

## TABLE OF CONTENTS SUPPLEMENTAL MATERIAL

Supplemental Table 1	Antibodies used for flow cytometry
Supplemental Table 2	Primer sequences
Supplemental Table 3	Measured parameters after low salt diet and after high salt diet in <i>study II</i> .
Supplemental Figure 1	Pilot experiments comparing anti-CD163 with anti-CD68
Supplemental Figure 2	Effect of high salt diet on monocyte markers
Supplemental Figure 3-4	Effect of salt on macrophages <i>in vitro</i> , in which the macrophages were differentiated for 7 days in the presence of high salt (whereas Figure 6 in the main text shows the results of salt stimulation after differentiation <i>without</i> any high salt medium)

**Supplemental Table 1: Antibodies used for flow cytometry.**

<b>Marker</b>	<b>Label</b>	<b>Dilution</b>	<b>Clone</b>	<b>Supplier</b>
CD14	PE-Cy7	1/50	M5E2	BD
CD16	APC-Cy7	1/50	3G8	BD
HLA-DR	PerCp-Cy5.5	1/50	G46-6	BD
CCR2	APC	1/50	48607	BD
CD206	FITC	1/25	15-2	Sony
CCR7	PE	1/25	G043H7	Biolegend
CCR5	FITC	1/10	2D7/CCR5	BD
CX3CR1	PE	1/50	2A9-1	Biolegend
CD86	FITC	1/25	IT2.2	Biolegend
CD45RA	APC	1/100	HI100	BD
TLR-4	PE	1/25	HTA125	Biolegend
CD32	FITC	1/10	FLI8.26 (2003)	BD
CD36	APC	1/25	CB38	BD
CD64	APC	1/50	10,1	Biolegend
SRA	PE	1/10	351615	R&D
CD163	APC	1/25	RM3/1	Biolegend
CD200R	PE	1/25	OX-108	Biolegend
CD18	APC	1/50	6,7	BD
CD11c	APC	1/10	B-ly6	BD
CD29	APC	1/10	MAR4	BD
CD49d	FITC	1/25	9F10	BD
CD11b	PE	1/25	ICRF44	BD
CD62L	PE	1/50	DREG-56	BD
Mouse IgG1k	FITC/PE/APC	1/50	MOPC-21	BD
isotype control				

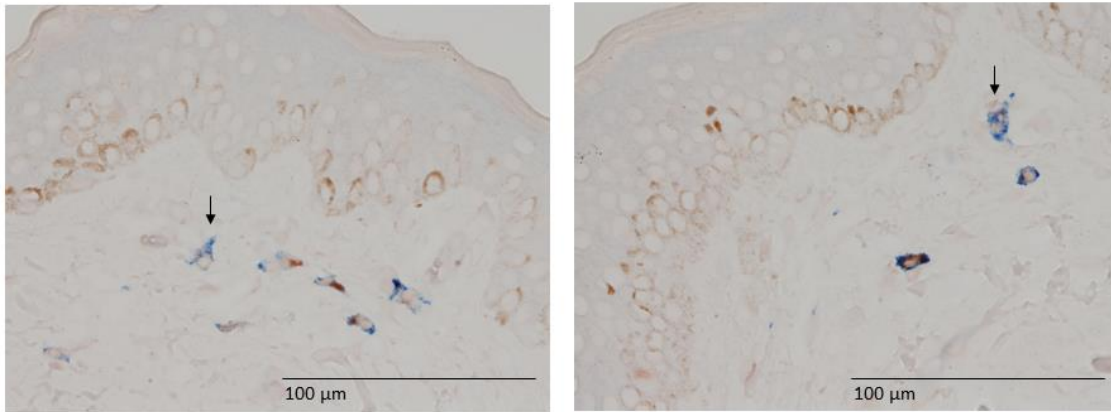
APC, allophycocyanin. CD, cluster of differentiation. CR, chemokine receptor. Cy, CyChrome. FITC, Fluorescein isothiocyanate. HLA, human leucocyte antigen. PE, Phycoerythrin. PerCP, peridinin-chlorophyll-protein. SR, scavenger receptor.

