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Title: Changing glucocorticoid action: 11 β -hydroxysteroid dehydrogenase type 1 in acute and chronic inflammation

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1 Title: **Changing glucocorticoid action: 11 β -hydroxysteroid**
2 **dehydrogenase type 1 in acute and chronic inflammation.**

3
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18
19
20 Disclosures: Jonathan R Seckl holds IP on the use of 11 β -HSD1 inhibitors in
21 diabetes, atherosclerotic disease and age-associated cognitive
22 impairment. None of the other authors have anything to disclose.

23
24 Abbreviations: 11 β -HSD, 11 β -hydroxysteroid dehydrogenase; H6PD, hexose-6-
25 phosphate dehydrogenase; TNF- α , tumour necrosis factor- α ; LPS,
26 lipopolysaccharide; IL-, interleukin, C/EBP, CCAAT/enhancer
27 binding protein; NF- κ B, nuclear factor kappa-light-chain-enhancer
28 of activated B cells; EGR-1, early growth response-1; HPA,
29 hypothalamic-pituitary-adrenal; MCP, monocyte chemotactic
30 protein; VCAM, vascular cell adhesion molecule.

31
32 Keywords: glucocorticoid, mineralocorticoid, 11 β -hydroxysteroid
33 dehydrogenase, macrophage, inflammation, arthritis

34
35
36
37 **ABSTRACT**

38 Since the discovery of cortisone in the 1940s and its early success in treatment of
39 rheumatoid arthritis, glucocorticoids have remained the mainstay of anti-inflammatory
40 therapies. However, cortisone itself is intrinsically inert. To be effective, it requires
41 conversion to cortisol, the active glucocorticoid, by the enzyme 11 β -hydroxysteroid
42 dehydrogenase type 1 (11 β -HSD1). Despite the identification of 11 β -HSD in liver in
43 1953 (which we now know to be 11 β -HSD1), its physiological role has been little
44 explored until recently. Over the past decade, however, it has become apparent that
45 11 β -HSD1 plays an important role in shaping endogenous glucocorticoid action. Acute
46 inflammation is more severe with 11 β -HSD1-deficiency or inhibition, yet in some
47 inflammatory settings such as obesity or diabetes, 11 β -HSD1-deficiency/inhibition is
48 beneficial, reducing inflammation. Current evidence suggests both beneficial and
49 detrimental effects may result from 11 β -HSD1 inhibition in chronic inflammatory
50 disease. Here we review recent evidence pertaining to the role of 11 β -HSD1 in
51 inflammation.

52

53 *Introduction*

54 The discovery of the anti-inflammatory effects of cortisone, a glucocorticoid hormone,
55 by Hench and colleagues in the 1940s, opened the door to the longest and most
56 successful drug development programme in history. Glucocorticoids remain the most
57 widely prescribed treatment for inflammatory disease. They potently affect both
58 immune and non-immune cells, shaping their responses. Glucocorticoid actions are
59 highly dependent on context and can be very different during acute and chronic
60 inflammation. In the short term at least, many of their effects promote the resolution of
61 inflammation. Several years ago, we hypothesised that the glucocorticoid metabolising
62 enzyme, 11 β -hydroxysteroid dehydrogenase type 1 (11 β -HSD1), is induced early
63 during an inflammatory response and shapes its subsequent trajectory [1]. How well
64 has that hypothesis stood the test of time? Reasonably well as it turns out, but not in
65 quite the way we had envisaged.

66

67 *Glucocorticoids and inflammation*

68 Synthetic glucocorticoids exert potent anti-inflammatory and immunosuppressive
69 effects and are widely prescribed to treat both acute and chronic inflammation. Yet the
70 well known side effects of glucocorticoid excess include type 2 diabetes, visceral
71 obesity, hypertension and atherosclerosis which are themselves, somewhat
72 paradoxically, inflammatory conditions. Quite how glucocorticoids provoke
73 inflammatory metabolic diseases at the same time as suppressing chronic inflammatory
74 conditions such as rheumatoid arthritis or inflammatory bowel disease remains unclear.
75 It is likely to involve more complex mechanisms than the commonly held view that the
76 “adverse” metabolic effects involve gene activation by glucocorticoid receptor (GR),
77 whereas the “beneficial” anti-inflammatory effects rely on gene repression. Fully
78 understanding how glucocorticoids cause “metabolic inflammation” will be crucial for
79 the development and optimal exploitation of future anti-inflammatory therapies, which
80 could manipulate glucocorticoid action in a more sophisticated manner than current
81 therapies.

82

83 Understanding the role of endogenous glucocorticoids during inflammation is key to
84 achieving this aim. Endogenous glucocorticoids are vital to survive trauma or certain
85 bacterial infections; they suppress pro-inflammatory cytokine production, binding to
86 GR in immune cells to prevent potentially lethal overshoot of immune responses [2, 3].
87 Acutely, circulating pro-inflammatory cytokines are a potent stimulus to the
88 hypothalamic-pituitary-adrenal (HPA) axis to increase endogenous glucocorticoid
89 production [4, 5]. However, this normal response is lost or attenuated in chronic
90 inflammation [6]. In this respect, the treatment of chronic inflammatory disease with
91 exogenous glucocorticoids can be regarded as replacement therapy for an inadequate
92 endogenous glucocorticoid response [7].

93

94 Acute inflammation is an immediate response of the body to injury or infection that
95 serves to remove the injurious stimulus, then restore homeostasis by removal of dead
96 and damaged cells/tissues and engagement of repair processes. It is initiated at the site
97 of injury by the release of proinflammatory mediators such as bioactive amines, lipids
98 and cytokines: typically tumour necrosis factor (TNF)- α and interleukin (IL)-1. These
99 cause vasodilation, increase vascular permeability allowing exudation of plasma, and
100 elicit leukocyte recruitment, activation and emigration from the microcirculation to the
101 damaged tissue. The initial response is typically predominated by neutrophils, which
102 are replaced by monocytes/macrophages during the resolution and repair stages.
103 Resolution of acute inflammation requires the engagement of mechanisms early in the
104 inflammatory response that shape the subsequent resolution (reviewed, [8-10]). Chronic

105 inflammation results from persistence of the initiating stimulus with associated
106 lymphocyte and macrophage activation. Excessive tissue damage contributes to
107 continuing inflammation, failure of resolution and dysregulated repair processes such
108 as angiogenesis and fibrosis and can thus form a “vicious” cycle. Whilst acute
109 inflammation frequently occurs and is contained entirely at the local level, chronic
110 inflammation invariably involves a systemic response.

111
112 Glucocorticoids limit acute inflammation. They repress a large number of
113 proinflammatory genes, including pro-inflammatory cytokines and chemokines, cell
114 adhesion molecules and enzymes involved in the initiation and/or maintenance of
115 inflammation, many of which are over-expressed in chronic non-resolving
116 inflammation. Conversely, they activate a number of genes encoding anti-inflammatory
117 mediators, such as IL-10 and annexin I (reviewed, [11-13]). Thus, acutely,
118 glucocorticoids inhibit the initial vasodilation and increased vascular permeability
119 during inflammation. They also alter the balance between survival and apoptosis of
120 leukocytes as well as their distribution between the circulation and immune tissues and
121 they decrease leukocyte emigration into sites of injury [13-18]. Importantly,
122 glucocorticoids potently influence the differentiation and phenotype of immune cells,
123 especially monocytes/macrophages and T lymphocytes, thereby polarising, or shaping,
124 immune responses [19]. Glucocorticoid treatment of human monocytes promotes an
125 anti-inflammatory, pro-resolution phenotype, characterised by high migratory and
126 phagocytic capacity, expression of CD163 (haemoglobin scavenger receptor) and high
127 production of IL-10 [20-23]. Similarly, in mice, pro-resolving macrophage functions
128 are enhanced by glucocorticoid treatment [24, 25], thus shaping the trajectory of an
129 inflammatory response and its outcome. Because glucocorticoids inhibit production of
130 “Th1” cytokines, which promote a cell-mediated immune response (activation of
131 phagocytes, antigen-specific T lymphocytes) whilst preserving or promoting “Th2”
132 cytokine production (aiding antibody production), they also shape the adaptive immune
133 response.

134
135 Most research on the anti-inflammatory actions of glucocorticoids has utilised
136 dexamethasone, a potent synthetic glucocorticoid with powerful immunosuppressive
137 properties. However, the endogenous glucocorticoids, cortisol (the main glucocorticoid
138 in humans) and corticosterone (in rats and mice), are immunomodulatory rather than
139 immunosuppressive [14, 26], particularly when administered at physiologically relevant
140 concentrations. Indeed, low doses of corticosterone stimulate whereas higher doses
141 suppress macrophage activity [27]. This could, in part, reflect the higher affinity
142 binding of endogenous glucocorticoids to the mineralocorticoid receptor (MR)
143 (dexamethasone poorly activates MR [28]) than to GR as both are expressed in
144 macrophages [27, 29, 30]. However, whereas knock-down or antagonism of GR in
145 macrophages abrogates responses to both high and low doses of corticosterone, knock-
146 down or antagonism of MR has little effect [27], suggesting GR-mediated effects, at
147 least in the rat macrophages tested. The interplay between GR and MR in macrophage
148 function and polarisation is likely to be complex (see below).

