

### **Electronic supplementary materials**

*ESM Table 1* Effect of adrenaline increase on the acute increase in count of granulocytes, lymphocytes and monocytes during hypoglycemia.

	Effect	<i>p-value</i>
Granulocytes	2.029	0.004
Lymphocytes	1.212	0.001
Monocytes	0.196	0.041

## *ESM methods*

### Inclusion criteria - overall

- Ability to provide written informed consent
- Must be able to speak and read Danish (for Hillerød-site) and Dutch (for Nijmegen-site)
- Insulin treatment according to basal-bolus insulin regimen (injections or insulin pump) (except for healthy participants)
- BMI: 19-40 kg/m<sup>2</sup>
- Age  $\geq 18$  years,  $\leq 80$  years
- Blood pressure:  $< 140/90$  mmHg
- Duration of diabetes  $> 1$  year (except for healthy participants)
- HbA<sub>1c</sub>  $< 100$  mmol/mol (11.3%)

### Inclusion criteria – subgroup specific

- T2DM: Insulin treatment for at least 1 year and age  $\geq 18$  years,  $\leq 80$  years
- Healthy volunteers: HbA<sub>1c</sub>  $< 42$  mmol/mol (6%) and age  $\geq 18$  years,  $\leq 80$  years

### Exclusion criteria for participants with diabetes

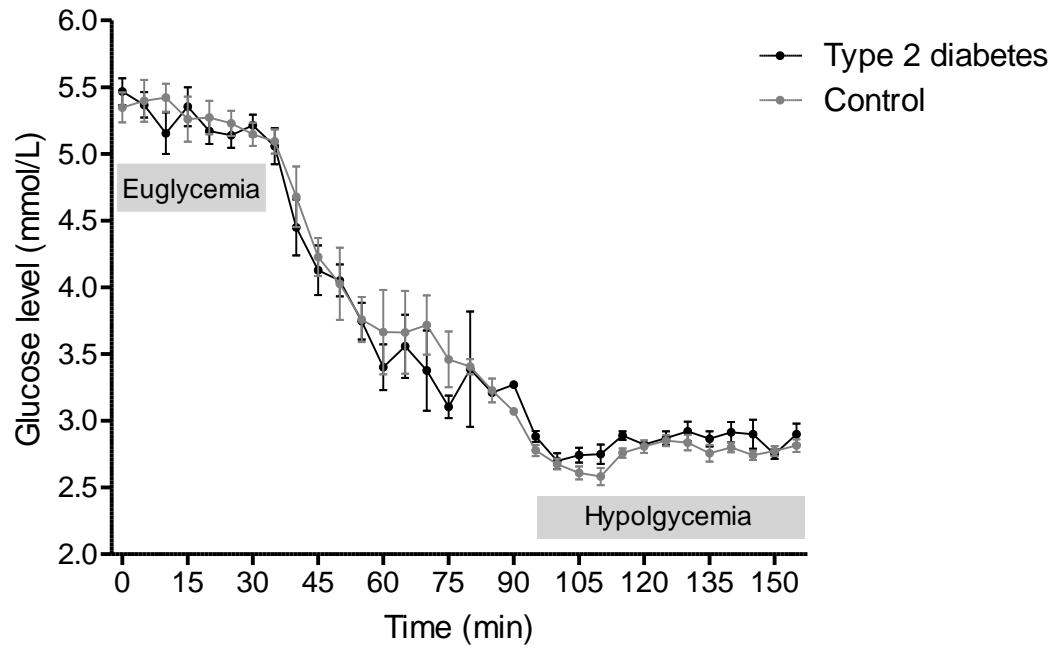
- Severe medical or psychological conditions interfering with the perception of hypoglycemia other than IAH such as brain injuries, epilepsy, a major cardiovascular disease event or anxiety disorders
- Use of immune-modifying drugs or antibiotics
- Treatment with glucose-modifying (other than insulin, SGLT-2 inhibitors and metformin) agents (e.g. prednisolone)