**Supplemental Table 2.** Primer sequences.

<b>Gene</b>	<b>Forward</b>	<b>Reverse</b>
CD200R	TGGATGAAAAACAGATTACACAGAA	TAATGCGATAGGAGGGCAAC
CCL22	CCTACTCTGATGACCGTGGC	AGGGAATGCAGAGAGTTGGC
CCL24	CACATCATCCCTACGGGCTC	GGTAGCTGACCACTCGGTTC
CCR2	CCACATCTCGTTCTCGGTTTATC	CAGGGAGCACCGTAATCATAATC
CD206	CACCATCGAGGAATTGGACT	ACAATTCGTCATTTGGCTCA
CLEC10A	GAACGGCAGGCAGTTCATTC	AGGCTTCCTCACCATTGTTGT
FCER2	CCAGGAATTGAACGAGAGGAAC	TTGATCCACTTTTCAGGGCAC
GNB2L1	GAGTGTGGCCTTCTCCTCTG	GCTTGCAGTTAGCCAGGTTC
HPRT1	GACCAGTCAACAGGGGACAT	AACACTTCGTGGGGTCCTTTTC
SOCS1	TCTCACCTCTTGAGGGGGTC	AGAGGTAGGAGGTGCGAGTT
TGM2	ATAAGTTAGCGCCGCTCTCC	CTCTCTAAGACCAGCTCCTCG