149
150 *11 β -hydroxysteroid dehydrogenases modulate glucocorticoid action*
151 Endogenous glucocorticoids differ from dexamethasone in another important respect;
152 dexamethasone is not inactivated by 11 β -HSD activity [31] whereas endogenous
153 glucocorticoids are substrates for the 11 β -HSDs, which are important modulators of
154 physiological glucocorticoid action [32]. The 11 β -HSD “shuttle” interconverts active
155 glucocorticoids (cortisol, corticosterone) with their 11-keto forms (cortisone, 11-
156 dehydrocorticosterone), which bind poorly to receptors and are therefore intrinsically
157 inert. In intact cells, 11 β -HSD1 exhibits oxo-reductase activity, converting cortisone

158 and 11-dehydrocorticosterone into active cortisol and corticosterone respectively,
159 increasing intracellular glucocorticoid levels. In contrast, 11 β -HSD2 is exclusively a
160 dehydrogenase, inactivating cortisol and corticosterone. Expression of 11 β -HSD2 is
161 largely restricted to mineralocorticoid-target tissues, most notably the distal nephron of
162 the kidney where it protects the non-selective MR from activation by glucocorticoids,
163 conferring aldosterone-specificity upon MR, which is otherwise a high affinity
164 glucocorticoid receptor [33, 34]. Of the synthetic glucocorticoids in widespread use as
165 anti-inflammatory drugs, it is worth noting that some, including
166 prednisone/prednisolone, are excellent substrates for the 11 β -HSDs.

167
168 11 β -HSD1 is widely expressed, including in immune cells, where its activity is
169 dynamically regulated depending on cell activation state (reviewed, [35]). 11 β -HSD1 is
170 up-regulated upon activation of monocytes/macrophages, neutrophils or lymphocytes
171 [35, 36] (and see Figure 1). Circulating leukocytes in mice and healthy humans do not
172 express 11 β -HSD2 [1, 37]. Both 11 β -HSD isozymes are regulated by pro-inflammatory
173 signalling in non-immune cells (see below for details).

174
175 *11 β -HSD1 expression in monocytes/macrophages depends on cell activation state*
176 Monocytes and macrophages are essential during an inflammatory response. In
177 response to diverse environmental signals, “resting” or naïve macrophages adopt
178 distinct phenotypes. These are broadly categorised based on *in vitro* experiments into
179 two states, M1 (or classically activated) and M2 (or alternatively activated) (reviewed
180 [38, 39]). M1 macrophages, induced by interferon- γ and Toll-like receptor (TLR)
181 activation (eg by lipopolysaccharide, LPS), are vital for host defence, expressing pro-
182 inflammatory cytokines, inducible nitric oxide synthase (iNOS) and demonstrating
183 strong microbicidal activity. M2 macrophages, polarised with IL-4 and/or IL-13,
184 restore homeostasis in the repair phase of inflammation. They are also vital for parasite
185 elimination. Other stimuli induce M2-like anti-inflammatory phenotypes, distinct from
186 IL-4/IL-13 polarised macrophages. Macrophage phenotype *in vivo* may be more
187 complex and heterogeneous [40], especially macrophages with M2-like characteristics,
188 reflecting the diversity of signalling and context *in vivo*. Glucocorticoids restrain M1
189 macrophages, dampening pro-inflammatory cytokine expression, and in naïve
190 monocytes/macrophages, induce a highly phagocytic, highly motile, M2-like phenotype
191 [21, 24, 41]. Conditional deletion of GR in macrophages increases pro-inflammatory
192 cytokine production and mortality following LPS administration [3, 42]. Conversely,
193 conditional deletion of MR in macrophages promotes polarisation to an alternatively
194 activated (M2) phenotype [43], suggesting a possible reciprocal relationship between
195 GR and MR activation in macrophages. There is therefore considerable potential for
196 11 β -HSD1 (which can potentially supply ligand to either receptor) to modulate
197 monocyte/macrophage phenotype by increasing intracellular glucocorticoid levels, even
198 in the absence of elevated circulating glucocorticoid levels.

199
200 Expression of 11 β -HSD1 is low in circulating mouse leukocytes but is higher in
201 macrophages [44]. Though negligible in non-stimulated human monocytes, 11 β -HSD1
202 expression is induced upon differentiation into resting or naïve (ie unstimulated)
203 macrophages [37]. M1 polarisation of naïve macrophages with LPS further induces
204 11 β -HSD1 (Figure 1). In contrast, polarisation to an M2 phenotype with IL-4 has little
205 effect on 11 β -HSD1 expression [45, 46]. However, in human monocytes differentiated
206 into macrophages in the presence of IL-4 (which may induce a distinct anti-
207 inflammatory macrophage phenotype from M2 polarisation of resting macrophages),
208 11 β -HSD1 activity is as high or higher than in M1, and is further increased by
209 peroxisome proliferator-activated receptor (PPAR)- γ activation [47]. In contrast, in
210 mouse bone marrow-derived macrophages (resting macrophages) PPAR- γ agonists

211 down-regulate 11! -HSD1 expression [47]; whether this reflects a mouse/human species
212 difference or the different macrophage phenotypes (resting mouse macrophages *versus*
213 human macrophages differentiated in the presence of IL-4) is currently unclear.
214 Nevertheless these studies illustrate a complex dependence of 11! -HSD1 expression
215 upon macrophage activation state. The significance is currently unknown but might
216 reflect (or influence) differences in energy metabolism between glycolytic M1 and
217 oxidative M2 macrophages [48, 49]. Recent evidence suggests manipulation of glucose
218 metabolism in macrophages directly alters polarisation [49]. Whether alterations in
219 11! -HSD1 expression influence macrophage glucose metabolism, for example through
220 the coupling of 11! -HSD1 oxo-reductase activity to hexose 6-phosphate activity in the
221 endoplasmic reticulum (see below) is an important question to address as it may
222 directly affect polarisation or the extent of activation of macrophages. Dynamic
223 regulation of 11! -HSD1 in macrophages could therefore be crucial to the ability to
224 shape an ongoing inflammatory response, either through intracellular regeneration of
225 glucocorticoids or indirectly by diversion of glucose-6-phosphate (Figure 2). Evidence
226 for dynamic regulation of 11! -HSD1 during an inflammatory response *in vivo* comes
227 from the rapid induction of 11! -HSD1 activity in neutrophils and
228 monocytes/macrophages during sterile peritonitis in mice; 11! -HSD1 activity decreases
229 as the inflammation resolves [1, 36]. The latter is possibly an active process; 11! -
230 HSD1 activity is rapidly down-regulated in macrophages that have phagocytosed
231 apoptotic neutrophils [35], a highly pro-resolution process [50]. This reasoning led to
232 the hypothesis that the early induction of 11! -HSD1 in macrophages increases
233 glucocorticoid action within these cells, promoting an anti-inflammatory phenotype and
234 leading to more rapid resolution of inflammation [1, 51].

235

236 *11! -HSD1 in acute inflammation; regulation*

237 In most animal models of acute inflammation 11! -HSD1 activity is up-regulated in the
238 inflamed tissue, whereas 11! -HSD2 (if expressed at all) is down-regulated. This is true
239 of the inflamed colon and the arthritic joint [52-54], but not the vasculature [55]. This
240 switch in the balance of 11! -HSD1 and 2 activities is predicted to increase
241 paracrine/autocrine glucocorticoid action, though this has not been directly tested.
242 Induction of 11! -HSD1 (and repression of 11! -HSD2) at inflamed sites is probably due
243 to local release of the pro-inflammatory cytokines IL-1 and TNF! which stimulate
244 transcription of the 11! -HSD1 gene promoter through increased binding of the
245 transcriptional regulators CCAAT/enhancer binding protein (C/EBP)! and nuclear
246 factor kappa-light-chain-enhancer of activated B cells (NF! B) [56-58] and repress the
247 11! -HSD2 gene promoter through an early growth response (EGR)-1 and NF! B-
248 dependent mechanism [59]. Normally glucocorticoids antagonise TNF! or IL-1
249 action, but they act together with the pro-inflammatory cytokines to synergistically
250 increase 11! -HSD1 expression in a variety of cell types [60-63]. This is predicted to
251 amplify the effect of glucocorticoid within a given cell or tissue, more rapidly
252 promoting the repair and resolution phase. Whether 11! -HSD1 expression in
253 inflammatory cells is regulated by similar mechanisms is an interesting question.
254 Neither TNF! nor IL-1 affect 11! -HSD1 activity in monocytes [37] and the
255 signalling pathways that regulate macrophage 11! -HSD1 expression have not been
256 characterised. C/EBP! , a key regulator of 11! -HSD1 transcription in a variety of cell
257 types [56, 57, 64-68], mediates M2 polarisation and arginase expression [69] yet also
258 plays a role in pro-inflammatory cytokine expression in M1 macrophages [70].
259 However, genetic deletion of C/EBP! abolishes both the liver-enriched inhibitor
260 protein (LIP) and liver-enriched activator protein (LAP) C/EBP! isoforms, the balance
261 of which potently influences 11! -HSD1 mRNA levels *in vivo* [71] and also regulates
262 osteoclast differentiation [72], a process akin to macrophage differentiation. The
263 C/EBP! -LIP:LAP ratio is regulated by mTOR [73], an integrator of cellular nutrient

264 and energy metabolism, that is downstream of phosphatidylinositol 3-kinase (PI3K)
265 and Akt, both capable of polarising macrophages [74, 75]. Plausibly, the C/EBP β -
266 LIP:LAP ratio differs according to the activating stimulus and may govern the
267 expression level of 11 β -HSD1 in polarised macrophages. The coupling within the
268 endoplasmic reticulum of 11 β -HSD1 activity to the supply of NADP(H) cofactor
269 generated by hexose-6-phosphate dehydrogenase (H6PD) [76-78] is particularly
270 intriguing in this respect, as it raises the possibility that cellular glucose availability and
271 flux through the endoplasmic reticulum pentose phosphate pathway (the first 2 steps of
272 which are catalysed by H6PD) controls 11 β -HSD1 activity [79] which may therefore
273 differ irrespective of expression levels in M1 and M2 macrophages.

