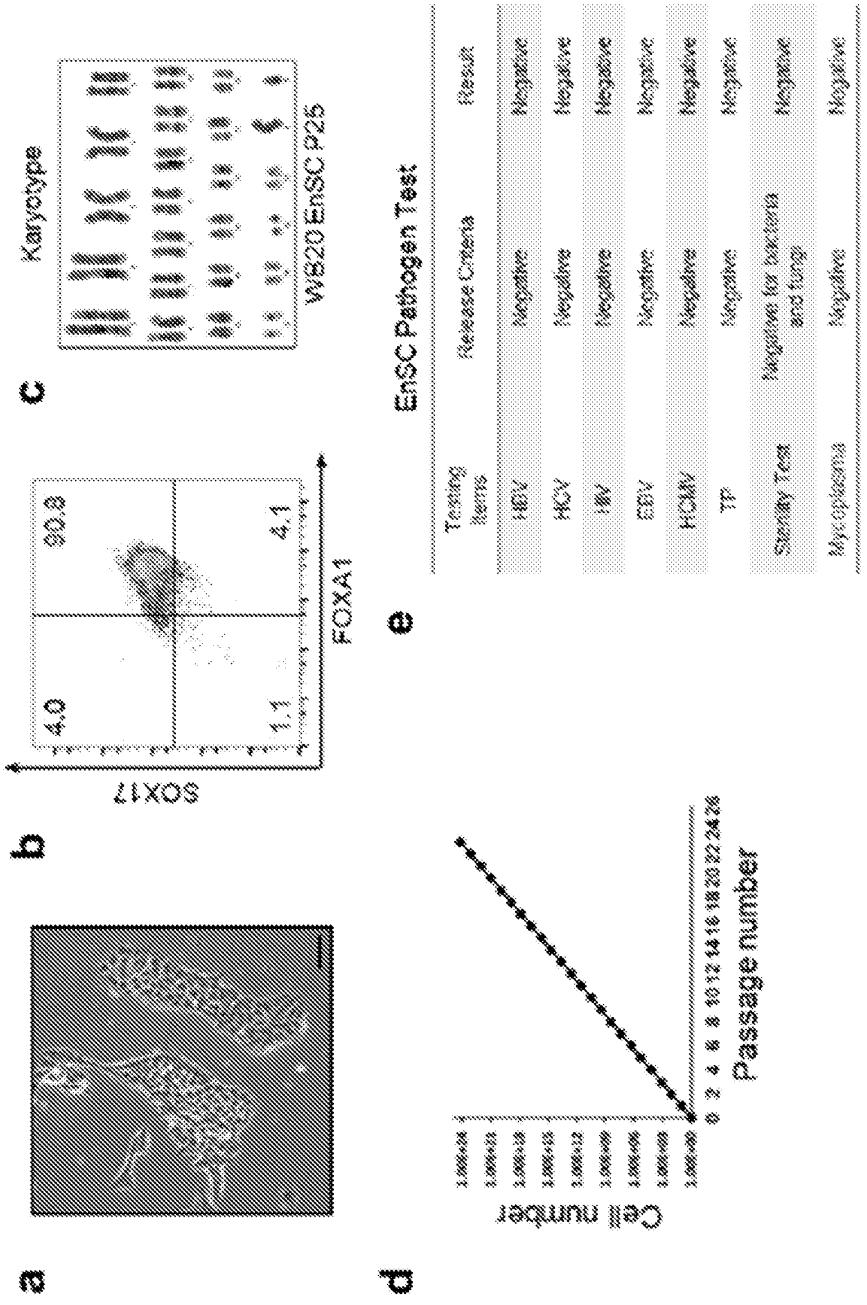

a
b
SOX17
FOXA1
4.0
90.8
1.1
4.1
c
Karyotype
WB20 EnSC P25
d
Cell number
Passage number
0 2 4 6 8 10 12 14 16 18 20 22 24 26
e
EnSC Pathogen Test
Testing Items
Release Criteria
Result
HBV
Negative
Negative
HCV
Negative
Negative
HIV
Negative
Negative
EBV
Negative
Negative
HCMV
Negative
Negative
TP
Negative
Negative
Sterility Test
Negative for bacteria and fungi
Negative
Mycoplasma
Negative
Negative

FIG. 1

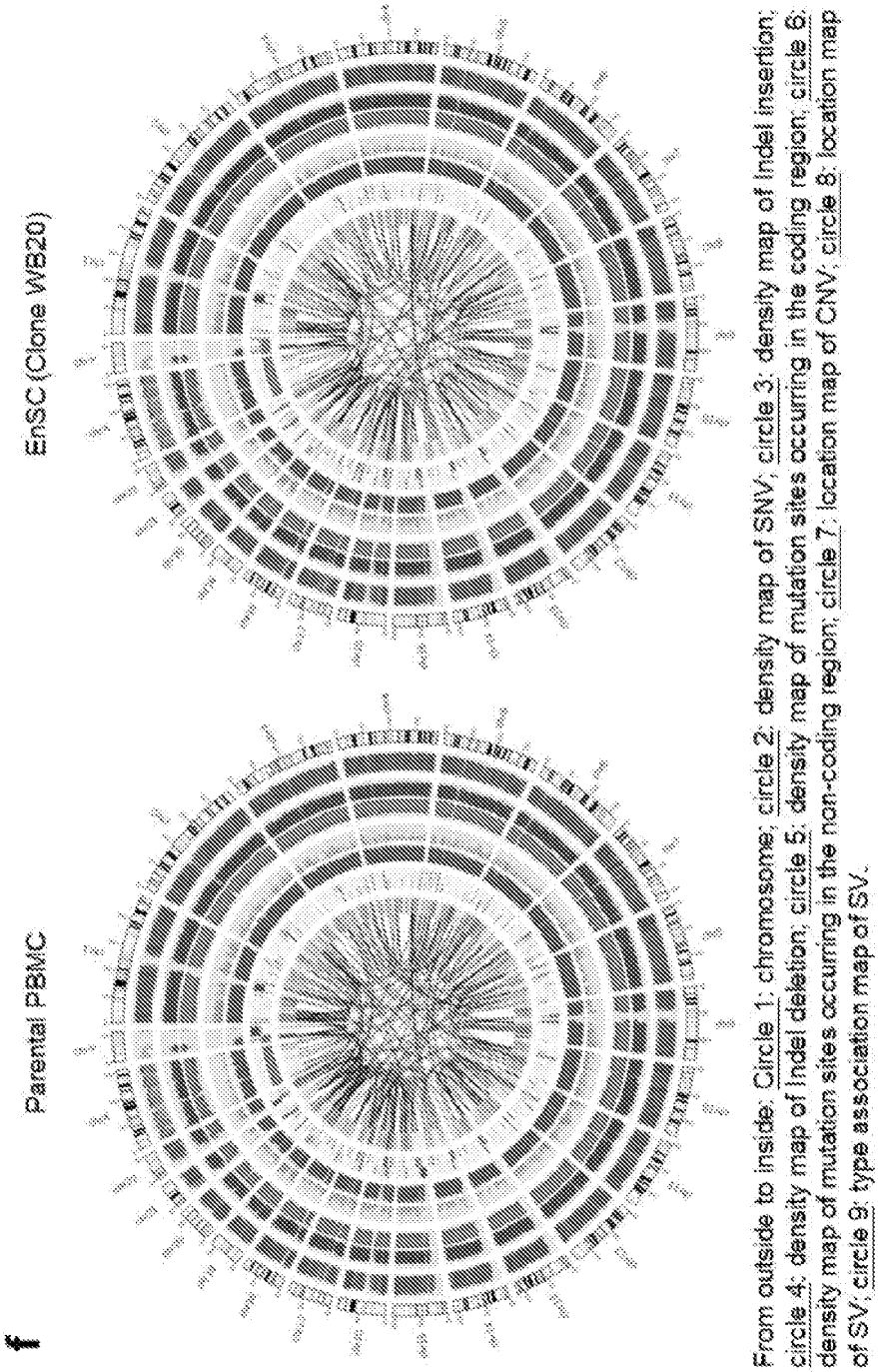


FIG. 1 (continued)

