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healthy subjects. In the presence of 4 g/l glucose, diabetic macrophage migration toward FMLP was significantly higher than that observed with healthy subjects' macrophages: 2.96 ± 0.15 vs. 1.96 ± 0.63, $p<0.05$. For migration toward native insulin the data were: 0.95 ± 0.06 vs. 0.73 ± 0.07. Neither native nor insulin recovered from reservoir stimulated IL-1β release from diabetic macrophages. TNF-α release in the presence of both kinds of insulin was 115.8 ± 13.3% and 100 ± 11.5% and not different from those of healthy subjects. Similar results were observed with diabetic macrophages differentiated in presence of 4 g/l glucose. Pump-reservoir insulin failed to modify both chemotaxis and TNF-α or IL-1β release from macrophages of diabetic patients with catheter obstruction. Under hyperglycaemic environment, diabetic macrophages exhibited high chemotactic responses toward both FMLP and pump insulin.

No loss of human (mantle-) islets by OptiPrep-UWS purification following isolation in UWS

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Human islet purification is hampered by many donor and isolation factors that may lead to deterioration of the density difference between islets and acini, or the isolation of islets covered with a mantle of acinar cells at the periphery ("mantle-islets") which generally results in a poor purity and islet loss after density separation. We recently reported a first series of 5 experiments with human islets isolated by the automated method and collected in cold RPMI before purification in our novel density gradient of iodixanol (OptiPrep) in University of Wisconsin solution (UWS) which resulted in ≥ 80% purity and recovery. The digests in this first series contained, however, no mantle-islets. We now report the efficacy of the gradient in a series of 6 consecutive experiments performed with a simple manual digestion method which facilitated testing whether using the UWS during all cold isolation steps better preserves the tissue and, furthermore, because of the frequent high proportion of mantle-islets in this study, whether the gradient may also save these islets from pelleting with the acinar tissue. Pancreases were obtained from multiorgan cadaveric donors (13-58 years old) and cold preserved with UWS for 3-11 h. Tail segments were intraductally digested with 1.4 mg/ml Liberase-HI in HBSS by static incubation at 37° C for 21-32 min. The tissue was dispersed in cold UWS (Viaspan) by shaking and sieving (500 µm). Aliquots were taken for assessment of the digest and for testing the purification in 1 to 3 different discontinuous test-gradients before final purification of the bulk of the digest bottom-loaded in a discontinuous gradient of 3 layers: a bottom with a density for the different isolations of 1100-1080 mg/ml, a barrier layer of 1090-1072.5 mg/ml, and UWS on top. The solutions were prepared by mixing UWS with an OptiPrep-UWS (equal volumes of OptiPrep and double-strength UWS). After a 5-min, 500 *g* centrifugation at 4° C all (mantle-) islets were recovered at a density lower than 1075, 1080, 1090, 1086, 1100, 1086, respectively for the consecutive isolations. The digest yield was 1797 ± 410 IEQs/g, the islet diameter was 163 ± 7 µm, and 40% (10%-60%) were mantle-islets. After purification, 1702 ± 334 IEQs were recovered (104 ± 10% recovery), islet diameter was 167 ± 11 µm and purity was 85 ± 7% not taking into account the contamination with acinar mantles (74% purity including mantles). Thus, the consistent high efficacy (no loss of islets, mantle-islets, islet integrity, and a high purity) indicates that the gradient may become a new powerful tool for human islet purification.

Preservation of the spleen after pancreas resection and islet autotransplantation for chronic pancreatitis

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Preservation of the spleen is not routine after pancreas resection for chronic pancreatitis (CP). Splenic preservation often requires devascularisation and with the addition of an islet autotransplant (IAT) this may result in portal hypertension, thrombosis or haemorrhage. The aim of this study was to assess the feasibility of splenic preservation after pancreas resection and IAT.

All patients underwent per-operative measurement of portal pressure along with post-operative abdominal ultrasound with power Doppler to assess splenic vascularity and the patency of the remaining portal and splenic vessels.

Of 36 patients having pancreas resection, the spleen was preserved in 30 patients (mean age 41 years). Eighteen of these underwent simultaneous islet autotransplantation. In this cohort portal pressure increased by 8 mm Hg. The aetiology of CP was mainly idiopathic or alcohol-related. All patients presented with chronic abdominal pain (mean 4.5 years) and required opiate-derived analgesia for pain relief (e.g. MST, pethidine, dihydrocodeine).

The spleen was preserved with an intact splenic artery and vein in 19 patients (60%) and in the remainder by the short gastric vessels (n=10). In 1 patient the splenic artery was sacrificed but the splenic vein remained intact. The mean duration of the procedure was 7 hours (range 5-11 h) and mean blood loss was 925 ml. The 30-day mortality was 4% (n=1).

Six patients developed splenic and portal complications. These included splenectomy (n=2), intra-splenic collection (n=2), a wedge splenic infarct and a partial portal venous thrombosis. Three of these complications occurred in IAT recipients. Splenic ultrasound and power Doppler did not demonstrate any other abnormalities. Flow was detected in all patients with intact splenic arteries and veins (n=19). The mean duration of hospital stay was 24 days. Of the 24 patients with at least 6 months of follow-up after pancreas resection, 82% (n=20) have complete relief of pain. Preservation of the spleen is possible after pancreas resection and IAT. The rise in portal pressure after IAT does not increase the risk of splenic complications.

Temporal relationship of insulin, intact proinsulin and split proinsulin after islet autotransplantation

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Islet autotransplantation (IAT) provides insulin independence after pancreas resection but is critically dependent on the islet mass. Previously, patients have demonstrated insulin deficiency, raised intact proinsulin (IPI) and very high split proinsulin (SPI) in the presence of hyperglycaemia. The aim of this study was to assess the temporal relationship of insulin, intact proinsulin and split proinsulin after IAT.

Patients underwent a pre-operative oral glucose tolerance test (75 g) which was repeated post-operatively at 6 months and yearly thereafter. Insulin was measured by an immunoenzymetric assay, SPI by a two-site specific immunometric assay, and IPI by a time-resolved fluorescence assay (Delfia).

A total of 17 patients were used in the analysis, 8 of which developed insulin independence at some point. In addition, patients with NIDDM and non-diabetic controls were also assessed. The results are summarised below.

	Glucose (mmol/l)		Insulin (pmol/l)	IPI (pmol/l)	SPI (pmol/l)*
	0 min	2 h	30 min	0 min	0 min
Pre-operative (n=12)	4.49	4.61	268.7	3.59	4.43
6 months (n=13)	8.68	16.54	84.8	8.68	25.93
1 year (n=5)	6.88	13.22	105.0	7.00	15.92
2 years (n=10)	10.2	20.67	121.4	8.89	17.66
3 years (n=8)	9.49	19.14	193.4	4.56	10.18
Control	5.5	<7.8	304	3.80	6.30
NIDDM	>7.8	>11.1	136	10.60	8.10

All values are means. *$p<0.05$