274

275 *11 β -HSD1 in acute inflammation; function*

276 Based on the expression of 11 β -HSD1 in macrophages, its induction early during an
277 inflammatory response and the well-known anti-inflammatory effects of
278 glucocorticoids, it was anticipated that 11 β -HSD1 deficiency or inhibition would
279 attenuate local glucocorticoid production and thus worsen acute inflammation. This is
280 indeed what is seen in 11 β -HSD1-deficient (*Hsd11b1*^{-/-}) mice, with more severe LPS-
281 induced endotoxaemia (classically repressed by glucocorticoids [3, 80]), an earlier
282 onset of inflammation in the K/BxN serum transfer model of inflammatory arthritis and
283 more inflammatory cells (both neutrophils and monocytes/macrophages) recruited in
284 sterile peritonitis or pleuritis and in the injured myocardium following myocardial
285 infarction [81-83] (and see Figure 2). This increase in inflammation could reflect
286 greater recruitment and/or delayed clearance/apoptosis of neutrophils [1, 84]. In support
287 of the latter, *Hsd11b1*^{-/-} mice show delayed macrophage acquisition of phagocytic
288 capacity for apoptotic neutrophils as well as an increase in the number of free apoptotic
289 neutrophils during sterile peritonitis, although surprisingly the peritonitis resolves at the
290 same time as in wild-type mice [1]. Also surprising was the finding that despite the
291 increased inflammation early following myocardial infarction or possibly because of it,
292 heart function post-infarction is much better preserved in *Hsd11b1*^{-/-} mice than in
293 controls. Underlying the improved recovery from myocardial infarction is an increased
294 angiogenic response to injury [85], probably as a consequence of an earlier
295 accumulation of reparative M2 (Ym1⁺) macrophages and higher levels of the pro-
296 angiogenic cytokine IL-8 in the hearts of *Hsd11b1*^{-/-} mice [82]. It will be important to
297 determine how generally this accelerated switch in macrophage phenotype from M1 to
298 M2 applies to inflammation in *Hsd11b1*^{-/-} mice; so far it has only been reported in
299 myocardial infarction and M2-like polarisation is not a general feature of 11 β -HSD1-
300 deficient macrophages, at least *in vitro* [1, 81] or *in vivo*, in adipose tissue of high fat
301 fed obese mice [86]. Despite the lack of detectable difference in adipose tissue
302 macrophage phenotype, an increased angiogenic response to tissue ischemia is also
303 seen in adipose tissue of obese *Hsd11b1*^{-/-} mice and underlies their resistance to some
304 of the adverse metabolic consequences of obesity [87], suggesting the pro-angiogenic
305 phenotype may be at least partly independent of macrophages.

306 How is the improved recovery of *Hsd11b1*^{-/-} mice from inflammation following
307 myocardial infarction reconciled with our original hypothesis? As predicted by the
308 hypothesis, deficiency in 11 β -HSD1 causes greater release of pro-inflammatory
309 cytokines from LPS-treated macrophages [1, 81], suggesting an exaggerated M1
310 macrophage phenotype. However, the earlier switch to an M2 phenotype was
311 unexpected. Whether this reflects a switch to M2 phenotype *in situ* or recruitment of a
312 distinct subset of monocytes is currently unknown. It is possible that this is a
313 consequence of prolonged activation of the HPA axis in *Hsd11b1*^{-/-} mice. However,
314 these mice show little perturbation of plasma corticosterone levels, even following

315 stress, on this genetic background [88], so the earlier switch is unlikely to be mediated
316 by plasma glucocorticoids. Moreover, as discussed above, intracellular amplification of
317 glucocorticoid signalling by 11 β -HSD1 is predicted to accelerate repair and resolution
318 processes, not attenuate them. Several key factors implicated in macrophage
319 polarisation [89] are differentially expressed in *Hsd11b1*^{-/-} mice. The Src homology 2-
320 containing inositol-5'-phosphatase (SHIP)-1 negatively regulates the PI3K pathway. It
321 represses the generation of M2 macrophages [74] yet restrains LPS-induced (M1)
322 activation of bone marrow-derived (naïve) macrophages [90]. Moreover, elevated
323 SHIP1 expression induces endotoxin tolerance [90] with reduced pro-inflammatory
324 cytokine production with subsequent endotoxin challenge [90]. The increased LPS-
325 responsiveness of thioglycollate elicited peritoneal (TEP) macrophages from *Hsd11b1*^{-/-}
326 mice was attributed to elevated SHIP1 levels as a consequence of higher levels of
327 TGF β 1 [81] though SHIP1 levels appear to decrease more rapidly following LPS in
328 *Hsd11b1*^{-/-} macrophages than in wild-type. In splenic macrophages, basal SHIP1 levels
329 are normal in *Hsd11b1*^{-/-} mice, but unlike wild-type splenic macrophages, those from
330 *Hsd11b1*^{-/-} mice fail to down-regulate SHIP1 following LPS [81]. Whether this induces
331 endotoxin tolerance [90] to a greater extent in *Hsd11b1*^{-/-} macrophages is something
332 that requires testing. Thus, SHIP1 appears abnormally regulated in *Hsd11b1*^{-/-}
333 macrophages, though why is currently unclear. Nevertheless, these somewhat confusing
334 data illustrate that M1/M2 macrophage polarisation in *Hsd11b1*^{-/-} mice may be highly
335 dependent upon the macrophage population and context.

336 Hypoxia-inducible factor (HIF1)- α , which promotes M1 polarisation, is decreased in
337 adipose tissue of *Hsd11b1*^{-/-} mice, whereas levels of PPAR α (which promotes the M2
338 phenotype) are increased [87, 91]. Whether these factors are differentially expressed in
339 macrophages of *Hsd11b1*^{-/-} mice will be important to determine.

340 The outcome of acute inflammation is not invariably improved in *Hsd11b1*^{-/-} mice. At
341 the stage when arthritis has largely resolved in wild-type mice following K/BxN serum
342 transfer, joints of *Hsd11b1*^{-/-} mice show greater periarticular fibrosis, more extensive
343 exostoses and ganglion cyst formation. Following carageenan-induced pleurisy,
344 *Hsd11b1*^{-/-} mice show persistence of inflammation at a stage when it is resolving in
345 wild-type controls, as well as lymphoid aggregates within the lung and formation of
346 fibrous adhesions between lung lobes, the latter not present in control mice [83].
347 Whether these disadvantageous features result from greater inflammation in *Hsd11b1*^{-/-}
348 mice, an earlier switch to a pro-repair (pro-fibrotic) M2 phenotype, a greater response
349 of the non-immune tissue or a combination of all of these will be an interesting
350 question for the future. Moreover, the consequences of more extended inflammation
351 will be interesting to determine. The preliminary findings in arthritis and carageenan
352 induced pleurisy suggest that 11 β -HSD1-deficiency or inhibition may aggravate
353 diseases associated with a dysregulated angiogenic and pro-fibrotic phenotype,
354 including rheumatoid arthritis.

355

356 *Chronic inflammation*

357 Chronic inflammation results from a failure to resolve acute inflammation.
358 Atherosclerosis, diabetes, metabolic syndrome and Alzheimer's disease are all now
359 recognised as chronic inflammatory diseases. Even simple obesity is frequently
360 associated with low level chronic inflammation within the adipose tissue. The elevation
361 in systemic pro-inflammatory cytokines during chronic inflammation might be
362 expected to activate the HPA axis. However, plasma cortisol is normal in both the
363 "classic" inflammatory diseases (rheumatoid arthritis, inflammatory bowel disease etc)
364 and in the "metabolic" inflammatory diseases (atherosclerosis, metabolic syndrome,
365 diabetes), at least until these become complicated by additional pathologies. HPA axis
366 activity may be elevated in metabolic inflammation, with increased clearance of

367 glucocorticoids maintaining normal plasma cortisol levels [92] but possibly increasing
368 plasma cortisone levels (and thus 11 β -HSD1 substrate), though this has only been
369 indirectly measured. In rheumatoid arthritis and other inflammatory diseases however,
370 the HPA axis appears relatively suppressed, especially given the level of systemic
371 inflammation expected to activate the axis [4, 93]. Edwards has recently hypothesised
372 that this apparent deficiency in HPA activation is a result of the systemic increase in
373 TNF- α in chronic inflammation inducing a widespread increase in 11 β -HSD1
374 expression, including in the hypothalamus, thus amplifying negative feedback by
375 glucocorticoids on the HPA axis [93]. Whether this is indeed the case requires
376 experimental testing, but consistent with this hypothesis, whole body conversion of
377 cortisone to cortisol (relative to cortisol to cortisone) is increased in patients with
378 inflammatory disease [94] suggesting altered balance of 11 β -HSD activities in favour
379 of 11 β -reductase (11 β -HSD1).