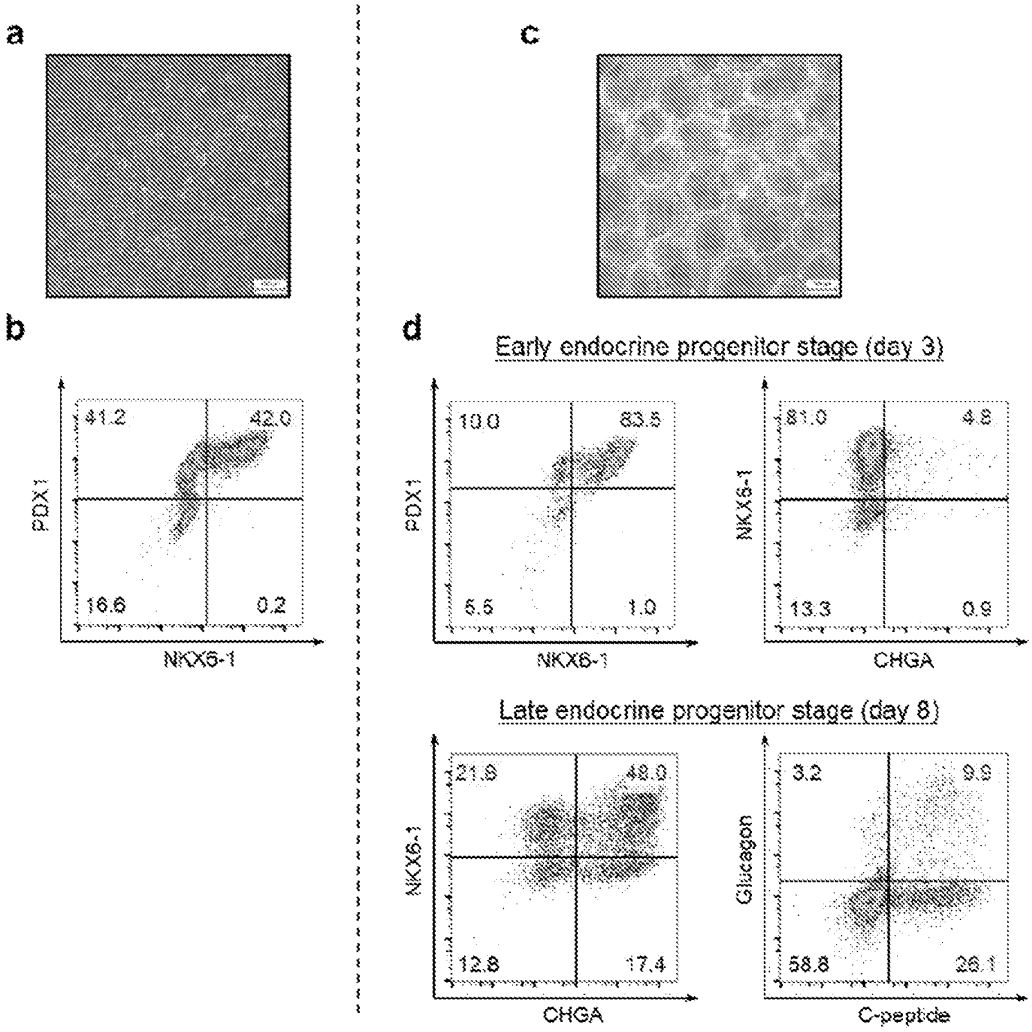

a
c
b
d
Early endocrine progenitor stage (day 3)
41.2
42.0
16.6
0.2
PDX1
NKX6-1
10.0
83.5
5.5
1.0
81.0
4.8
13.3
0.9
CHGA
Late endocrine progenitor stage (day 8)
21.8
48.0
12.8
17.4
3.2
9.9
58.8
28.1
C-peptide
Glucagon

FIG. 2

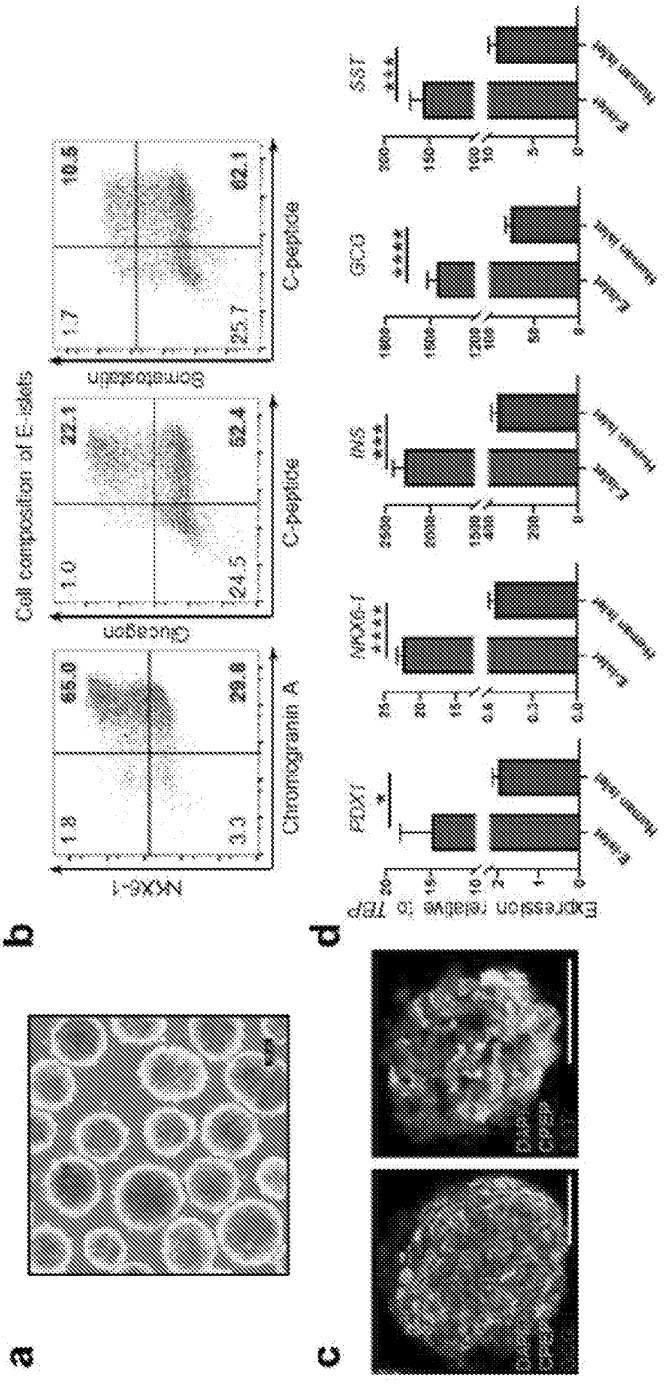

a
b
c
d
Cell composition of E-islets
NKX6-1
Chromogranin A
Glucagon
C-peptide
Somatostatin
C-peptide
65.0
29.8
1.8
3.3
22.1
62.4
1.0
24.5
16.5
62.1
1.7
25.7
PDX1
NKX6-1
INS
GCG
SST
Expression relative to TBP
Human islet
Exvivo
Human islet
Exvivo