- Use of anti-depressive drugs
- Pregnancy or breastfeeding or unwillingness to undertake measures for birth control
- Use of statins (e.g. stop statins >2 weeks before performing blood sampling. This can be safely done in the context of primary prevention)
- Any event of cardiovascular disease in the 5 years before screening (e.g. myocardial infarction, stroke, heart failure, symptomatic peripheral arterial disease)
- Auto-inflammatory or auto-immune diseases
- Any infection in the three months before screening
- Previous vaccination in the three months before screening
- Laser coagulation for proliferative retinopathy in the six months before screening
- Proliferative retinopathy
- Diabetic nephropathy as reflected by an albumin-creatinine ratio > 30 mg/g or an estimated glomerular filtration rate (by MDRD) <60ml/min/1.73m<sup>2</sup>
- History of pancreatitis (acute or chronic) or pancreatic cancer

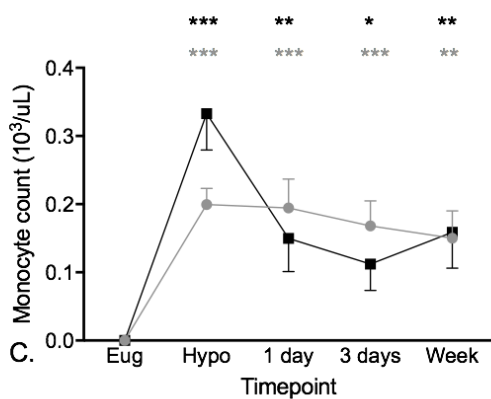
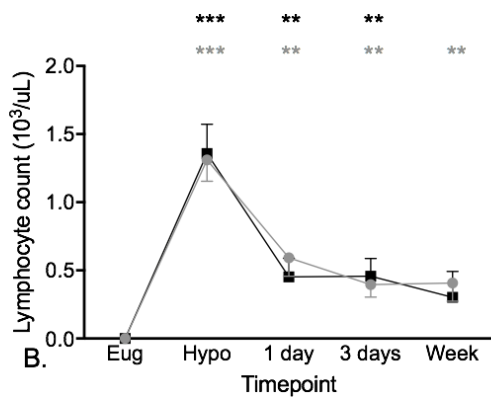
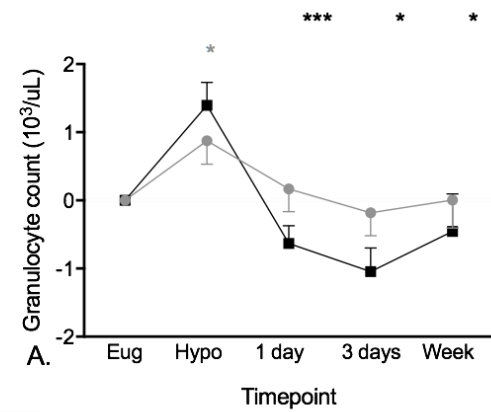
#### Exclusion criteria for participants without diabetes

- Presence of any medical condition that might interfere with the study protocol, such as brain injuries, epilepsy, a major cardiovascular disease event or anxiety disorders
- Use of any medication, except for oral contraceptives, stable dose of thyroxine
- Pregnancy or breastfeeding or unwillingness to undertake measures for birth control
- Any infection in the three months before screening
- Vaccination in the three months before screening

