 = 0.9966$) and for method B (slope $379,847 \pm 1620$; intercept $4,513,995 \pm 423,999$; $R^2 = 0.9956$) was observed.

3.4. Accuracy

Accuracy was studied using two different sets of four solutions of concentration 150, 200, 250 and 500 IU/mL Hum R for method A and 150, 200, 300 and 400 IU/mL Hum R for method B. The recovery was $100.48 \pm 0.63\%$ and $101.27 \pm 1.46\%$ for Hum R for methods A and B using corresponding AUPs to insulin concentrations.

3.5. Precision

Table 2 shows precision data for Hum R samples during intra- and inter-day runs for methods A and B. The %RSD for intra-day precision was ≤ 1.01 and inter-day precision was ≤ 0.75 for method A; the intra-day precision was ≤ 0.74 and inter-day precision was ≤ 0.30 for method B. The two methods showed suitable repeatability and intermediate precision.

Table 2. Precision for determination of insulin in Hum R for methods A and B.

Method A					
Std conc.	Intra-Day		Inter-Day		Overall RSD
IU/mL	AUP Mean \pm SD	%RSD	AUP Mean \pm SD	%RSD	%
200	80,511,102 \pm 8,10,141	1.01	81,130,312 \pm 321,212	0.40	0.80
300	114,116,991 \pm 502,655	0.44	114,648,789 \pm 310,602	0.27	0.41
500	197,502,242 \pm 1,337,885	0.68	197,061,936 \pm 1,478,956	0.75	0.65
Method B					
100	45,340,751 \pm 79,090	0.17	44,156,436 \pm 133,078	0.30	1.47
200	82,944,396 \pm 184,091	0.22	86,743,903 \pm 37,292	0.04	2.46
400	156,927,295 \pm 1,166,927	0.74	162,635,381 \pm 141,610	0.09	2.01

3.6. Detection and Quantitation Limits (Sensitivity)

LOD and LOQ were calculated based on the standard deviation from the AUP for the lowest concentration used in linearity studies as well as the slope of the calibration curve. The LOD and LOQ for Hum R for method A was 0.79 and 2.41 IU/mL, respectively, and for method B was 0.98 and 2.97 IU/mL, respectively. The results thus demonstrate the sensitivity of the two methods.

3.7. Hum R Stability

Table 3 shows that insulin stability determined by comparing Hum R 400 IU and 500 IU at 20 °C over 31 days was within the criteria highlighted in Table 1 and that in the absence of any materials was stable.

Table 3. Stability data for Hum R insulin samples stored at 20 °C for 31 days using methods A and B.

Time After Preparation (Days)	Method A			
	Hum R 500 IU/mL		Hum R 400 IU/mL	
	Concentration (IU/mL)	Concentration %	Concentration (IU/mL)	Concentration %
Fresh	494.25	100.00	401.16	100.00
1	491.87	99.52	400.88	99.93
3	492.93	99.73	401.10	99.99
7	492.64	99.67	400.15	99.75
11	491.63	99.47	400.77	99.90
31	497.35	100.63	398.16	99.25
	Method B			
Fresh	500.87	100.00	400.58	100.00
1	499.09	99.64	399.63	99.76
3	499.62	99.75	400.61	100.01
7	502.49	100.32	399.66	99.77
11	502.59	100.35	403.72	100.78
31	501.81	100.19	399.18	99.65

3.8. Effect on Insulin Recovery over Time

The Hum R insulin recovery values were normalised over the entire time period based on the values observed from the initial time point. The Hum R recovery results for solutions in contact with PC, R, SS and Ti show negligible loss of Hum R after 31 days at 20 °C (Figure 2a). However, PU showed a loss of ~74% after 31 days at 20 °C. At 37 °C, solutions in contact with Ti showed negligible loss of Hum R, with SS showing a loss of ~7%, R showing ~16%, PC showing ~67% and PU showing complete loss of Hum R after 31 days (Figure 2b).