FIG. 3

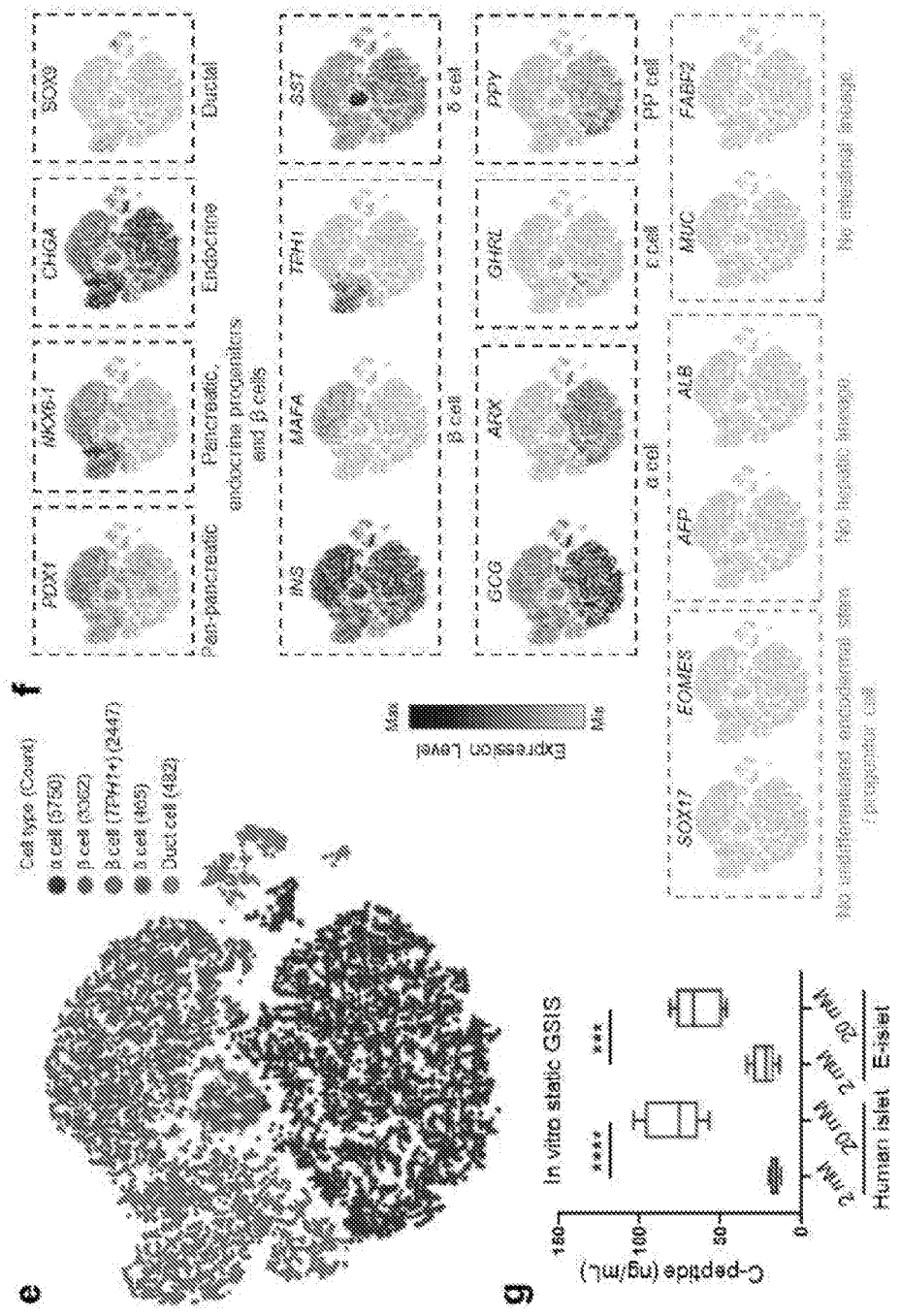
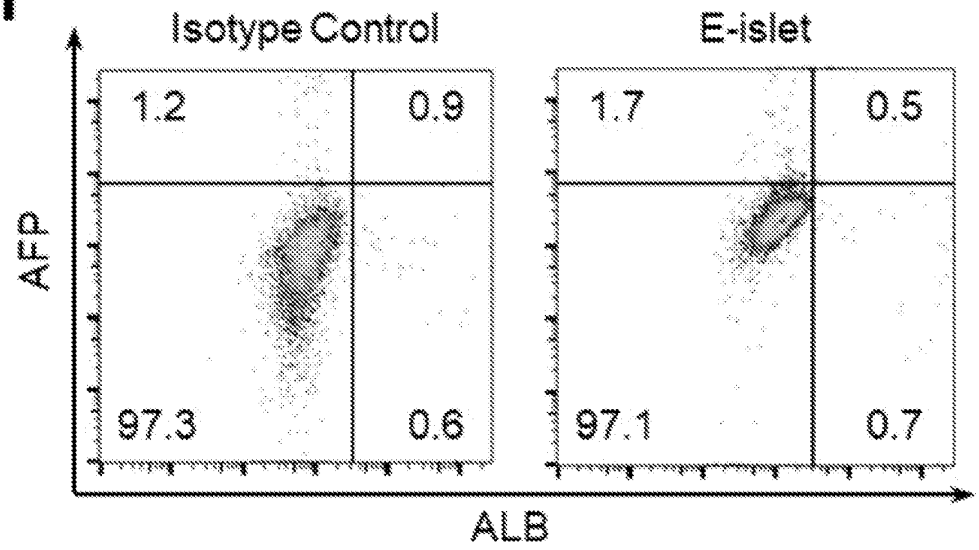


FIG. 3 (continued)

h



i

E-islet Pathogen Test

Testing Items	Release Criteria	Result
HBV	Negative	Negative
HCV	Negative	Negative
HIV	Negative	Negative
EBV	Negative	Negative
HCMV	Negative	Negative
TP	Negative	Negative
Sterility Test	Negative for bacteria and fungi	Negative
Mycoplasma	Negative	Negative
Endotoxin	< 1 EU/mL	< 0.01 EU/mL

FIG. 3 (continued)

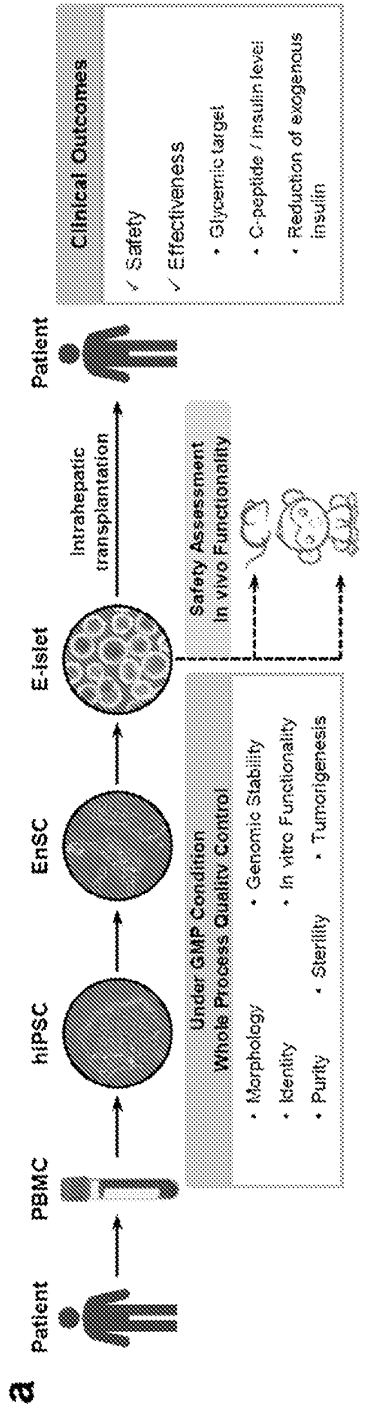

a
Patient
PBMC
hiPSC
EnSC
E-islet
Intrahepatic transplantation
Patient
Clinical Outcomes
✓ Safety
✓ Effectiveness
• Glycemic target
• C-peptide / insulin level
• Reduction of exogenous insulin
Under GMP Condition
Whole Process Quality Control
• Morphology
• Identity
• Purity
• Sterility
• Genomic Stability
• In vitro Functionality
• Tumorigenesis
Safety Assessment
In vivo Functionality

FIG. 4

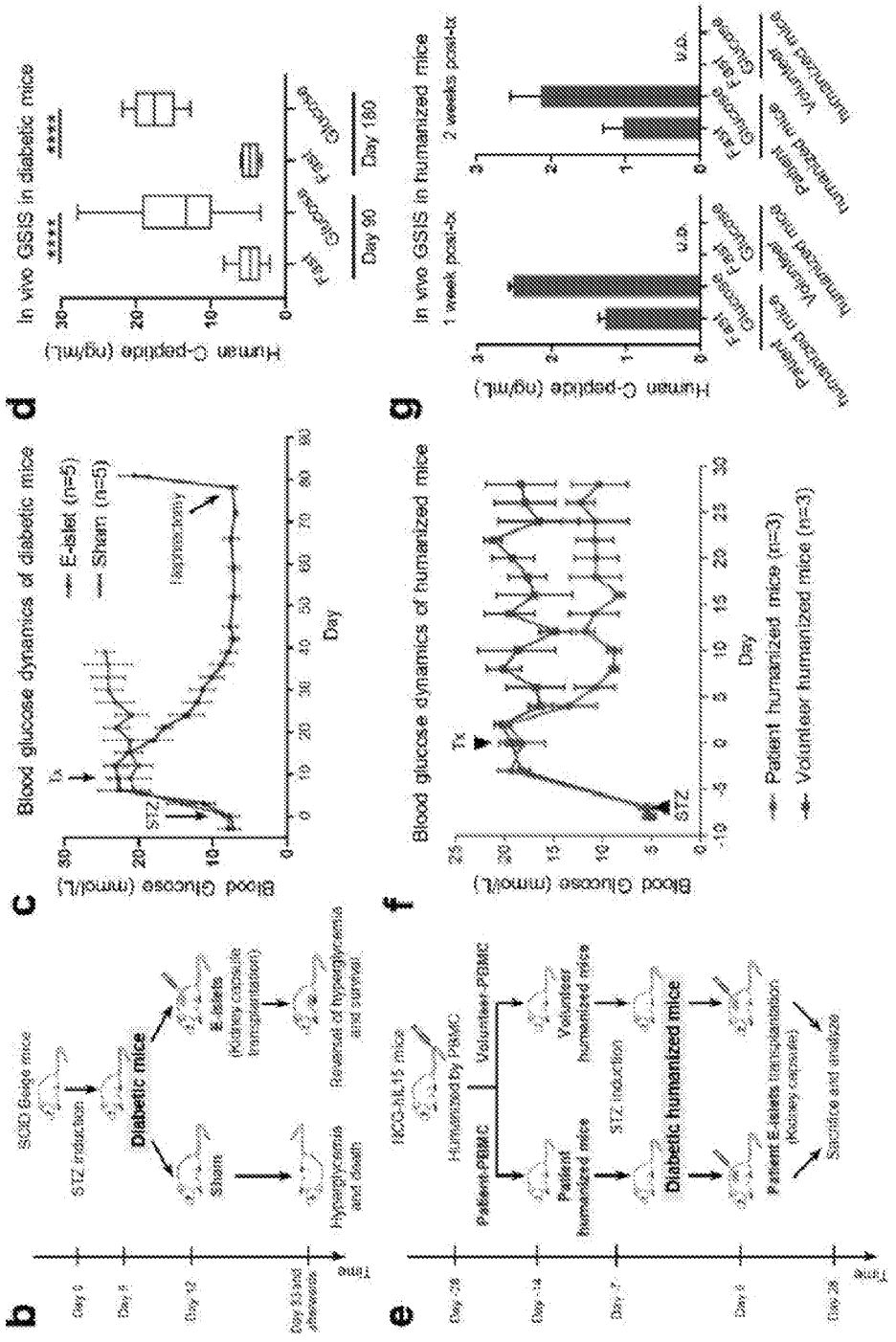


FIG. 4 (continued)