380

381 *Metabolic Syndrome, type 2 diabetes and atherosclerosis*

382 11 β -HSD1 deficiency or inhibition is metabolically beneficial in rodent models of diet-
383 induced obesity or diabetes. It improves hepatic and adipose insulin sensitivity,
384 attenuates hepatic gluconeogenesis, skews to a “cardioprotective” plasma lipid profile,
385 shifts hepatic lipid metabolism from lipogenesis to fatty acid oxidation and causes a
386 preferential gain of peripheral adipose tissue at the expense of visceral [86, 91, 95-101]
387 (and see Figure 2). Similarly, in patients with type 2 diabetes, 11 β -HSD1 inhibition
388 lowers plasma glucose and lipids, consistent with rodent studies. It also modestly
389 reduces blood pressure in human hypertension [102-104]. Intriguingly, an 11 β -HSD1
390 inhibitor more effectively improved glucose homeostasis in obese mice when
391 administered close to the time of the diurnal peak of plasma glucocorticoid levels [105].
392 Given that 11 β -HSD1 mRNA probably does not vary with the circadian rhythm [105,
393 106] (though one study suggests it may in rats [107]), this is much more likely to reflect
394 high 11 β -HSD1 substrate levels at peak HPA axis activity [108]. Indeed, 11 β -HSD1
395 may contribute to normal circadian control of the HPA axis, at least in some genetic
396 backgrounds [88, 108]. 11 β -HSD1 is expressed in the paraventricular nucleus of the
397 human hypothalamus, suggesting a conserved role in HPA axis regulation [109].

398

399 Recent data suggest that the liver is not the sole or even predominant target of the
400 metabolically beneficial effects of 11 β -HSD1-deficiency or inhibition; conditional
401 deletion of 11 β -HSD1 in hepatocytes of mice produces only minimal improvements in
402 glucose homeostasis in diet-induced obesity [110]. Instead, increased glucocorticoid
403 activity in adipose tissue is implicated. In obese humans, numerous studies have
404 reported elevated 11 β -HSD1 expression in subcutaneous adipose tissue (reviewed [92])
405 and in human omental fat, 11 β -HSD1 expression correlates with adipocyte hypertrophy
406 [111, 112], itself associated with a more pro-inflammatory state [113, 114]. In mice, a
407 two to three-fold elevation of 11 β -HSD1 selectively in adipose tissue phenocopies the
408 metabolic syndrome, with central obesity, insulin resistance, dyslipidaemia and
409 hypertension [115, 116] whereas similar transgenic expression of 11 β -HSD2 in
410 adipocytes (it is not normally expressed in adipocytes), presumably lowering intra-
411 adipose glucocorticoid action, causes insulin sensitisation in high fat fed mice [117].

412

413 11 β -HSD1-deficiency protects against pro-inflammatory changes in adipose tissue in
414 obesity. Inflammatory cell (macrophages, lymphocytes) infiltration of mesenteric
415 adipose tissue is lower in high fat-fed 11 β -HSD1-deficient mice than in controls,
416 probably due to reduced adipocyte secretion of the pro-inflammatory chemokine,
417 monocyte chemoattractant-1 (MCP-1) [86]. This is associated with higher levels of
418 AMP-activated protein kinase activation in this depot [86], likely to contribute to the
419 maintained lipid oxidation with obesity [118] in 11 β -HSD1-deficiency. Whether these

420 changes are a cause or a consequence of the increase in angiogenesis and reduction in
421 hypoxia and fibrosis recently described in the adipose tissue of these mice [87] is an
422 interesting question. Adipose tissue hypoxia is associated with a local pro-
423 inflammatory environment and leads to fibrosis though not necessarily angiogenesis
424 [119-121], suggesting that it is the greater angiogenic response in *Hsd11b1*^{-/-} mice that
425 is protective against adipose tissue hypoxia and fibrosis. PPAR α mRNA levels are
426 higher and the pro-angiogenic response to PPAR α activation is much greater in
427 *Hsd11b1*^{-/-} adipocytes than in controls, placing the adipocyte at the heart of the
428 response. Whether there are also beneficial roles for macrophage and/or vascular 11 β -
429 HSD1 is important to determine.

430 As well as improving metabolic risk factors, deficiency in or inhibition of 11 β -HSD1
431 also reduces atherosclerosis and systemic inflammation and lowers macrophage and T
432 cell infiltration of atherosclerotic lesions in *Apoe*^{-/-} mice [122-124]. This is the converse
433 of what happens with 11 β -HSD2-deficiency, which is pro-inflammatory in the
434 endothelium and accelerates atherosclerosis in *Apoe*^{-/-} mice, an effect at least partly
435 mediated through activation of the MR as it is blocked by eplerenone, an MR
436 antagonist [125]. The atheroprotective effects of 11 β -HSD1-deficiency are likely to be
437 mediated through both systemic (reduced circulating monocyte chemotactic protein
438 (MCP)-1 and number of pro-inflammatory Ly6C^{hi} monocytes) and local (reduced aortic
439 vascular cell adhesion molecule (VCAM)-1 expression) mechanisms [124]. It is
440 interesting to speculate that reduced visceral adipose tissue inflammation may
441 contribute to the reduction in systemic inflammation – as in diet-induced obesity,
442 mesenteric adipose tissue MCP-1 mRNA levels are reduced in western diet-fed 11 β -
443 HSD1-deficient *Apoe*^{-/-} mice [124].

444

445 “Classic” inflammatory diseases - rheumatoid arthritis

446 If 11 β -HSD1-deficiency is beneficial in chronic “cardiometabolic inflammation”, what
447 of the classical inflammatory diseases, in which a glucocorticoid-insufficient state is
448 suggested and glucocorticoid therapy remains highly effective? Inevitably, studies in
449 animals are predominantly short term, modelling the disease, whereas the disease in
450 patients frequently reflects years of accumulated damage and inflammation. These
451 situations may be quite different. Nevertheless, accumulating evidence in both patients
452 and animal models is consistent with dysregulated 11 β -HSD1 in the inflamed joint in
453 rheumatoid arthritis as well as increased colonic expression of 11 β -HSD1 at sites of
454 inflammation in inflammatory bowel disease (reviewed [35]). So far, studies in
455 inflammatory bowel disease have gone little beyond observation, though they do
456 suggest that at least some of the increase in 11 β -HSD1 expression occurs in activated
457 lymphocytes that migrate from the inflamed colon to the draining lymph nodes [54].
458 Studies in human patients with rheumatoid arthritis suggest differential regulation of
459 11 β -HSDs in immune and mesenchymal cells. Comparison of cortisone and cortisol
460 levels in synovial fluid and serum suggest the balance favours intra-articular generation
461 of cortisol in the rheumatic joint [126] although it seems that even so, the overall
462 capacity to convert cortisone to cortisol may be reduced in the inflamed arthritic
463 synovium compared to non-inflamed. However, within inflamed rheumatic joints,
464 synovial inflammation still correlates with conversion of cortisone to cortisol [127].
465 This complex relationship probably reflects the balance between high expression of
466 11 β -HSD1 in synovial fibroblasts from arthritic patients (almost certainly as a result of
467 the pro-inflammatory cytokine environment) and expression of 11 β -HSD2 in synovial
468 macrophages from patients with rheumatoid arthritis [126-128]. This latter finding
469 accords with other studies identifying 11 β -HSD2 as a peripheral blood mononuclear
470 cell marker of early rheumatoid arthritis and highly expressed in the arthritic joint [129,
471 130]. 11 β -HSD2-positive macrophages have also been described in the lungs of
472 patients who died of acute respiratory distress syndrome [131]. Similar cells

473 (macrophages, lymphocytes) from healthy humans do not express 11 β -HSD2 [37, 130],
474 nor has 11 β -HSD2 been found in mouse leukocytes [1]. 11 β -HSD2 expression in
475 leukocytes may reflect a species difference between mouse and human, or could, in
476 humans, reflect an adaptive response to chronic inflammation. The biological reason
477 for this apparently pro-inflammatory change is unknown but it is likely to cause
478 resistance to endogenous glucocorticoids, which might be overcome by
479 pharmacological levels of synthetic glucocorticoids like prednisolone or bypassed with
480 non-metabolised synthetic glucocorticoids like dexamethasone.