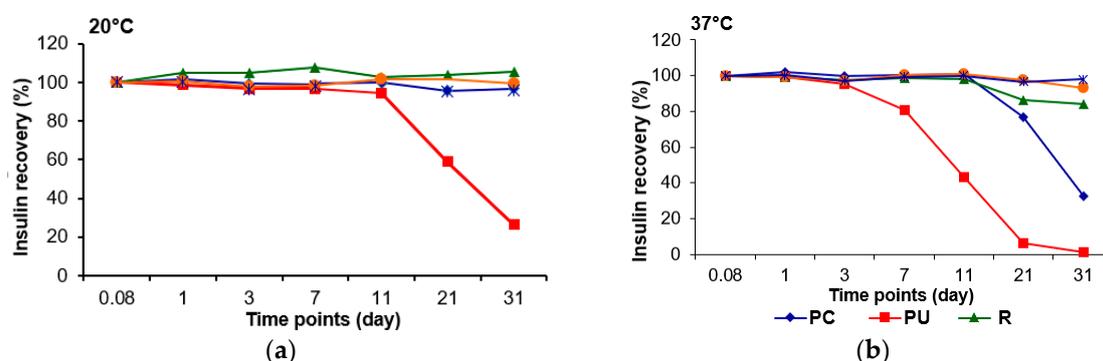


Figure 2. (a) Hum R insulin recovery over time for each test material at 20 °C and (b) 37 °C for method A. PC: polycarbonate; PU: polyurethane; R: resin.

Figure 3a shows solutions in contact with PC and Ti had a negligible loss of Hum R after 31 days at 20 °C. However, R showed a loss of ~25%, SS showed a loss of ~15% and PU showed a loss of ~82% after 31 days at 20 °C. At 37 °C, solutions in contact with PC and Ti showed negligible loss of

Hum R, with SS and R showing a loss of ~22% and PU showing complete loss of Hum R after 31 days (Figure 3b). The insulin recovery results suggest that at 20 °C Hum R solution is compatible with PC and Ti for up to 31 days and with R and SS up to 21 days. At 37 °C Hum R solution is compatible with Ti for up to 31 days, with SS for up to 21 days and PC and R up to 11 days. The results indicate that PU looks like the least compatible of all the materials.

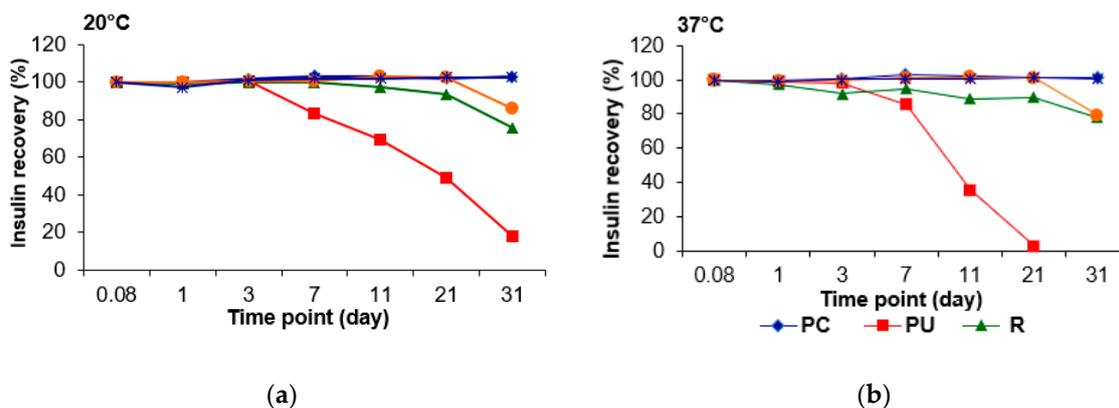


Figure 3. (a) Hum R insulin recovery over time for each test material at 20 °C and (b) 37 °C for method B.