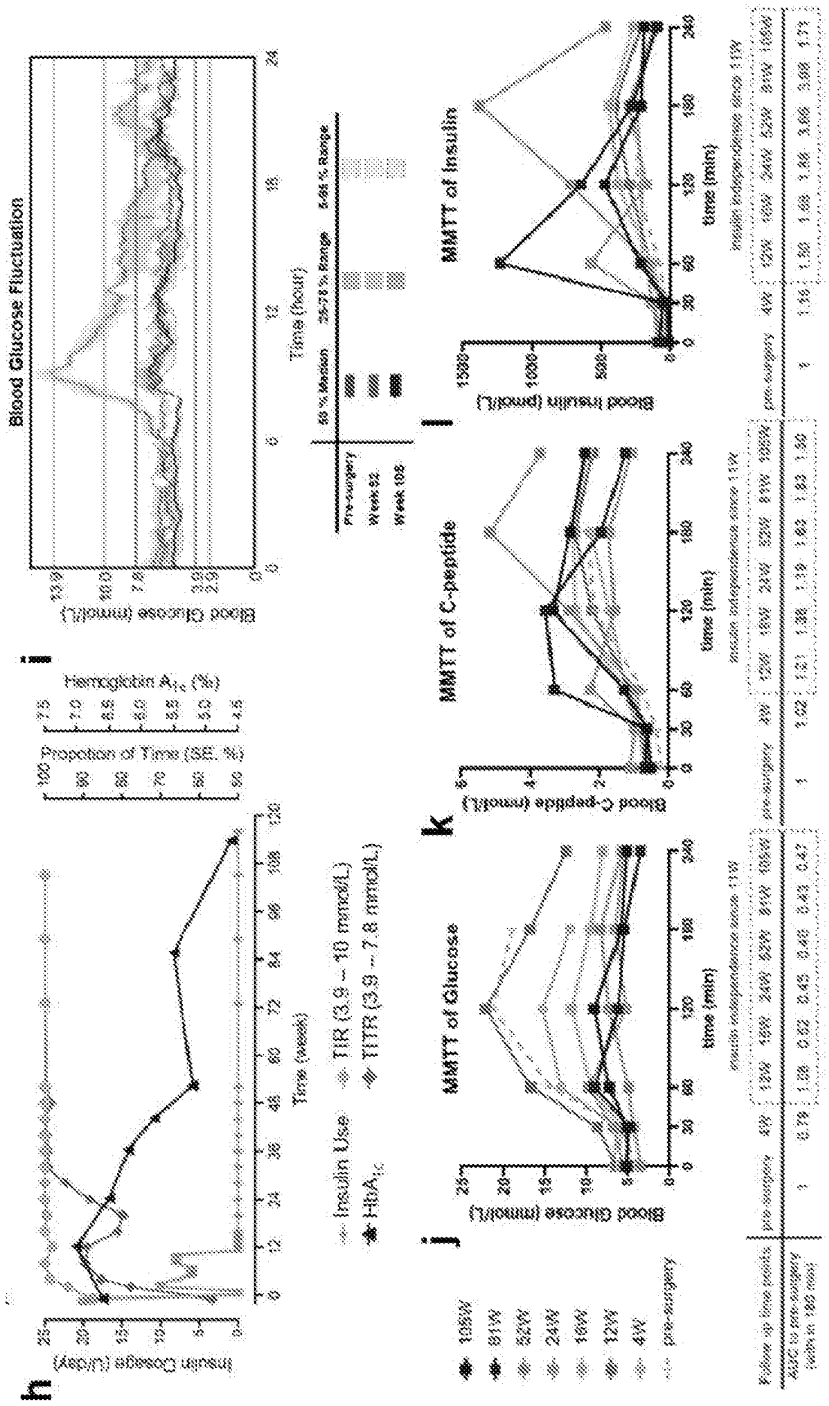


FIG. 4 (continued)

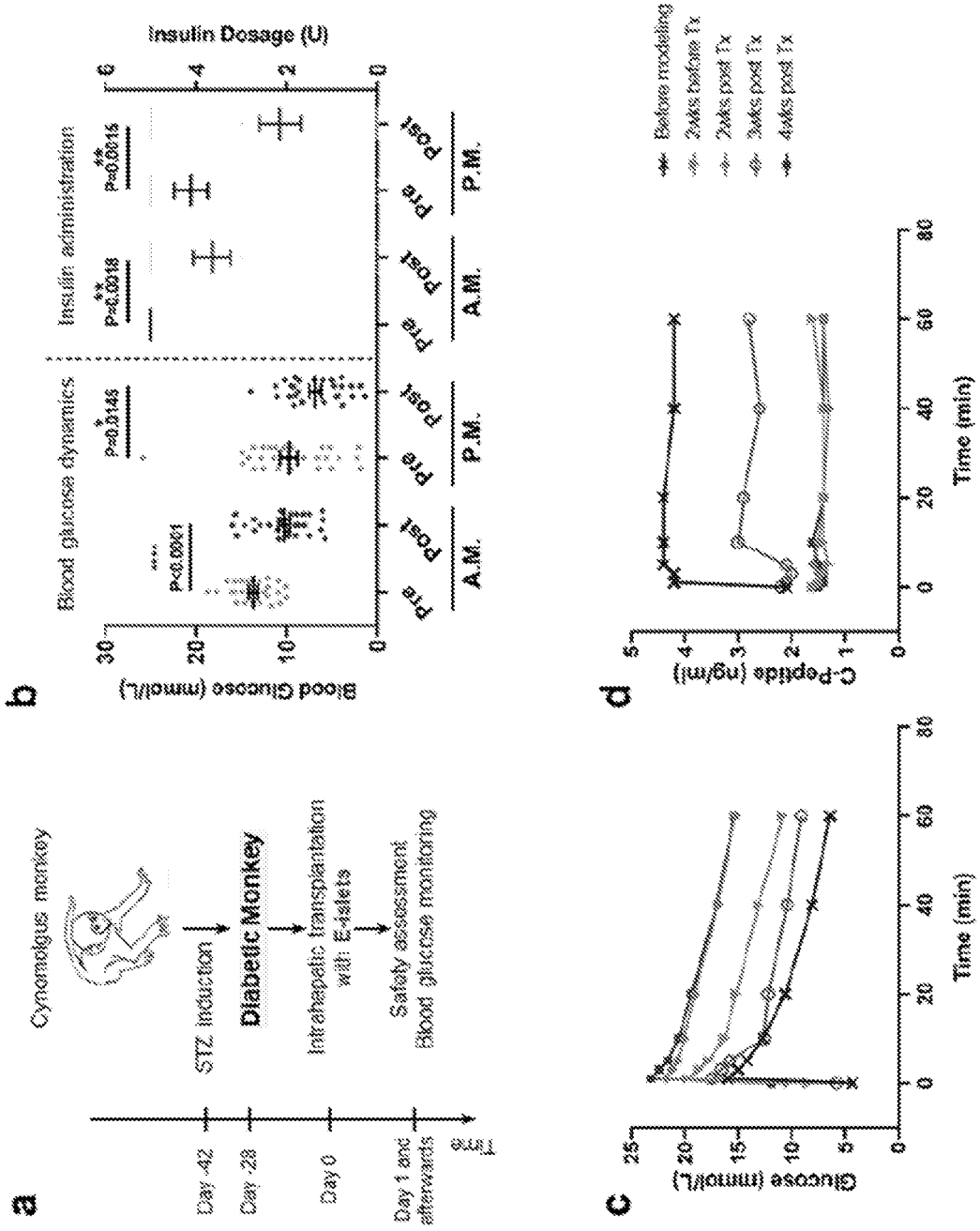

a
Cynomolgus monkey
STZ induction
Diabetic Monkey
Intrahepatic transplantation with E-islets
Safety assessment Blood glucose monitoring
Time
Day -42
Day -28
Day 0
Day 1 and afterwards
b
Blood glucose dynamics
Insulin administration
Insulin Dosage (U)
Blood Glucose (mmol/L)
P<0.0001
P=0.0145
P=0.0018
P=0.0015
Pre
Post
A.M.
P.M.
c
Glucose (mmol/L)
Time (min)
d
Before modeling
2wks before Tx
2wks post Tx
3wks post Tx
4wks post Tx
C-Peptide (ng/ml)
Time (min)