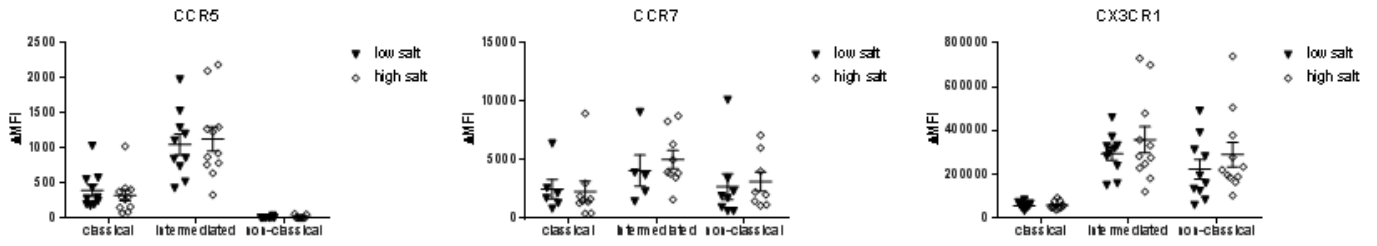
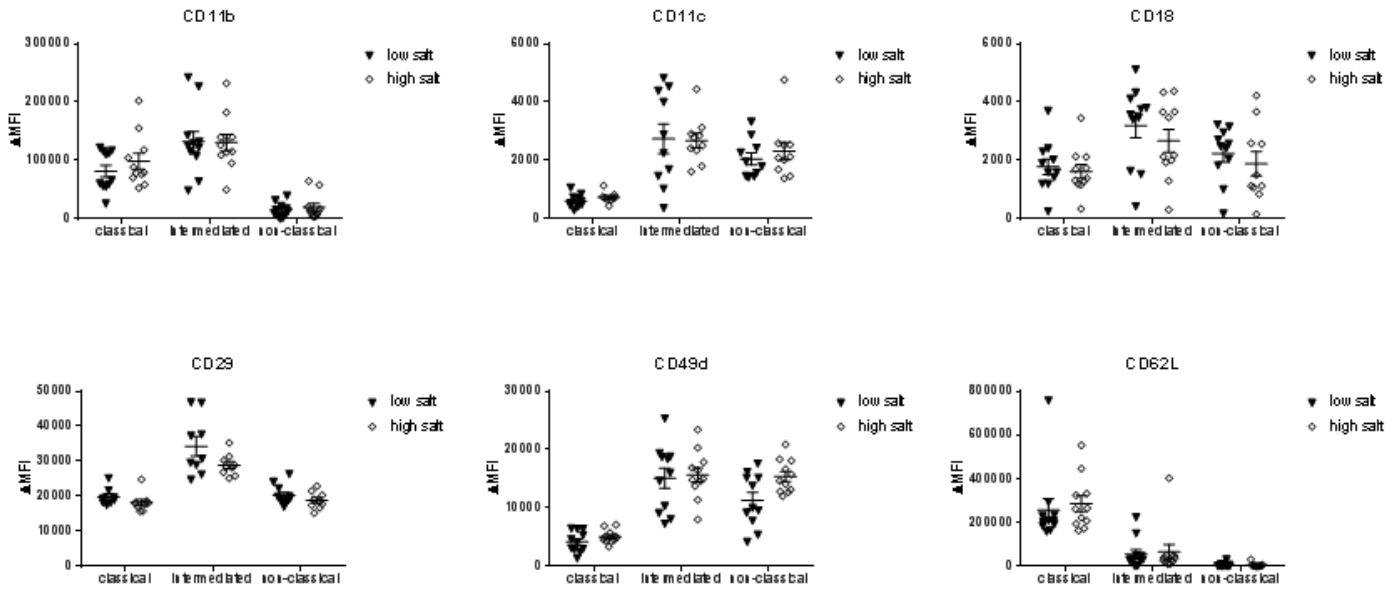
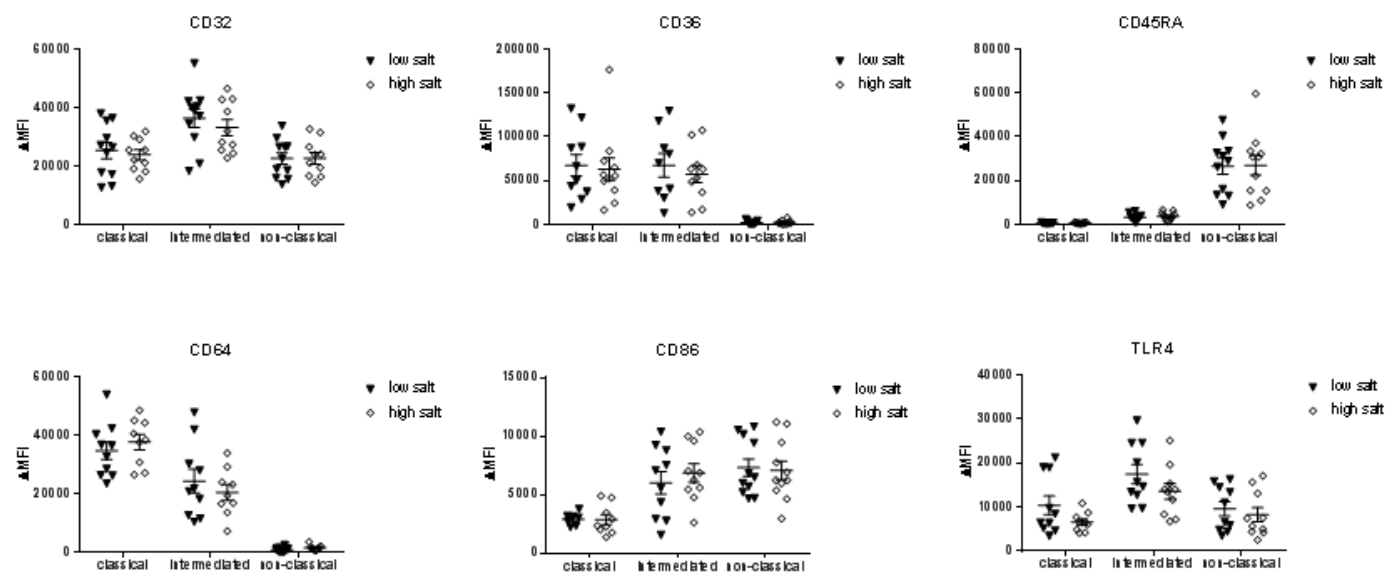
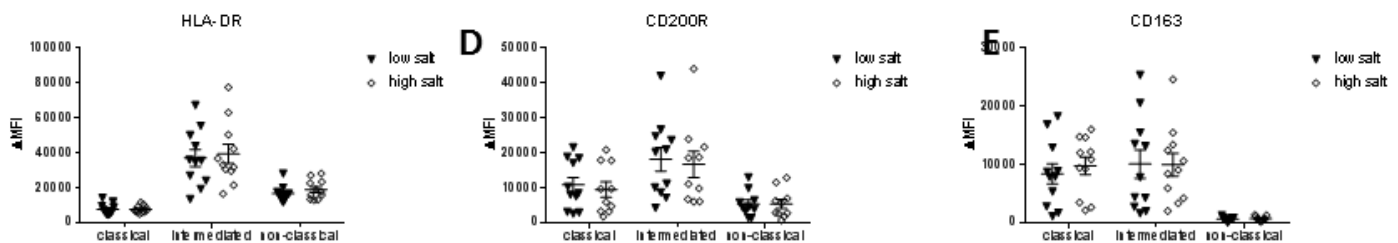
**Supplemental Table 3.** Measured parameters after low salt diet and after high salt diet in *study II*. Data are presented as mean (SD). BP, blood pressure. MAP, mean arterial pressure. eGFR, estimated glomerular filtration rate.