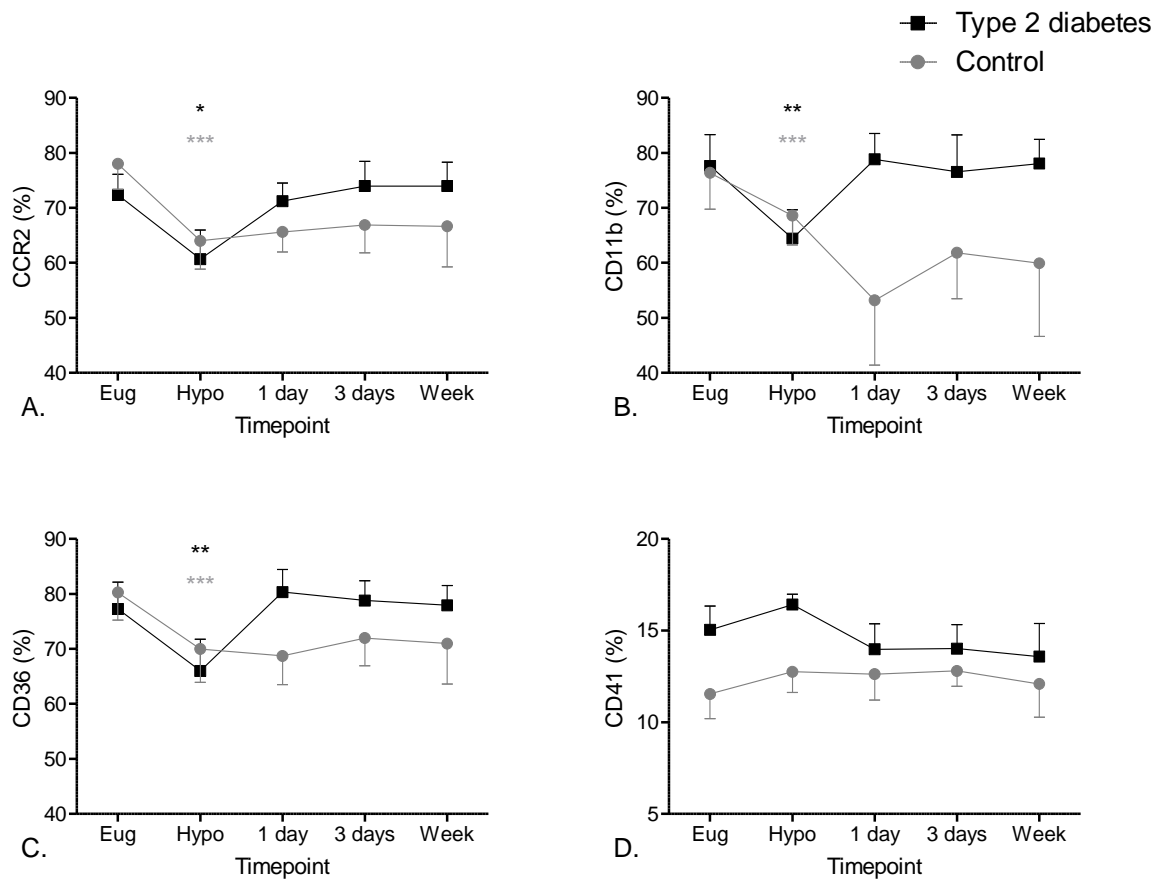
*ESM Figure 1* Determined glucose levels of the hyperinsulinemic-hypoglycemic clamp of participants with type 2 diabetes and healthy controls.



*ESM Figure 2* Counts ( $\cdot 10^3/\mu\text{L}$ ) of granulocytes (A), lymphocytes (B), and monocytes (C) in participants with type 2 diabetes (black) and controls (grey). Data are presented as the  $\Delta \pm \text{SEM}$ , \* $p < 0.05$ , \*\* $p < 0.01$  and \*\*\* $p < 0.001$  change versus euglycemia.