FIG. 5

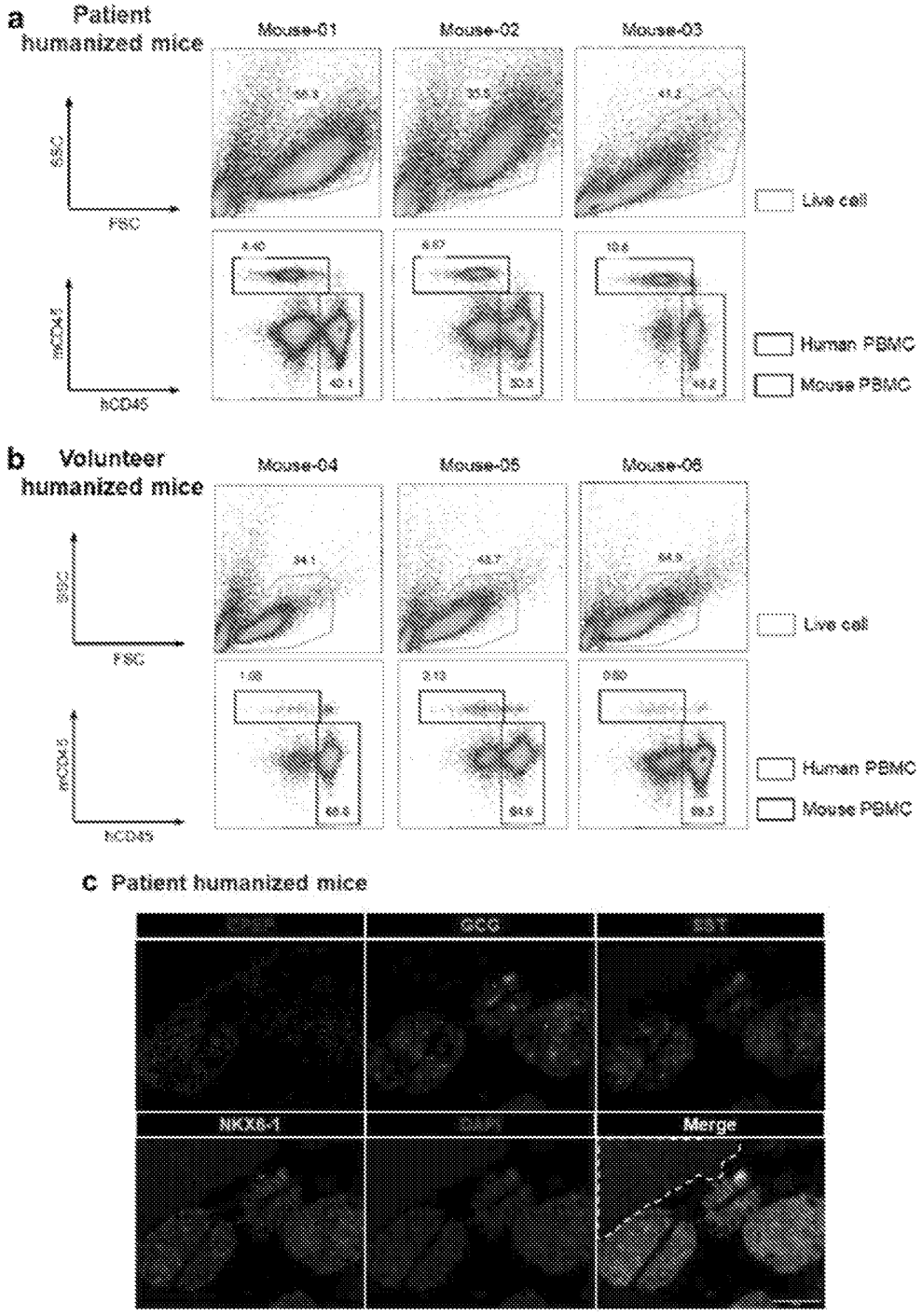

a Patient humanized mice
Mouse-01
Mouse-02
Mouse-03
SSC
FSC
Live cell
hCD45
Human PBMC
Mouse PBMC
b Volunteer humanized mice
Mouse-04
Mouse-05
Mouse-06
c Patient humanized mice
NKX6-1
Merge

FIG. 6

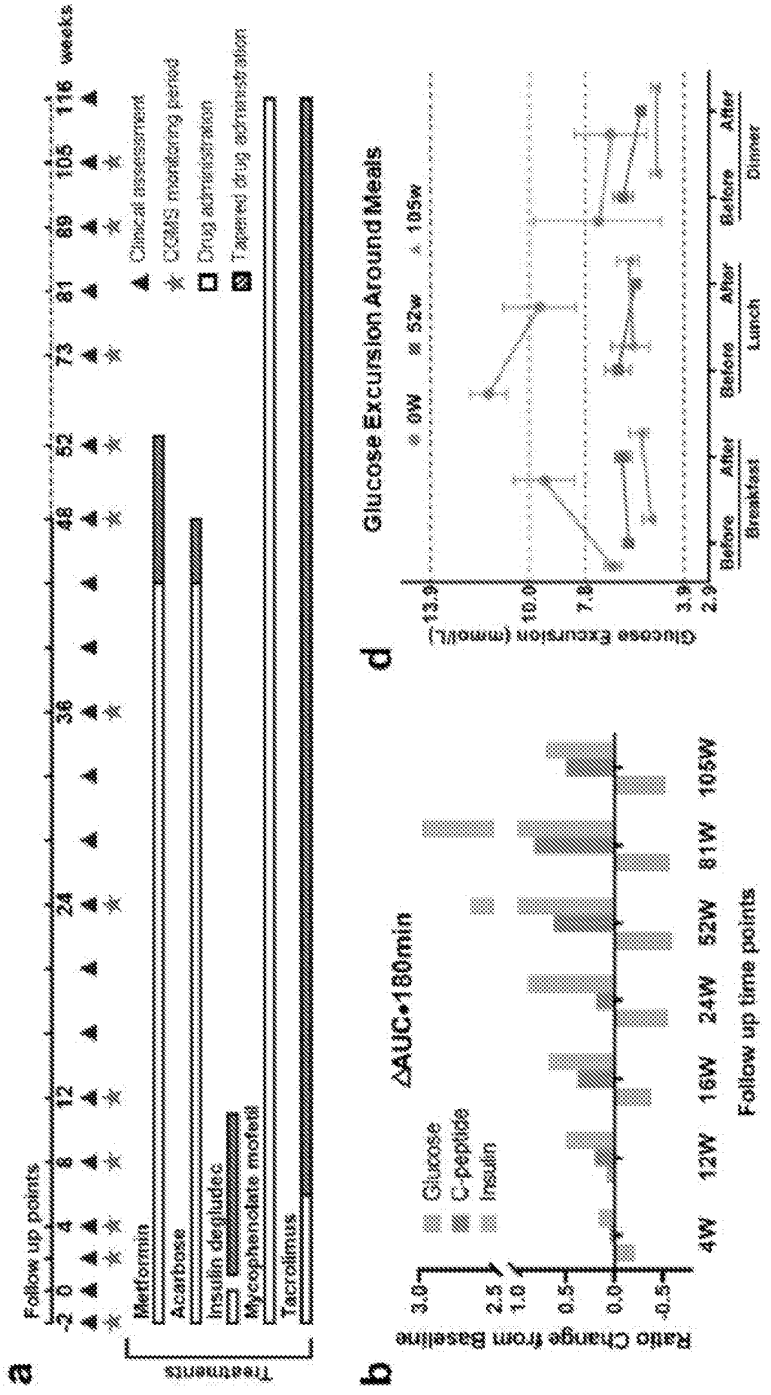

a
Follow up points
-2 0 4 8 12 24 36 48 62 73 81 89 105 116 weeks
Clinical assessment
CGMS monitoring period
Drug administration
Tapered drug administration
Metformin
Acarbose
Insulin degludec
Mycophenolate mofetil
Tacrolimus
Treatments
b
ΔAUC•180min
Glucose
C-peptide
Insulin
Ratio Change from Baseline
3.8
2.5
1.0
0.5
0.0
-0.5
4W 12W 16W 24W 52W 81W 105W
Follow up time points
d
Glucose Excursion Around Meals
0W 52w 105w
Glucose Excursion (mmol/L)
13.9
10.0
7.8
3.9
2.9
Before After Before After Before After
Breakfast Lunch Dinner