481

482 What might 11 β -HSD1 inhibition do in chronic inflammatory disease? If the Edwards
483 hypothesis [93] is correct, then systemic inhibition of 11 β -HSD1, particularly if
484 administered during the night (in humans), should correct the HPA axis abnormality
485 and boost the plasma cortisol levels. This might be enough to dampen down some of
486 the inflammation, though 11 β -HSD1 inhibition would also deprive inflamed tissues of
487 the 11 β -HSD1-mediated increase in intracellular glucocorticoid levels. Moreover,
488 given that cortisol also activates MR (in the absence of 11 β -HSD2), this could further
489 exacerbate inflammation which could be particularly damaging within the vasculature
490 (see below). In chronic inflammatory disease, continuing tissue injury is frequently
491 associated with fibrosis and angiogenesis. Both may be exacerbated by 11 β -HSD1
492 inhibition. As mentioned above, 11 β -HSD1-deficient mice show an increased
493 angiogenic response to adipose tissue hypoxia, to ischaemia following myocardial
494 infarction, in wound healing and in sub-cutaneously implanted sponges [85, 87]. They
495 also show a pro-fibrotic response to pleural inflammation and following inflammatory
496 arthritis [83]. Whilst it is currently unclear whether the increased fibrosis in 11 β -HSD1-
497 deficient mice will resolve completely during recovery from inflammation, it is likely
498 that if the injurious stimulus persists, fibrosis will be more severe with 11 β -HSD1-
499 deficiency or inhibition. In continuing liver injury, a population of macrophages with
500 "M2"-like properties drives the fibrotic response, probably mediated at least in part
501 through TGF β 1 [40]. Higher macrophage expression of TGF β 1 with 11 β -HSD1-
502 deficiency [81] may be an important contributor to the pro-fibrotic phenotype of these
503 mice.

504

505 *Glucocorticoid receptor or mineralocorticoid receptor activation?*

506 Activation of MR, most notably in the heart and vasculature, has pro-inflammatory and
507 pro-fibrotic consequences [132, 133]. Unlike synthetic glucocorticoids, most of which
508 show selectivity for GR over MR, endogenous glucocorticoids bind with higher affinity
509 to MR than to GR. Thus, MR is usually considered near saturated at circulating
510 glucocorticoid levels, even at the diurnal nadir [134]. Aldosterone activates MR
511 irrespective of which cells it is expressed in, but cortisol activation of MR is normally
512 prevented if 11 β -HSD2 is co-expressed with MR. However, under conditions of
513 oxidative stress, endogenous glucocorticoids can activate MR, at least in the
514 cardiovascular system [135]. A crucial question therefore, central to the function of
515 11 β -HSD1, is which receptor binds the ligand it generates, GR or MR? This may differ
516 according to tissues. MR is absent from liver, so in this tissue, 11 β -HSD1 provides
517 ligand to GR. However, MR is expressed in some classical glucocorticoid targets,
518 including adipocytes and macrophages, normally in the absence of 11 β -HSD2, where it
519 presumably functions as a glucocorticoid receptor. A pro-inflammatory role for
520 glucocorticoid-activated MR is suggested; eplerenone treatment of *ob/ob* mice
521 prevented the obesity-associated increases in MCP-1, TNF- α and other inflammatory
522 markers in adipose tissue [136]. Whether the relevant cell is the adipocyte, however, is
523 unclear. Whereas MR activation (presumably by glucocorticoids) in macrophages
524 appears pro-inflammatory, macrophage-specific deletion of MR appears anti-
525 inflammatory - it causes M2 polarisation of macrophages [43] and reduces cerebral

526 infarct area following ischaemia in mice, concomitant with reduced expression of M1
 527 macrophage markers (TNF- α , IL-1, MCP-1 etc) but maintained M2 markers (Ym1,
 528 Arg1) [137]. Thus, the consequences of 11 β -HSD1-mediated glucocorticoid generation
 529 could differ greatly, depending on cellular oxidation/stress state and the relative levels
 530 of GR *versus* MR.

531
 532

533 SUMMARY AND CONCLUSIONS

534 Consistent with the adverse metabolic effects of glucocorticoid excess, 11 β -HSD1
 535 deficiency or inhibition is clearly beneficial in cardiometabolic disease. The extent to
 536 which this is dependent on inhibition/deficiency within inflammatory cells will be
 537 interesting to discover. Also, whether 11 β -HSD1 deficiency/inhibition is beneficial in
 538 other types of inflammation remains to be seen. Current evidence suggests that the
 539 acute response to injury is more severe. The subsequent recovery phase may depend on
 540 whether the injurious stimulus persists as in patients with rheumatoid arthritis (in which
 541 case 11 β -HSD1 deficiency/inhibition may worsen the disease), or whether recovery and
 542 tissue remodelling occur, as for example follows myocardial infarction (when 11 β -
 543 HSD1 deficiency/inhibition may aid recovery). The application of Cre/Lox technology
 544 to generate tissue- and cell-specific “knock-out” of 11 β -HSD1 will be invaluable in
 545 dissecting the contributions of immune cells, particularly macrophages and neutrophils,
 546 to the pro-angiogenic and pro-fibrotic phenotype. In the future, such studies could lead
 547 to better targeting of glucocorticoid therapy, perhaps even targeting macrophages
 548 separately from host tissues at specific temporal stages of disease. As already suggested
 549 [1], targeted delivery of inactive glucocorticoid precursors to macrophages might
 550 provide an effective future therapy for chronic inflammatory disease.

551
 552

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559 REFERENCES

- 560 [1] J.S. Gilmour, A.E. Coutinho, J.F. Cailhier, T.Y. Man, M. Clay, G. Thomas, H.J.
 561 Harris, J.J. Mullins, J.R. Seckl, J.S. Savill, K.E. Chapman, Local amplification of
 562 glucocorticoids by 11 β -hydroxysteroid dehydrogenase type 1 promotes
 563 macrophage phagocytosis of apoptotic leukocytes, *J Immunol* 176 (2006) 7605-
 564 7611.
- 565 [2] J.A. Brewer, B. Khor, S.K. Vogt, L.M. Muglia, H. Fujiwara, K.E. Haegele, B.P.
 566 Sleckman, L.J. Muglia, T-cell glucocorticoid receptor is required to suppress COX-
 567 2-mediated lethal immune activation, *Nat Med* 9 (2003) 1318-1322.
- 568 [3] S. Bhattacharyya, D.E. Brown, J.A. Brewer, S.K. Vogt, L.J. Muglia, Macrophage
 569 glucocorticoid receptors regulate Toll-like receptor-4-mediated inflammatory
 570 responses by selective inhibition of p38 MAP kinase, *Blood* 109 (2007) 4313-
 571 4319.
- 572 [4] G.P. Chrousos, The hypothalamic-pituitary-adrenal axis and immune-mediated
 573 inflammation, *N Engl J Med* 332 (1995) 1351-1362.
- 574 [5] A.V. Turnbull, C.L. Rivier, Regulation of the hypothalamic-pituitary-adrenal axis
 575 by cytokines: actions and mechanisms of action, *Physiol Rev* 79 (1999) 1-71.
- 576 [6] M.S. Harbuz, A.J. Chover-Gonzalez, D.S. Jessop, Hypothalamo-pituitary-adrenal
 577 axis and chronic immune activation, *Ann N Y Acad Sci* 992 (2003) 99-106.