3.9. Formation of A21 and B3 Desamido Insulin over Time

3.9.1. A21 Desamido Insulin

Low levels of A21 desamido insulin were observed over time for all materials at 20 °C and 37 °C (Table 4); the maximum values observed were well below Pharmacopoeia [15] specified limits for insulin preparations of not more than 5% of total area of peaks. The maximum levels observed over 31 days at 20 °C was with R (0.34%) and the maximum levels observed at 37 °C was with SS and Ti (0.49%).

Table 4. A21 desamido insulin formed over time at 20 °C and 37 °C as a percentage of the total insulin peak area for method A.

Time (days)	SS 20 °C	SS 37 °C	Ti 20 °C	Ti 37 °C	PU 20 °C	PU 37 °C	PC 20 °C	PC 37 °C	R 20 °C	R 37 °C
0.08	0.11	0.12	0.11	0.12	0.11	0.12	0.12	0.12	0.11	0.12
1	0.09	0.10	0.09	0.10	0.09	0.11	0.09	0.10	0.11	0.13
3	0.09	0.12	0.09	0.11	0.13	0.19	0.09	0.12	0.13	0.19
7	0.11	0.17	0.10	0.11	0.16	0.05	0.06	0.04	0.02	0.05
11	0.10	0.19	0.07	0.20	0.15	0.09	0.10	0.20	0.06	0.21
21	0.11	0.32	0.12	0.33	0.04	0.04	0.13	0.25	0.10	0.22
31	0.16	0.45	0.14	0.47	0.10	0.00	0.18	0.00	0.34	0.30

3.9.2. B3 Desamido Insulin

The levels of B3 desamido insulin formed are as presented in Table 5. Formation of B3 desamido insulin is largely unaffected by any of the materials at 20 °C and 37 °C except for R, which shows an increase after three days. Polycarbonate seems to contribute to the formation of B3 desamido insulin after 21 days at 37 °C. For all materials, except R (maximum level was 7.5%), the B3 desamido insulin values observed were below the Pharmacopoeia [15] specified limit which states that all other peaks (other than A21 and insulin peak) should not be more than 6%.

Table 5. B3 desamido insulin (iso-aspartate (isoAsp) and aspartate (Asp)) formed over time at 20 °C and 37 °C as a percentage of the total insulin peak area for method B.

Time (days)	SS 20 °C	SS 37 °C	Ti 20 °C	Ti 37 °C	PU 20 °C	PU 37 °C	PC 20 °C	PC 37 °C	R 20 °C	R 37 °C
0.08	2.30	2.37	2.34	2.33	2.33	2.34	2.33	2.35	1.91	2.39
1	2.04	2.10	2.01	2.08	2.01	2.05	2.12	2.03	2.06	2.08
3	2.08	2.18	2.19	2.25	2.07	2.15	2.16	2.07	2.14	2.34
7	2.13	2.42	2.28	2.43	1.86	2.36	2.27	2.49	2.42	3.07
11	1.70	2.15	1.54	1.50	1.32	1.06	1.69	1.94	2.17	4.26
21	1.58	1.99	1.61	1.99	1.41	0.09	1.58	0.09	4.56	7.01
31	1.39	2.11	1.55	2.11	0.75	0.31	1.53	5.29	2.67	7.43

3.10. Effect on *m*-Cresol Recovery over Time

The *m*-cresol recovery values were normalised over the entire time period based on values observed at initial time point. The *m*-cresol recovery levels from the Hum R solutions with the test materials over 31 days at 20 °C and 37 °C are presented in Figure 4a,b.