FIG. 7

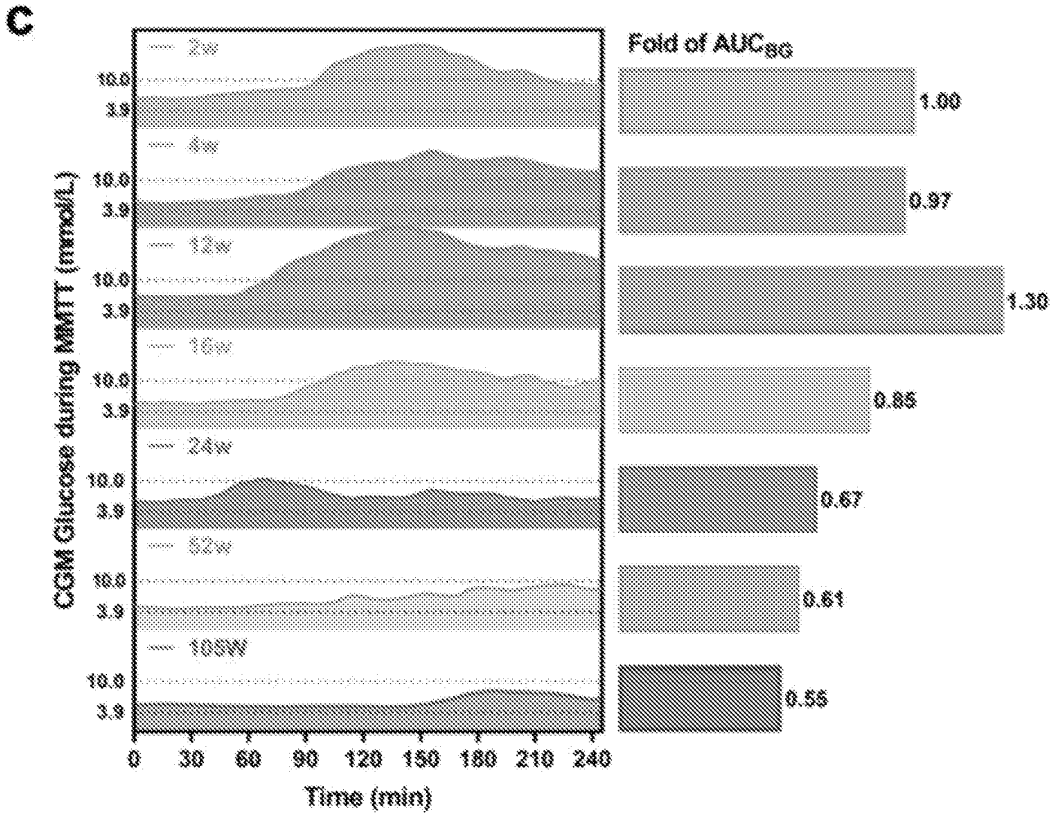


FIG. 7 (continued)

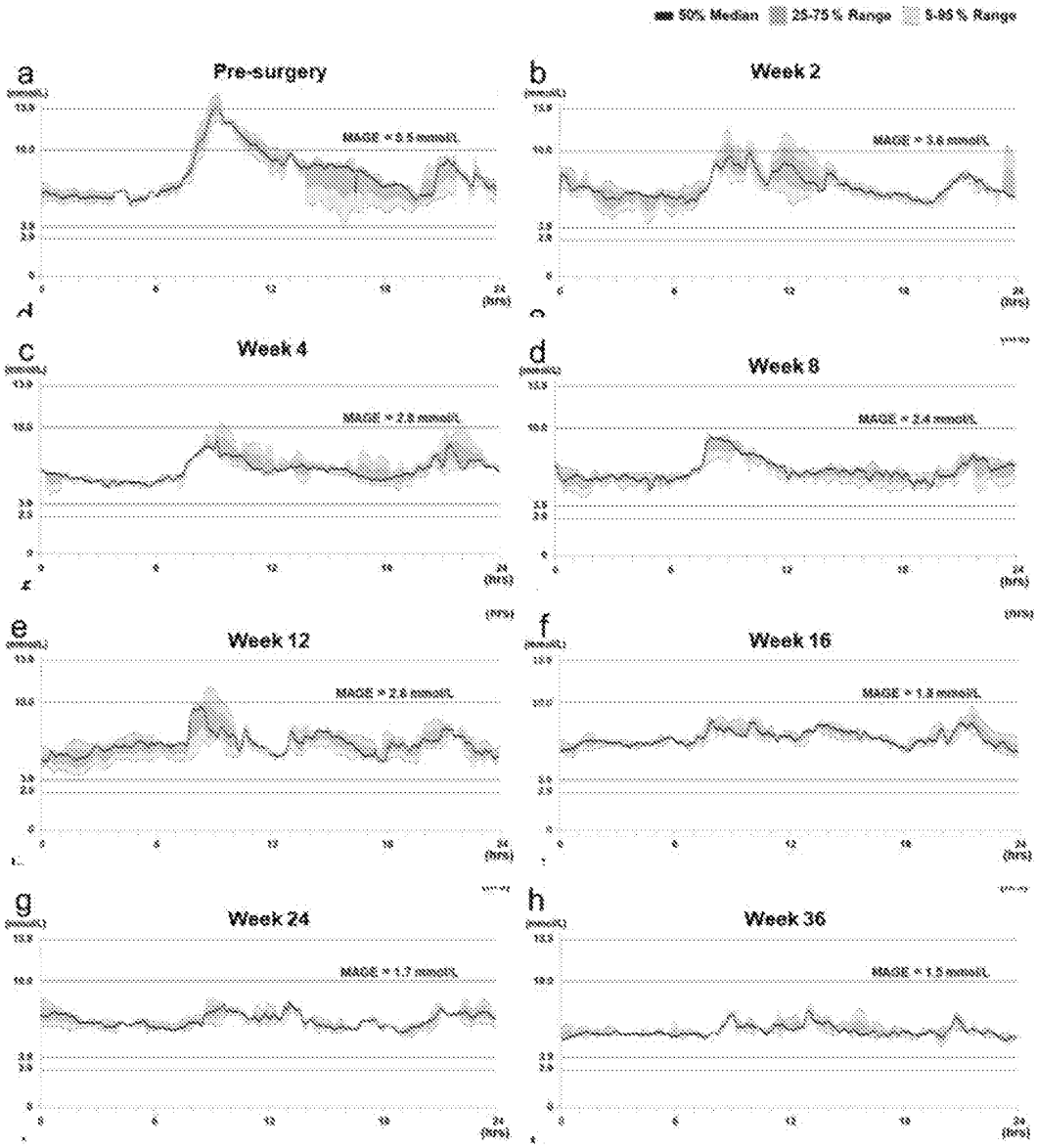

50% Median
25-75 % Range
5-95 % Range
a
Pre-surgery
MAGE = 3.5 mmol/L
b
Week 2
MAGE = 3.6 mmol/L
c
Week 4
MAGE = 2.8 mmol/L
d
Week 8
MAGE = 2.4 mmol/L
e
Week 12
MAGE = 2.8 mmol/L
f
Week 16
MAGE = 1.8 mmol/L
g
Week 24
MAGE = 1.7 mmol/L
h
Week 36
MAGE = 1.5 mmol/L

FIG. 8

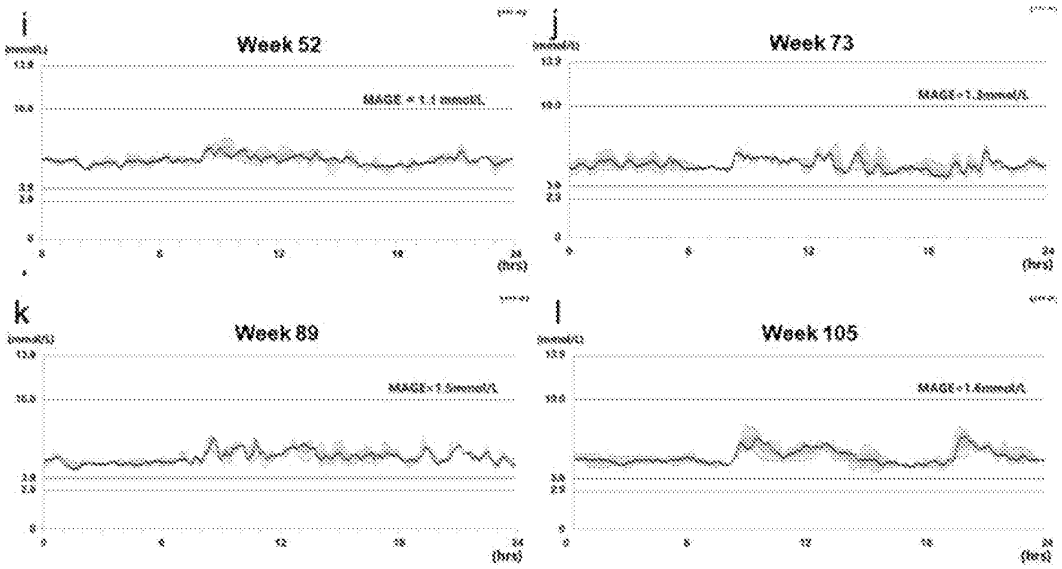


FIG. 8 (continued)

Cell Types (cell number)	Animal model (transplantation site, period)	Number	Teratoma forming percentage
hPSC (1×10^5)	SCID Beige mice (cervical subcutaneous or intramuscular, 4-6 weeks)	20	100%
EnSC (1×10^7)	SCID Beige mice (cervical subcutaneous or intramuscular, 6 months)	20	0
E-islet (1000 IEQ)	SCID Beige mice (kidney capsule, 4-6 months)	20	0
	NCG-hIL15 humanized mice (kidney capsule, 4 weeks)	6	0
E-islet (6000 IEQ)	Monkey (intrahepatic, 2 months)	1	0
E-islet (30000 IEQ)	Monkey (intrahepatic, 2 months)	1	0

FIG. 9

Quality control release criteria of EnSCs

Test	Item or method	Release Criteria
Cell morphology	Phase contract observation	Typical epithelial morphology with clear cell boundary. Dense clones consist of irregular cells with diameter of 3-5 μm.
Cell characterization and purity	Flow cytometry	FOXA1+ ≥ 90%
Genomic stability	Karyotype test	Intact karyotype
	Whole genome sequencing	Devoid of known cancer-related mutations and the lowest overall mutational burden compared to patient PBMC.
Cell viability	Flow cytometry	Live cell proportion >90% before frozen and >60% after thaw.
Pathogen test	Mycoplasma	Undetectable
	Sterility	Undetectable
	Virus	Negative
Safety	Teratoma formation test	No teratoma found.