- 578 [7] M. Cutolo, A. Sulli, C. Pizzorni, M.E. Secchi, S. Soldano, B. Seriola, R.H. Straub,
579 K. Otsa, G.J. Maestroni, Circadian rhythms: glucocorticoids and arthritis, *Ann N Y*
580 *Acad Sci* 1069 (2006) 289-299.
- 581 [8] J. Savill, I. Dransfield, C. Gregory, C. Haslett, A blast from the past: clearance of
582 apoptotic cells regulates immune responses, *Nat Rev Immunol* 2 (2002) 965-975.
- 583 [9] D.W. Gilroy, The endogenous control of acute inflammation - from onset to
584 resolution, *Drug Discovery Today: Therapeutic Strategies* 1 (2004) 313.
- 585 [10] C.N. Serhan, J. Savill, Resolution of inflammation: the beginning programs the
586 end, *Nat Immunol* 6 (2005) 1191-1197.
- 587 [11] P.J. Barnes, Anti-inflammatory actions of glucocorticoids: molecular mechanisms,
588 *Clin Sci (Lond)* 94 (1998) 557-572.
- 589 [12] K.A. Smoak, J.A. Cidlowski, Mechanisms of glucocorticoid receptor signaling
590 during inflammation, *Mech Ageing Dev* 125 (2004) 697-706.
- 591 [13] M. Perretti, A. Ahluwalia, The microcirculation and inflammation: site of action
592 for glucocorticoids, *Microcirculation* 7 (2000) 147-161.
- 593 [14] B.S. McEwen, C.A. Biron, K.W. Brunson, K. Bulloch, W.H. Chambers, F.S.
594 Dhabhar, R.H. Goldfarb, R.P. Kitson, A.H. Miller, R.L. Spencer, J.M. Weiss, The
595 role of adrenocorticoids as modulators of immune function in health and disease:
596 neural, endocrine and immune interactions, *Brain Res Rev* 23 (1997) 79-133.
- 597 [15] S.L. Planey, G. Litwack, Glucocorticoid-induced apoptosis in lymphocytes,
598 *Biochem Biophys Res Commun* 279 (2000) 307-312.
- 599 [16] J.D. Ashwell, F.W. Lu, M.S. Vacchio, Glucocorticoids in T cell development and
600 function, *Ann Rev Immunol* 18 (2000) 309-345.
- 601 [17] M.J. Herold, K.G. McPherson, H.M. Reichardt, Glucocorticoids in T cell apoptosis
602 and function, *Cell Mol Life Sci* 63 (2006) 60-72.
- 603 [18] A. McColl, S. Michlewska, I. Dransfield, A.G. Rossi, Effects of glucocorticoids on
604 apoptosis and clearance of apoptotic cells, *ScientificWorldJournal* 7 (2007) 1165-
605 1181.
- 606 [19] A.E. Coutinho, K.E. Chapman, The anti-inflammatory and immunosuppressive
607 effects of glucocorticoids, recent developments and mechanistic insights, *Mol Cell*
608 *Endocrinol* 335 (2011) 2-13.
- 609 [20] K.M. Giles, K. Ross, A.G. Rossi, N.A. Hotchin, C. Haslett, I. Dransfield,
610 Glucocorticoid augmentation of macrophage capacity for phagocytosis of apoptotic
611 cells is associated with reduced p130Cas expression, loss of paxillin/pyk2
612 phosphorylation, and high levels of active Rac, *J Immunol* 167 (2001) 976-986.
- 613 [21] J. Ehrchen, L. Steinmuller, K. Barczyk, K. Tenbrock, W. Nacken, M. Eisenacher,
614 U. Nordhues, C. Sorg, C. Sunderkotter, J. Roth, Glucocorticoids induce
615 differentiation of a specifically activated, anti-inflammatory subtype of human
616 monocytes, *Blood* 109 (2007) 1265-1274.
- 617 [22] F. Vallelian, C.A. Schaer, T. Kaempfer, P. Gehrig, E. Duerst, G. Schoedon, D.J.
618 Schaer, Glucocorticoid treatment skews human monocyte differentiation into a
619 hemoglobin-clearance phenotype with enhanced heme-iron recycling and
620 antioxidant capacity, *Blood* 116 (2011) 5347-5356.
- 621 [23] A. Tsianakas, G. Varga, K. Barczyk, G. Bode, N. Nippe, N. Kran, J. Roth, T.A.
622 Luger, J. Ehrchen, C. Sunderkoetter, Induction of an anti-inflammatory human
623 monocyte subtype is a unique property of glucocorticoids, but can be modified by
624 IL-6 and IL-10, *Immunobiology* 217 (2012) 329-335.
- 625 [24] G. Varga, J. Ehrchen, A. Tsianakas, K. Tenbrock, A. Rattenholl, S. Seeliger, M.
626 Mack, J. Roth, C. Sunderkoetter, Glucocorticoids induce an activated, anti-

- 627 inflammatory monocyte subset in mice that resembles myeloid-derived suppressor
628 cells, *J Leukoc Biol* 84 (2008) 644-650.
- 629 [25] S. Schif-Zuck, N. Gross, S. Assi, R. Rostoker, C.N. Serhan, A. Ariel, Saturated-
630 efferocytosis generates pro-resolving CD11b low macrophages: Modulation by
631 resolvins and glucocorticoids, *Eur J Immunol* 41 (2011) 366-379.
- 632 [26] M.P. Yeager, P.M. Guyre, A.U. Munck, Glucocorticoid regulation of the
633 inflammatory response to injury, *Acta Anaesthesiol Scand* 48 (2004) 799-813.
- 634 [27] H.Y. Lim, N. Muller, M.J. Herold, J. van den Brandt, H.M. Reichardt,
635 Glucocorticoids exert opposing effects on macrophage function dependent on their
636 concentration, *Immunology* 122 (2007) 47-53.
- 637 [28] M. Lombes, S. Kenouch, A. Souque, N. Farman, M.E. Rafestin-Oblin, The
638 mineralocorticoid receptor discriminates aldosterone from glucocorticoids
639 independently of the 11 β -hydroxysteroid dehydrogenase, *Endocrinology* 135
640 (1994) 834-840.
- 641 [29] A.H. Miller, R.L. Spencer, M. Stein, B.S. McEwen, Adrenal steroid receptor
642 binding in spleen and thymus after stress or dexamethasone, *Am J Physiol* 259
643 (1990) E405-E412.
- 644 [30] G.D. Barish, M. Downes, W.A. Alaynick, R.T. Yu, C.B. Ocampo, A.L. Bookout,
645 D.J. Mangelsdorf, R.M. Evans, A Nuclear Receptor Atlas: macrophage activation,
646 *Mol Endocrinol* 19 (2005) 2466-2477.
- 647 [31] S. Diederich, E. Eigendorff, P. Burkhardt, M. Quinkler, C. Bumke-Vogt, M.
648 Rochel, D. Seidelmann, P. Esperling, W. Oelkers, V. Bahr, 11 β -Hydroxysteroid
649 dehydrogenase types 1 and 2: an important pharmacokinetic determinant for the
650 activity of synthetic mineralo- and glucocorticoids, *J Clin Endocrinol Metab* 87
651 (2002) 5695-5701.
- 652 [32] J.R. Seckl, 11 β -hydroxysteroid dehydrogenases: changing glucocorticoid action.,
653 *Curr Opin Pharmacol* 4 (2004) 597-602.
- 654 [33] C.R.W. Edwards, P.M. Stewart, D. Burt, L. Brett, M.A. McIntyre, W.S. Sutanto,
655 E.R. de Kloet, C. Monder, Localisation of 11 β -hydroxysteroid dehydrogenase-
656 tissue specific protector of the mineralocorticoid receptor, *Lancet* ii (1988) 986-
657 989.
- 658 [34] J.W. Funder, P.T. Pearce, R. Smith, A.I. Smith, Mineralocorticoid action: target
659 tissue specificity is enzyme, not receptor, mediated, *Science* 242 (1988) 583-585.
- 660 [35] K.E. Chapman, A.E. Coutinho, M. Gray, J.S. Gilmour, J.S. Savill, J.R. Seckl, The
661 role and regulation of 11 β -hydroxysteroid dehydrogenase type 1 in the
662 inflammatory response., *Mol Cell Endocrinol* 301 (2009) 123-131.
- 663 [36] A.E. Coutinho, T. Kipari, Z. Zhang, C. Esteves, J.S. Gilmour, J.-F. Cailhier, J.
664 Hughes, J.R. Seckl, J.S. Savill, K.E. Chapman, Dynamic Regulation of 11 β -
665 Hydroxysteroid Dehydrogenase Type 1 in Neutrophils during an Inflammatory
666 Response, *Endocrine Reviews* 32 (2011) P2-575.
- 667 [37] R. Thieringer, C.B. Le Grand, L. Carbin, T.Q. Cai, B. Wong, S.D. Wright, A.
668 Hermanowski-Vosatka, 11 β -Hydroxysteroid dehydrogenase type 1 is induced in
669 human monocytes upon differentiation to macrophages, *J Immunol* 167 (2001) 30-
670 35.
- 671 [38] F.O. Martinez, A. Sica, A. Mantovani, M. Locati, Macrophage activation and
672 polarization, *Front Biosci* 13 (2008) 453-461.
- 673 [39] F.O. Martinez, L. Helming, S. Gordon, Alternative activation of macrophages: an
674 immunologic functional perspective, *Annu Rev Immunol* 27 (2009) 451-483.