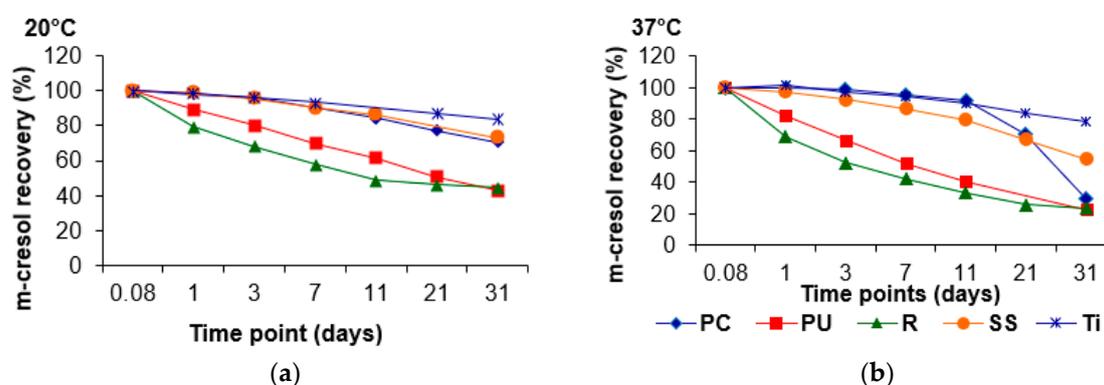


Figure 4. (a) *m*-Cresol recovery over time for each test material at 20 °C and (b) 37 °C using analytical method A.

Hum R solutions in contact with all five materials show loss of *m*-cresol over the 31-day test period at 20 °C and 37 °C. At 20 °C, solutions with Ti showed the least loss of *m*-cresol, with PU and R showing a loss of ~56% and SS and PC showing a loss of ~28% after 31 days. At 37 °C, solutions with Ti showed the least loss of *m*-cresol, with PC, PU and R showing a loss of ~74% and SS showing a loss of ~45% after 31 days. Comparison of the Hum R and *m*-cresol recovery trends reveal that recovery of *m*-cresol decreases with increase in loss of Hum R at 20 °C and 37 °C.

Combining all findings from Hum R recovery, *m*-cresol recovery, formation of A21 and B3 desamido insulin the following conclusions can be made for the materials tested:

PC: Hum R was found to be compatible with PC at 20 °C with negligible loss of insulin and very low levels of desamido insulin degradants formed though there was some loss of *m*-cresol observed. At 37 °C there appears to be insulin loss after 11 days (using method A) which was proportional to *m*-cresol loss observed after 11 days, here the insulin loss can be linked to the loss of protection of the insulin molecule by *m*-cresol and not the formation of degradants.

SS and Ti: At 20 °C and 37 °C, the Hum R solutions were compatible over 31 days with low levels of desamido insulin degradants observed and some loss in insulin observed for SS at 37 °C after 21 days can be linked to loss in *m*-cresol observed.

PU: Hum R loss from 11 days at 20 °C and 3 days at 37 °C. This loss in insulin was due to loss of protection by *m*-cresol observed and not due to formation of the desamido insulin degradants as very low levels of these were observed.

R: Hum R loss was less than compared to other materials. The *m*-cresol loss is linked to higher levels of B3 (>7%) formed.

4. Conclusions

- The validation capabilities, selectivity, system suitability, linearity, range, accuracy, precision, detection and quantitation limits determined for the two RP-HPLC methods were assessed based on the FDA guidelines. The RP-HPLC acidic and neutral analytical methods can be used to detect and quantify insulin and two of its main deamidation degradants.
- The validation studies have demonstrated the suitability of the two methods for the determination of insulin and its degradants with reduced analysis times and with accuracy across the concentration ranges intended to be studied for the InSmart device material compatibility studies.
- The objective of the compatibility studies was to investigate insulin stability and compatibility with materials that formed part of an implantable insulin delivery device or have a potential to be considered for use in future device designs. The main conclusions from the material compatibility studies show that SS and Ti are suitable for device design. The use of PC and PU could also be considered if the insulin reservoir in the device was only to remain in the device for less than 11 days after which time there is loss in cresol which acts as in a protecting capacity for insulin stability. As the insulin reservoir in the device would be refillable these materials could also be considered suitably biocompatible candidates for device incorporation. Resin was found to be wholly unsuitable a device material.

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