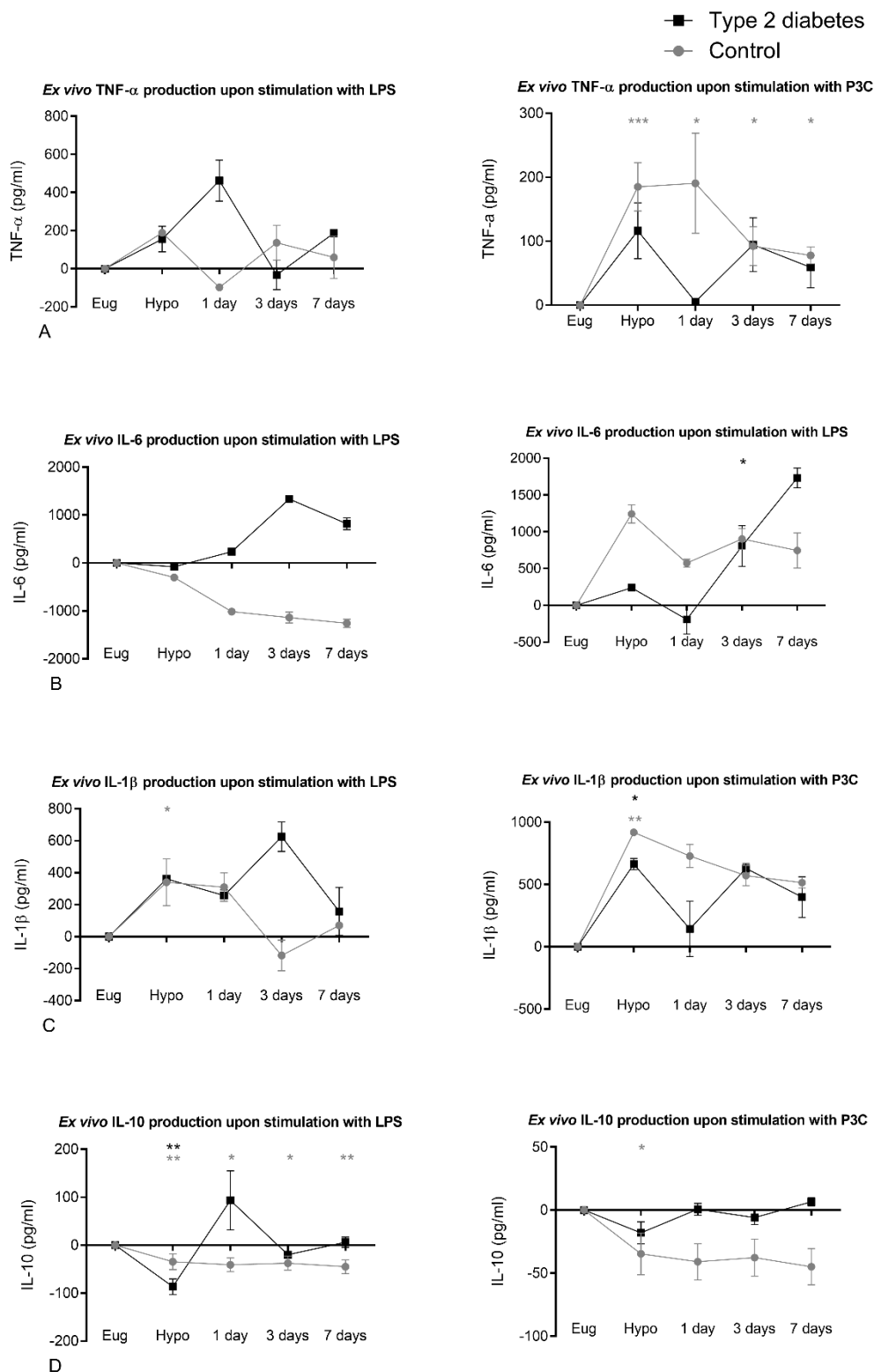


*ESM Figure 3* Percentages of monocytes at euglycemia for people with type 2 diabetes (black) and control (grey) and all timepoints characterized by CCR2 (A), CD11b (B), CD36 (C) and CD41 (D). Data presented as mean  $\pm$  SEM, \* $p < 0.05$ , \*\* $p < 0.01$  and \*\*\* $p < 0.01$  change versus euglycemia.



*ESM Figure 4 Ex vivo cytokine production of TNF- $\alpha$  (A), IL-6 (B), IL-1 $\beta$  (C) and IL-10 (D) upon LPS or P3C stimulation. Data presented as delta  $\pm$  SEM, \*p < 0.05, \*\*p < 0.01 and \*\*\*p < 0.001 change versus euglycemia.*

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[illegible]