- 675 [40] J.S. Duffield, S.J. Forbes, C.M. Constandinou, S. Clay, M. Partolina, S. Vuthoori,
676 S. Wu, R. Lang, J.P. Iredale, Selective depletion of macrophages reveals distinct,
677 opposing roles during liver injury and repair, *J Clin Invest* 115 (2005) 56-65.
- 678 [41] Y.Q. Liu, J.M. Cousin, J. Hughes, J. VanDamme, J.R. Seckl, C. Haslett, I.
679 Dransfield, J. Savill, A.G. Rossi, Glucocorticoids promote nonphlogistic
680 phagocytosis of apoptotic leukocytes, *J Immunol* 162 (1999) 3639-3646.
- 681 [42] A. Kleiman, S. Hubner, J.M. Rodriguez Parkitna, A. Neumann, S. Hofer, M.A.
682 Weigand, M. Bauer, W. Schmid, G. Schutz, C. Libert, H.M. Reichardt, J.P.
683 Tuckermann, Glucocorticoid receptor dimerization is required for survival in septic
684 shock via suppression of interleukin-1 in macrophages, *FASEB J* 26 (2012) 722-
685 729.
- 686 [43] M.G. Usher, S.Z. Duan, C.Y. Ivaschenko, R.A. Frieler, S. Berger, G. Schutz, C.N.
687 Lumeng, R.M. Mortensen, Myeloid mineralocorticoid receptor controls
688 macrophage polarization and cardiovascular hypertrophy and remodeling in mice,
689 *The Journal of Clinical Investigation* 120 (2010) 3350-3364.
- 690 [44] J.S. Gilmour, Glucocorticoids, 11 β -hydroxysteroid dehydrogenases and
691 macrophage function., PhD thesis, University of Edinburgh, 2003.
- 692 [45] F.O. Martinez, S. Gordon, M. Locati, A. Mantovani, Transcriptional profiling of
693 the human monocyte-to-macrophage differentiation and polarization: new
694 molecules and patterns of gene expression, *J Immunol* 177 (2006) 7303-7311.
- 695 [46] V. Joganathan, A. Al-Hakami, S. Rauz, P.M. Stewart, G.R. Wallace, I.J. Bujalska,
696 Local cortisol generation by human macrophage subsets by 11 β -hydroxysteroid
697 dehydrogenase type 1 enzyme and its role in ocular immune privilege, *Endocrine*
698 *Abstracts* 15 (2008) OC30.
- 699 [47] G. Chinetti-Gbaguidi, M.A. Bouhrel, C. Copin, C. Duhem, B. Derudas, B. Neve, B.
700 Noel, J. Eeckhoutte, P. Lefebvre, J.R. Seckl, B. Staels, Peroxisome Proliferator
701 Activated Receptor- α Activation Induces 11 β -Hydroxysteroid Dehydrogenase
702 Type 1 Activity in Human Alternative Macrophages, *Arterioscler Thromb Vasc*
703 *Biol* 32 (2012) 677-685.
- 704 [48] J.C. Rodriguez-Prados, P.G. Traves, J. Cuenca, D. Rico, J. Aragonés, P. Martín-
705 Sanz, M. Cascante, L. Bosca, Substrate fate in activated macrophages: a
706 comparison between innate, classic, and alternative activation, *J Immunol* 185
707 (2010) 605-614.
- 708 [49] A. Haschemi, P. Kosma, L. Gille, C.R. Evans, C.F. Burant, P. Starkl, B. Knapp, R.
709 Haas, J.A. Schmid, C. Jandl, S. Amir, G. Lubec, J. Park, H. Esterbauer, M. Bilban,
710 L. Brizuela, J.A. Pospisilik, L.E. Otterbein, O. Wagner, The Sedoheptulose Kinase
711 CARKL Directs Macrophage Polarization through Control of Glucose Metabolism,
712 *Cell Metab* 15 (2012) 813-826.
- 713 [50] V.A. Fadok, D.L. Bratton, A. Konowal, P.W. Freed, J.Y. Westcott, P.M. Henson,
714 Macrophages that have ingested apoptotic cells in vitro inhibit proinflammatory
715 cytokine production through autocrine/paracrine mechanisms involving TGF- β ,
716 PGE₂, and PAF, *J Clin Invest* 101 (1998) 890-898.
- 717 [51] K.E. Chapman, A. Coutinho, M. Gray, J.S. Gilmour, J.S. Savill, J.R. Seckl, Local
718 amplification of glucocorticoids by 11 β -hydroxysteroid dehydrogenase type 1 and
719 its role in the inflammatory response, *Ann N Y Acad Sci* 1088 (2006) 265-273.
- 720 [52] J. Bryndova, S. Zbankova, M. Kment, J. Pacha, Colitis up-regulates local
721 glucocorticoid activation and down-regulates inactivation in colonic tissue, *Scand J*
722 *Gastroenterol* 39 (2004) 549-553.

- 723 [53] P. Ergang, P. Leden, K. Vagnerova, P. Klusonova, I. Miksik, J. Jurcovicova, M.
724 Kment, J. Pacha, Local metabolism of glucocorticoids and its role in rat adjuvant
725 arthritis, *Mol Cell Endocrinol* 323 (2010) 155-160.
- 726 [54] P. Ergang, K. Vytackova, J. Svec, J. Bryndova, I. Miksik, J. Pacha, Upregulation
727 of 11 β -hydroxysteroid dehydrogenase 1 in lymphoid organs during inflammation
728 in the rat, *J Steroid Biochem Mol Biol* 126 (2011) 19-25.
- 729 [55] A.R. Dover, P.W. Hadoke, L.J. Macdonald, E. Miller, D.E. Newby, B.R. Walker,
730 Intravascular glucocorticoid metabolism during inflammation and injury in mice,
731 *Endocrinology* 148 (2007) 166-172.
- 732 [56] Z. Yang, X. Zhu, C. Guo, K. Sun, Stimulation of 11 β -HSD1 expression by IL-1!
733 via a C/EBP binding site in human fetal lung fibroblasts, *Endocrine* 36 (2009) 404-
734 411.
- 735 [57] I.D. Ignatova, R.M. Kostadinova, C.E. Goldring, A.R. Nawrocki, F.J. Frey, B.M.
736 Frey, Tumor necrosis factor- α upregulates 11 β -hydroxysteroid dehydrogenase type
737 1 expression by CCAAT/enhancer binding protein-1 in HepG2 cells, *Am J Physiol*
738 *Endocrinol Metab* 296 (2009) E367-377.
- 739 [58] M.M. Ahasan, R. Hardy, C. Jones, K. Kaur, D. Nanus, M. Juarez, S.A. Morgan, Z.
740 Hassan-Smith, C. Benezech, J.H. Caamano, M. Hewison, G. Lavery, E.H. Rabbitt,
741 A.R. Clark, A. Filer, C.D. Buckley, K. Raza, P.M. Stewart, M.S. Cooper,
742 Inflammatory regulation of glucocorticoid metabolism in mesenchymal stromal
743 cells, *Arthritis Rheum* 64 (2012) 2404-2413.
- 744 [59] R.M. Kostadinova, A.R. Nawrocki, F.J. Frey, B.M. Frey, Tumor necrosis factor
745 α and phorbol 12-myristate-13-acetate down-regulate human 11 β -
746 hydroxysteroid dehydrogenase type 2 through p50/p50 NF- κ B homodimers
747 and Egr-1, *FASEB J* 19 (2005) 650-652.
- 748 [60] K. Sun, L. Myatt, Enhancement of glucocorticoid-induced 11 β -hydroxysteroid
749 dehydrogenase type 1 expression by proinflammatory cytokines in cultured human
750 amnion fibroblasts, *Endocrinology* 144 (2003) 5568-5577.
- 751 [61] M.T. Rae, D. Niven, H.O. Critchley, C.R. Harlow, S.G. Hillier, Antiinflammatory
752 steroid action in human ovarian surface epithelial cells, *J Clin Endocrinol Metab*
753 89 (2004) 4538-4544.
- 754 [62] W. Li, L. Gao, Y. Wang, T. Duan, L. Myatt, K. Sun, Enhancement of cortisol-
755 induced 11 β -hydroxysteroid dehydrogenase type 1 expression by interleukin 1!
756 in cultured human chorionic trophoblast cells, *Endocrinology* 147 (2006) 2490-2495.
- 757 [63] K. Kaur, R. Hardy, M.M. Ahasan, M. Eijken, J.P. van Leeuwen, A. Filer, A.M.
758 Thomas, K. Raza, C.D. Buckley, P.M. Stewart, E.H. Rabbitt, M. Hewison, M.S.
759 Cooper, Synergistic induction of local glucocorticoid generation by inflammatory
760 cytokines and glucocorticoids: implications for inflammation associated bone loss,
761 *Ann Rheum Dis* 69: (2010) 1185-1190.
- 762 [64] L.J.S. Williams, V. Lyons, I. MacLeod, V. Rajan, G.J. Darlington, V. Poli, J.R.
763 Seckl, K.E. Chapman, C/EBP regulates hepatic transcription of 11 β -
764 hydroxysteroid dehydrogenase type 1; a novel mechanism for cross talk between
765 the C/EBP and glucocorticoid signalling pathways, *J Biol Chem* 275 (2000) 30232-
766 30239.
- 767 [65] J. Gout, J. Tirard, C. Thevenon, J.P. Riou, M. Begeot, D. Naville,
768 CCAAT/enhancer-binding proteins (C/EBPs) regulate the basal and cAMP-
769 induced transcription of the human 11 β -hydroxysteroid dehydrogenase encoding
770 gene in adipose cells, *Biochimie* 88 (2006) 1115-1124.