<b>Healthy males (n=12)</b>			
	<b>LSD</b>	<b>HSD</b>	<b>p-value</b>
Weight (kg)	74.0 (6.6)	76.5 (6.7)	<0.001
<b>Plasma</b>			
Sodium (mmol/L)	137.5 (1.6)	140.3 (1.8)	<0.01
Potassium (mmol/L)	3.9 (0.3)	3.9 (0.2)	0.76
Chloride (mmol/L)	99.6 (1.5)	103.3 (2.0)	<0.001
Bicarbonate (mmol/L)	25.5 (2.0)	25.4 (1.6)	0.88
Creatinine (umol/L)	83.9 (10.0)	77.0 (9.2)	<0.001
eGFR (CKD-EPI)	111.7 (14.1)	119.9 (11.7)	<0.01
Osmolality (mosm/kg)	284.9 (3.1)	289.6 (3.8)	<0.01
Glucose (mmol/L)	4.9 (0.3)	5.0 (0.4)	0.70
<b>24h urine</b>			
Volume (ml/24h)	1702 (551)	1909 (544)	0.24
Osmolality (mosm/kg)	430.9 (164.4)	743.7 (164.8)	<0.001
Creatinine (mmol/24h)	15.8 (2.4)	17.3 (2.5)	<0.05
Sodium (mmol/24h)	19.1 (9.5)	340.8 (104)	<0.001



**Supplemental Figure 1. Immunohistochemical staining with anti-CD68 and anti-CD163 in human skin.**

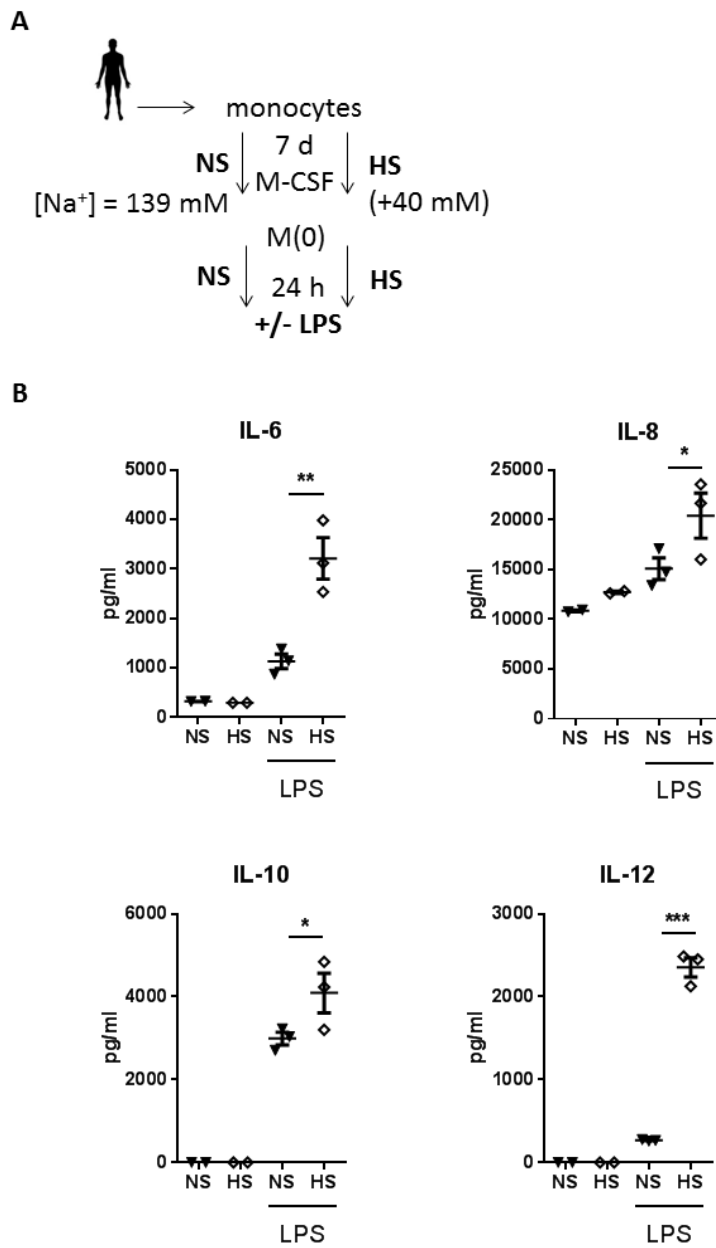
Anti-CD163 (blue) repeatedly proved to be more abundant than anti-CD68 (red). In some areas anti-CD68 staining was absent, but clearly a staining for anti-CD163 was present; examples of such regions are indicated by the black arrow heads.