Quality control release criteria of E-islets

Stage	Test	Method	Release Criteria
PP stage	Cell morphology	Phase contract observation	Typical epithelial morphology with unclear cell boundary. Cells with diameter of ≤ 3 μm.
	Cell characterization and purity	Flow cytometry	PDX1+ ≥ 60%
	Cell viability	Flow cytometry	Live cell proportion >90%
	Pathogen test	Mycoplasma	Undetectable
		Sterility	Undetectable
		Virus	Negative
		Endotoxin	≤1 EU/mL
EP stage	Cell characterization and purity	Flow cytometry	PDX1+/NKX6-1+ ≥ 60%
	Cell viability	Flow cytometry	Live cell proportion >90%
	Pathogen test	Mycoplasma	Undetectable
		Sterility	Undetectable
		Virus	Negative
		Endotoxin	≤1 EU/mL

FIG. 10

E-islet	Morphology	Phase contract observation	Dense spherical cell mass with unclear cell boundary. Average diameter at 150 μm.
	Cell proportion and purity	Flow cytometry	① Pancreatic lineage: PDX1+ ≥ 85% ② Endocrine lineage: CHGA+ ≥ 60% ③ Endocrine cell proportion: β cell (C-peptide+) ≥ 40%; α cell (Glucagon+) ≥ 20%; δ cell (Somatostatin+) ≤ 15% ④ Untargeted hepatic lineage: AFP+ ≤ 2%
	Cell viability	Flow cytometry	Live cell proportion >90%
	In vitro functionality	Static glucose stimulated insulin (C-peptide) secretion assay	Positive, ≥ 1.5 folds change
	Pathogen test	Mycoplasma	Undetectable
		Sterility	Undetectable
		Virus	Negative
		Endotoxin	≤1 EU/mL

FIG. 10 (continued)

Gene	Sequences (Forward, 5' to 3')	Sequences (Reverse, 5' to 3')
PDX1	GAACGCCACACAGTGCCAAATC	AACGCGCATGGGTCCTTGTAAA
NKX6-1	ACACACTGCTGTGCAACTAAAG	ACTTCTAGTCAGTGTAACTCACGATT
INS	TTTGTGAACCAACACCTGTGCGG	GCGGGTCTTGGGTGTGTAGAAGAA
GCG	TTCCCAGAAGAGGTCGCCATTGTT	CAACCAGTTTATAAAGTCCCTGGCGG
SST	GAGAATGATGCCCTGGAACCTGAAGA	ATTCTTGCAGCCAGCTTTGCGT
TBP	TTGCTGAGAAGAGTGTGCTGGAGATG	CGTAAGGTGGCAGGCTGTTGTT

FIG. 11

Antibodies	Source	Catalog No.
Anti-human Alpha-1-fetoprotein	Agilent	Cat# A0008
Anti-human Albumin	Bethyl	Cat# A80-129
Anti-mouse CD45	Biolegend	Cat# 103106
Anti-human CD45	Biolegend	Cat# 304010
Anti-Chromogranin A	Abcam	Cat# AB68271
Anti-C-peptide	Cell Signaling Technology	Cat# 4593S
Anti-Glucagon	Sigma-Aldrich	Cat# G2654
Anti-FOXA1	Santa Cruz Biotechnology	Cat# sc-101058
Anti-NKX6-1	DSHB	Cat# F55A12
Anti-PDX1	R & D Systems	Cat# AF2419
Anti-Somatostatin	Santa Cruz Biotechnology	Cat# sc-74556
Anti-SOX17	R & D Systems	Cat# BAF1924
Streptavidin APC	BD	Cat# 554067

FIG. 12

Alexa Fluor 488 Goat Anti-Mouse IgG2a	Jackson ImmunoResearch Labs	Cat# 115-545-206
Alexa Fluor 647 Donkey Anti-Mouse IgG	Jackson ImmunoResearch Labs	Cat# 715-605-151
Alexa Fluor 488 Donkey Anti-Rabbit IgG	Jackson ImmunoResearch Labs	Cat# 711-545-152
Alexa Fluor 647 Donkey Anti-Goat IgG	Thermo Fisher Scientific	Cat# A-21447
Alexa Fluor 488 Rabbit Anti-Mouse IgG	Jackson ImmunoResearch Labs	Cat# 315-545-003
Normal Goat IgG Biotinylated Control	R & D Systems	Cat# BAF108
Normal Mouse IgG2a	Santa Cruz Biotechnology	Cat# sc-3878
Normal Goat IgG Control	R & D Systems	Cat# AB-108-C
Rabbit IgG Isotype Control	Cell Signaling Technology	Cat# 3900s
Purified Mouse IgG1 Kappa Isotype Control	BD	Cat# 557273
Calcein Blue AM Viability Dye	Thermo Fisher Scientific	Cat# 650855

FIG. 12 (continued)

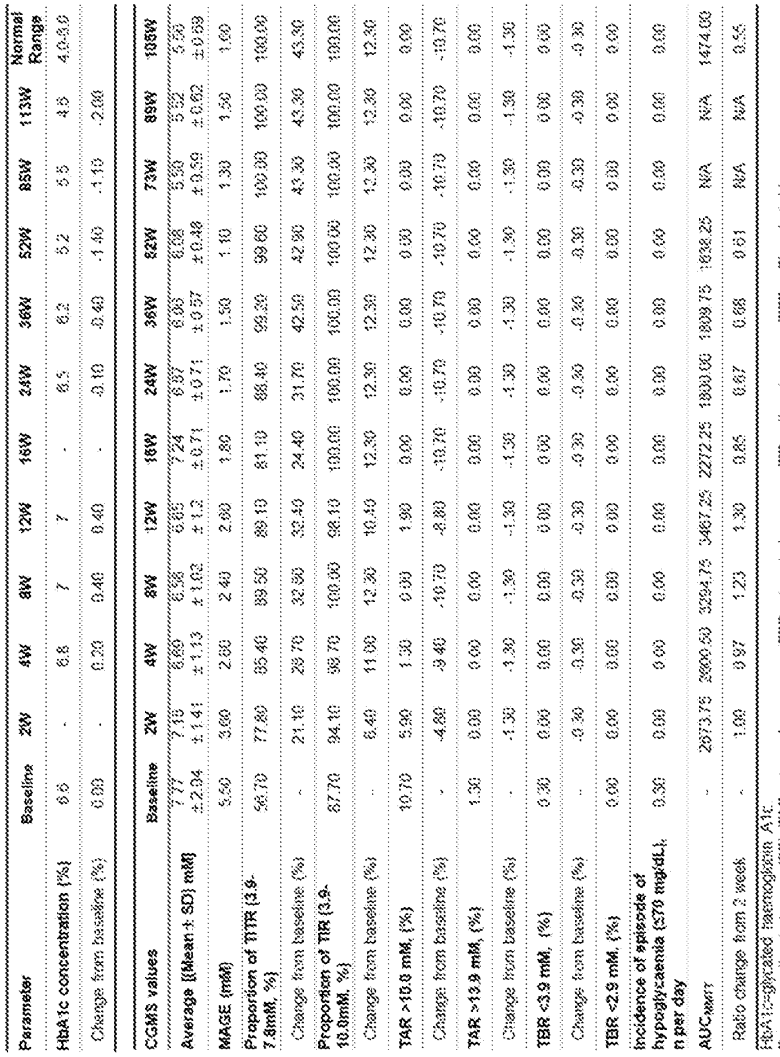
Parameter	Baseline	2W	4W	8W	12W	16W	24W	52W	81W	116W	Normal Range
White-cell count (10^9/Liter)	5.2	4.1	5.1	5.3	4.4	5.2	5.5	4.6	3.9	5.6	3.5-9.5
Hemoglobin (g/Liter)	143	140	136	144	134	131	131	125	116	130	130-175
Platelet count (10^9/Liter)	177	194	155	171	151	194	243	192	202	143	125-350
Total Bilirubin (umol/Liter)	13.4	8.3	6.6	8.2	14.1	11.1	11.2	15.2	12.2	8.0	3.4-17.1
Directed Bilirubin (umol/Liter)	2.7	0	1.5	1.7	2.1	1.8	0	3.4	2.6	3.5	0-3.4
Albumin (g/Liter)	38	39.1	39.9	40.7	37.8	41.7	45.9	41.8	34.1	40.9	40-55
Aspartate aminotransferase (U/Liter)	15	20	17	17	14	26	27	23	21	17	15-40
Alanine aminotransferase (U/Liter)	10	30	13	11	12	23	21	19	17	23	9-50
Creatinine (umol/Liter)	87	84	90	77	77	72	60	87	82	103	57-111
estimated GFR (CKD-EPI calculation, ml/min per 1.73 m^2)(ml/min)	84	82	80	94	94	97	-	83	89	72	>60
D-dimer (ug/ml)	0.56	-	-	0.64	1.35	0.29	0.5	0.99	-	-	0-1
CD4+ cell count (%)	26	29.7	27.8	28	31.9	35.8	-	21.3	28.7	-	25.8-41.6
CD8+ cell count (%)	41	37.3	38.1	38.3	39.9	39	-	39.6	40.6	-	18.1-29.6
CD4/CD8 ratio	0.63	0.8	0.73	0.73	0.8	0.92	-	0.43	0.71	-	1.57-2.93
Insulin specific antibodies*	Neg	-	-	-	Neg	Neg	-	-	-	-	Neg
FK506 concentration (ng/ml)	3.6	4.5	3.7	3.9	3	5.9	-	5.9	4.7	5.2	5-10