**A****B****C****D**

**Supplemental figure 2. Effects of a HSD on monocyte chemokine receptors, adhesion molecules, activation molecules, M2-associated markers, and scavenger receptors.**

The expression of various surface markers on the monocyte subsets was calculated as  $\Delta\text{MFI} = [\text{median fluorescence intensity}]_{\text{positive staining}} - [\text{median fluorescence intensity}]_{\text{isotype control}}$ .

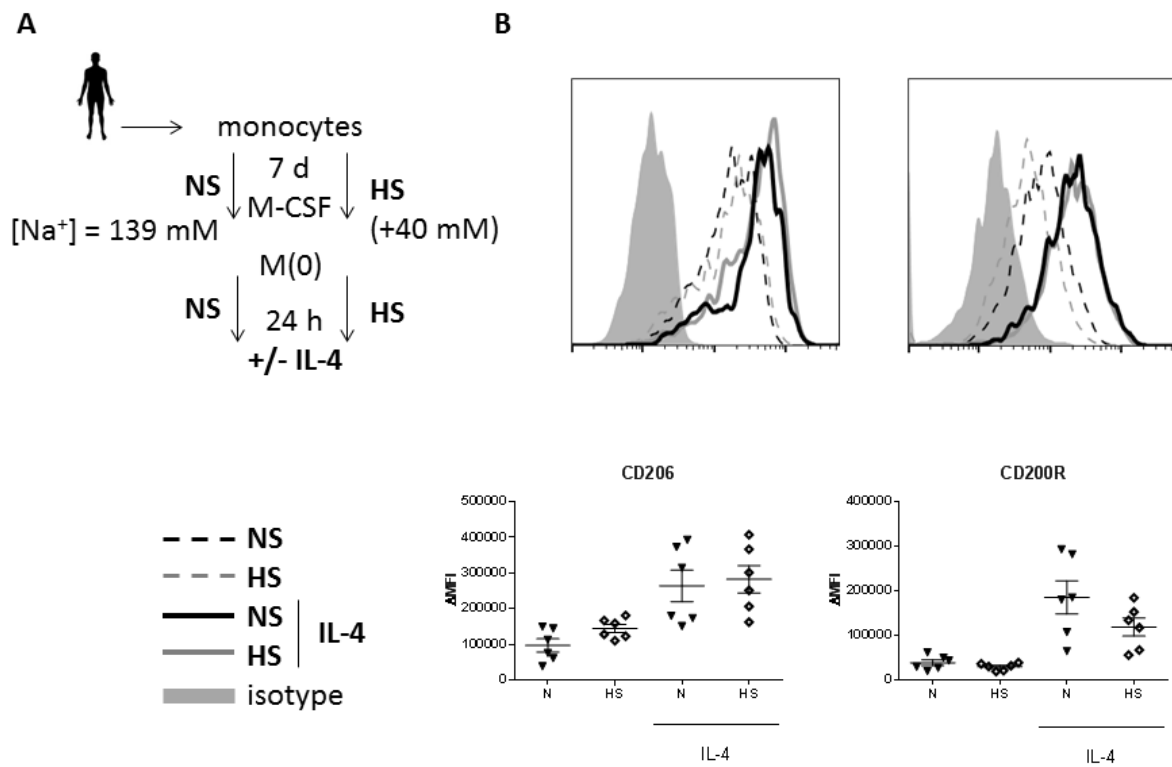
(A) HSD has no effect on monocyte chemokine receptors CCR5, CCR7, and CX3CR1. (B) HSD has no effect on monocyte adhesion molecules CD11b, CD11c, CD18, CD29, CD49d, and CD62L. (C) HSD has no effect on monocyte activation molecules CD32, CD36, CD45RA, CD64, CD86, TLR4, and HLA-DR. (D) HSD has no effect on monocyte M2-associated marker CD200R. (E) HSD has no effect on monocyte scavenger receptor SRA. HSD, high salt diet.



**Supplemental figure 3. High salt boosts LPS-induced cytokine secretion in macrophages *in vitro*.**

(A) Schematic overview of *in vitro* stimulation of macrophages. (B) HS increases secretion of IL-6, IL-8, IL-10 and IL-12 upon LPS exposure.





**Supplemental figure 4. High salt does not reduce IL-4 stimulated surface expression of CD206 and CD200R in macrophages *in vitro*.** (A) Schematic overview of *in vitro* stimulation of macrophages. (B) HS does not reduce surface expression of CD206 and CD200R.