* Included IA-2A, Zn-8, IAA, GADA, and ICA-40KDA, ICA-64KDA.

FIG. 13

Parameter	Baseline	2W	4W	8W	12W	16W	24W	36W	52W	81W	105W	Normal Range
Mixed meal Tolerance Test												
Fasting serum glucose concentration (mmol/L)	3.9	3.9	6.8	5.4	6.4	6	5.6	4.75	3.5	5.2	5.6	3.9-6.1
15min serum glucose concentration (mmol/L)	4.9	7.2	5.2	5.7	6.6	5.9	6.4	5.21	4.1	4.8	5.4	3.9-6.1
30min serum glucose concentration (mmol/L)	5.9	10.5	6.3	7.2	8.7	5.9	6.8	5.64	4.3	4.8	5.1	<10.5
60min serum glucose concentration (mmol/L)	14.3	8.3	13.1	17.1	16.7	9.4	9.8	5.96	4.9	8.0	7.3	<8.9
120min serum glucose concentration (mmol/L)	11.3	6.7	15.3	21.6	22.2	11.8	5.2	7.51	7.3	6.2	9.1	<7.8
180min serum glucose concentration (mmol/L)	10	5.3	12	17.8	16.8	9.5	6.6	6.61	8.3	5.4	5.8	<5.9
240min serum glucose concentration (mmol/L)	-	-	-	12.9	13.5	8.1	5.5	4.75	5.8	3.5	5.1	3.9-6.1
AUC 180min of Glucose	2727	1579	2138	2899	2930	1882	1242	1158	1092	1161	1277	-
Fold change in the AUC from baseline	0	-0.4219	-0.2160	0.0631	0.0744	-0.3932	-0.545	-0.5754	-0.5996	-0.5743	-0.5317	-
Fasting serum C-peptide (nmol/L)	0.23	0.43	0.49	0.47	0.66	0.7	1.09	0.29	0.53	0.68	0.51	0.37-1.47
15min serum C-peptide (nmol/L)	0.33	0.56	0.55	0.41	0.61	0.71	0.96	0.24	0.52	0.59	0.69	0.37-1.47
30min serum C-peptide (nmol/L)	0.35	0.5	0.61	0.54	0.77	0.85	0.97	0.45	0.8	0.62	0.69	-
60min serum C-peptide (nmol/L)	0.74	0.67	1.23	0.8	1.13	1.29	2.26	0.65	0.93	3.3	1.23	-
120min serum C-peptide (nmol/L)	2.15	3.33	1.71	1.91	2.2	2.7	1.56	1.82	2.88	3.56	3.34	-
180min serum C-peptide (nmol/L)	2.33	1.99	2.25	1.9	2.71	2.76	1.73	1.43	5.16	1.96	2.64	-
240min serum C-peptide (nmol/L)	-	-	-	1.8	2.19	2.35	1.07	1.12	3.72	1.24	2.41	0.37-1.47
AUC 180min of C-peptide	246.8	332.5	251.1	229.4	295.6	337.9	291.6	197.3	399.2	449.7	369.3	-
Fold change in the AUC from baseline	0	0.2662	0.0174	-0.0705	0.1977	0.3691	0.1815	-0.2006	0.6175	0.8265	0.4963	-
Fasting serum insulin (pmol/L)	16.84	22.6	16.71	16.64	52.96	48.62	94.32	32.53	41.73	83.79	12.36	18.11-173.43
15min serum insulin (pmol/L)	34.59	58.43	39.95	24.16	40.64	520.3	128.9	-	54.58	61.0	37.56	-
30min serum insulin (pmol/L)	29.74	36.04	46.61	60.86	72.63	38.56	114.4	163.07	138.7	56.32	27.05	-
60min serum insulin (pmol/L)	84.79	62.65	205.7	112	147.3	141.2	573.7	173.45	161.5	1232.0	215.9	-
120min serum insulin (pmol/L)	241.4	663.9	206.1	244.2	295	381	171.4	519.53	721.5	644.3	473.8	-
180min serum insulin (pmol/L)	229.5	138.5	345.8	204.7	408.1	441	298.4	436.71	1383	277.3	212.4	-
240min serum insulin (pmol/L)	-	-	-	132	210	282.8	118	-	469.1	98.1	189.3	18.11-173.43
AUC 180min of insulin	26619	48673	30770	27690	39212	51481	30267	56858	96300	105363	45512	-
Fold change in the AUC from baseline	0	0.8285	0.1559	0.0402	0.4731	0.9340	0.8884	1.1359	2.6177	2.9586	0.7097	-

FIG. 14



Parameter	Baseline	2W	4W	8W	12W	16W	24W	36W	52W	85W	113W	Normal Range
HbA1c concentration (%)	6.6	-	6.8	7	7	-	6.5	6.2	6.2	6.5	4.6	4.0-6.0
Change from baseline (%)	0.00	-	0.20	0.40	0.40	-	-0.10	-0.40	-1.40	-1.10	-2.00	

CGMS values	Baseline	2W	4W	8W	12W	16W	24W	36W	52W	73W	85W	113W
Average [(Mean ± SD) mM]	7.77 ± 2.04	7.16 ± 1.41	6.89 ± 1.13	6.98 ± 1.03	6.85 ± 1.2	7.24 ± 0.71	6.97 ± 0.71	6.95 ± 0.57	6.68 ± 0.48	5.58 ± 0.59	5.52 ± 0.62	5.55 ± 0.69
MAGE (mM)	5.50	3.60	2.80	2.40	2.60	1.80	1.70	1.50	1.10	1.30	1.50	1.60
Proportion of TITR (3.9-7.8mM, %)	56.70	77.80	85.40	89.50	89.10	91.10	88.40	89.20	99.60	100.00	100.00	100.00
Change from baseline (%)	-	21.10	28.70	32.80	32.40	24.40	31.70	42.50	42.90	43.30	43.30	43.30
Proportion of TIR (3.9-10.0mM, %)	87.70	94.10	96.70	100.00	98.10	100.00	100.00	100.00	100.00	100.00	100.00	100.00
Change from baseline (%)	-	6.40	11.00	12.30	10.40	12.30	12.30	12.30	12.30	12.30	12.30	12.30
TAR >10.0 mM, (%)	10.70	5.90	1.30	0.00	1.90	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Change from baseline (%)	-	-4.80	-9.40	-10.70	-8.80	-10.70	-10.70	-10.70	-10.70	-10.70	-10.70	-10.70
TAR >13.9 mM, (%)	1.30	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Change from baseline (%)	-	-1.30	-1.30	-1.30	-1.30	-1.30	-1.30	-1.30	-1.30	-1.30	-1.30	-1.30
TBR <3.9 mM, (%)	0.30	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Change from baseline (%)	-	-0.30	-0.30	-0.30	-0.30	-0.30	-0.30	-0.30	-0.30	-0.30	-0.30	-0.30
TBR <2.9 mM, (%)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Incidence of episode of hypoglycaemia (≤70 mg/dL), n per day	0.30	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
AUC_{last}	-	2573.75	2600.50	3294.75	3467.25	2272.25	1800.00	1809.75	1838.25	N/A	N/A	1474.00
Ratio change from 2 week	-	1.00	0.97	1.23	1.30	0.85	0.67	0.68	0.61	N/A	N/A	0.55

HbA1c=glycated haemoglobin A1c.
Data are estimated mean (SE). TAR = time above range; TBR = time below range; TIR = time in range; TITR = Time in tight range
Time (minutes), Percent (%), N/A: Not available

FIG. 15