- 771 [66] V.A. Payne, W.S. Au, S.L. Gray, E.D. Nora, S.M. Rahman, R. Sanders, D.
772 Hadaschik, J.E. Friedman, S. O'Rahilly, J.J. Rochford, Sequential regulation of
773 diacylglycerol acyltransferase 2 expression by CAAT/enhancer-binding protein
774 beta (C/EBP β) and C/EBP β during adipogenesis, *J Biol Chem* 282 (2007) 21005-
775 21014.
- 776 [67] N. Arai, H. Masuzaki, T. Tanaka, T. Ishii, S. Yasue, N. Kobayashi, T. Tomita, M.
777 Noguchi, T. Kusakabe, J. Fujikura, K. Ebihara, M. Hirata, K. Hosoda, T. Hayashi,
778 H. Sawai, Y. Minokoshi, K. Nakao, Ceramide and adenosine 5'-monophosphate-
779 activated protein kinase are two novel regulators of 11 β -hydroxysteroid
780 dehydrogenase type 1 expression and activity in cultured preadipocytes,
781 *Endocrinology* 148 (2007) 5268-5277.
- 782 [68] S. Sai, C.L. Esteves, V. Kelly, Z. Michailidou, K. Anderson, A.P. Coll, Y.
783 Nakagawa, T. Ohzeki, J.R. Seckl, K.E. Chapman, Glucocorticoid regulation of the
784 promoter of 11 β -hydroxysteroid dehydrogenase type 1 is indirect and requires
785 C/EBP β , *Mol Endocrinol* 22 (2008) 2049-2060.
- 786 [69] D. Ruffell, F. Mourkioti, A. Gambardella, P. Kirstetter, R.G. Lopez, N. Rosenthal,
787 C. Nerlov, A CREB-C/EBP β cascade induces M2 macrophage-specific gene
788 expression and promotes muscle injury repair, *Proc Natl Acad Sci U S A* 106
789 (2009) 17475-17480.
- 790 [70] B. Gorgoni, D. Maritano, P. Marthyn, M. Righi, V. Poli, C/EBP beta gene
791 inactivation causes both impaired and enhanced gene expression and inverse
792 regulation of IL-12 p40 and p35 mRNAs in macrophages, *J Immunol* 168 (2002)
793 4055-4062.
- 794 [71] C.L. Esteves, V. Kelly, V. Begay, T.Y. Man, N.M. Morton, A. Leutz, J.R. Seckl,
795 K.E. Chapman, Regulation of Adipocyte 11 β -Hydroxysteroid Dehydrogenase
796 Type 1 (11 β -HSD1) by CCAAT/Enhancer-Binding Protein (C/EBP) beta Isoforms,
797 LIP and LAP, *PLoS One* 7 (2012) e37953.
- 798 [72] J.J. Smink, V. Begay, T. Schoenmaker, E. Sterneck, T.J. de Vries, A. Leutz,
799 Transcription factor C/EBPbeta isoform ratio regulates osteoclastogenesis through
800 MafB, *EMBO J* 28 (2009) 1769-1781.
- 801 [73] J.J. Smink, A. Leutz, Rapamycin and the transcription factor C/EBPbeta as a
802 switch in osteoclast differentiation: implications for lytic bone diseases, *J Mol Med*
803 88 (2009) 227-233.
- 804 [74] M.J. Rauh, V. Ho, C. Pereira, A. Sham, L.M. Sly, V. Lam, L. Huxham, A.I.
805 Minchinton, A. Mui, G. Krystal, SHIP represses the generation of alternatively
806 activated macrophages, *Immunity* 23 (2005) 361-374.
- 807 [75] A. Arranz, C. Doxaki, E. Vergadi, Y. Martinez de la Torre, K. Vaporidi, E.D.
808 Lagoudaki, E. Ieronymaki, A. Androulidaki, M. Venihaki, A.N. Margioris, E.N.
809 Stathopoulos, P.N. Tsihchlis, C. Tsatsanis, Akt1 and Akt2 protein kinases
810 differentially contribute to macrophage polarization, *Proc Natl Acad Sci U S A* 109
811 (2012) 9517-9522.
- 812 [76] A.G. Atanasov, L.G. Nashev, R.A. Schweizer, C. Frick, A. Odermatt, Hexose-6-
813 phosphate dehydrogenase determines the reaction direction of 11 β -hydroxysteroid
814 dehydrogenase type 1 as an oxoreductase, *FEBS Lett* 571 (2004) 129-133.
- 815 [77] G. Banhegyi, A. Benedetti, R. Fulceri, S. Senesi, Cooperativity between 11 β -
816 hydroxysteroid dehydrogenase type 1 and hexose-6-phosphate dehydrogenase in
817 the lumen of the endoplasmic reticulum, *J Biol Chem* 279 (2004) 27017-27021.
- 818 [78] I.J. Bujalska, N. Draper, Z. Michailidou, J.W. Tomlinson, P.C. White, K.E.
819 Chapman, E.A. Walker, P.M. Stewart, Hexose-6-phosphate dehydrogenase confers

- 820 oxo-reductase activity upon 11 β -hydroxysteroid dehydrogenase type 1, *J Mol*
821 *Endocrinol* 34 (2005) 675-684.
- 822 [79] E.A. Walker, A. Ahmed, G.G. Lavery, J.W. Tomlinson, S.Y. Kim, M.S. Cooper,
823 J.P. Ride, B.A. Hughes, C.H. Shackleton, P. McKiernan, E. Elias, J.Y. Chou, P.M.
824 Stewart, 11 β -Hydroxysteroid Dehydrogenase Type 1 Regulation by Intracellular
825 Glucose 6-Phosphate Provides Evidence for a Novel Link between Glucose
826 Metabolism and Hypothalamo-Pituitary-Adrenal Axis Function, *J Biol Chem* 282
827 (2007) 27030-27036.
- 828 [80] R. Bertini, M. Bianchi, P. Ghezzi, Adrenalectomy sensitizes mice to the lethal
829 effects of Interleukin-1 and Tumor Necrosis Factor, *J Exp Med* 167 (1988) 1708-
830 1712.
- 831 [81] T.Y. Zhang, R.A. Daynes, Macrophages from 11 β -hydroxysteroid dehydrogenase
832 type 1-deficient mice exhibit an increased sensitivity to lipopolysaccharide
833 stimulation due to TGF- β -mediated up-regulation of SHIP1 expression, *J Immunol*
834 179 (2007) 6325-6335.
- 835 [82] S.J. McSweeney, P.W. Hadoke, A.M. Kozak, G.R. Small, H. Khaled, B.R. Walker,
836 G.A. Gray, Improved heart function follows enhanced inflammatory cell
837 recruitment and angiogenesis in 11 β -HSD1-deficient mice post-MI, *Cardiovasc*
838 *Res* 88 (2010) 159-167.
- 839 [83] A.E. Coutinho, M. Gray, D.G. Brownstein, D.M. Salter, D.A. Sawatzky, S. Clay,
840 J.S. Gilmour, J.R. Seckl, J.S. Savill, K.E. Chapman, 11 β -Hydroxysteroid
841 Dehydrogenase Type 1, But Not Type 2, Deficiency Worsens Acute Inflammation
842 and Experimental Arthritis in Mice, *Endocrinology* 153 (2012) 234-240.
- 843 [84] T. Kardon, S. Senesi, P. Marcolongo, B. Legeza, G. Banhegyi, J. Mandl, R.
844 Fulceri, A. Benedetti, Maintenance of luminal NADPH in the endoplasmic
845 reticulum promotes the survival of human neutrophil granulocytes, *FEBS Lett* 582
846 (2008) 1809-1815.
- 847 [85] G.R. Small, P.W. Hadoke, I. Sharif, A.R. Dover, D. Armour, C.J. Kenyon, G.A.
848 Gray, B.R. Walker, Preventing local regeneration of glucocorticoids by 11 β -
849 hydroxysteroid dehydrogenase type 1 enhances angiogenesis, *Proc Natl Acad Sci*
850 *U S A* 102 (2005) 12165-12170.
- 851 [86] M. Wamil, J.H. Battle, S. Turban, T. Kipari, D. Seguret, R. de Sousa Peixoto, Y.B.
852 Nelson, D. Nowakowska, D. Ferenbach, L. Ramage, K.E. Chapman, J. Hughes,
853 D.R. Dunbar, J.R. Seckl, N.M. Morton, Novel fat depot-specific mechanisms
854 underlie resistance to visceral obesity and inflammation in 11 β -hydroxysteroid
855 dehydrogenase type 1-deficient mice, *Diabetes* 60 (2011) 1158-1167.
- 856 [87] Z. Michailidou, S. Turban, E. Miller, X. Zou, J. Schrader, P.J. Ratcliffe, P.W.
857 Hadoke, B.R. Walker, J.P. Iredale, N.M. Morton, J.R. Seckl, Increased
858 Angiogenesis Protects against Adipose Hypoxia and Fibrosis in Metabolic
859 Disease-resistant 11 β -Hydroxysteroid Dehydrogenase Type 1 (HSD1)-deficient
860 Mice, *J Biol Chem* 287 (2012) 4188-4197.
- 861 [88] R. Carter, J.M. Paterson, U. Tworowska, D.J. Stenvers, J.J. Mullins, J.R. Seckl,
862 M.C. Holmes, Hypothalamic-pituitary-adrenal axis abnormalities in response to
863 deletion of 11 β -HSD1 is strain-dependent, *J Neuroendocrinol* 21 (2009) 879-
864 877.
- 865 [89] A. Sica, A. Mantovani, Macrophage plasticity and polarization: in vivo veritas, *J*
866 *Clin Invest* 122 (2012) 787-795.

- 867 [90] L.M. Sly, M.J. Rauh, J. Kalesnikoff, C.H. Song, G. Krystal, LPS-induced
868 upregulation of SHIP is essential for endotoxin tolerance, *Immunity* 21 (2004) 227-
869 239.
- 870 [91] N.M. Morton, J.M. Paterson, H. Masuzaki, M.C. Holmes, B. Staels, C. Fievet, B.R.
871 Walker, J.S. Flier, J.J. Mullins, J.R. Seckl, Novel adipose tissue-mediated
872 resistance to diet-induced visceral obesity in 11 β -hydroxysteroid dehydrogenase
873 type 1-deficient mice, *Diabetes* 53 (2004) 931-938.
- 874 [92] J.R. Seckl, N.M. Morton, K.E. Chapman, B.R. Walker, Glucocorticoids and 11 β -
875 hydroxysteroid dehydrogenase in adipose tissue, *Recent Prog Horm Res* 59 (2004)
876 359-393.
- 877 [93] C. Edwards, Sixty years after Hench--corticosteroids and chronic inflammatory
878 disease, *J Clin Endocrinol Metab* 97 (2012) 1443-1451.
- 879 [94] Y. Ichikawa, K. Yoshida, M. Kawagoe, E. Saito, Y. Abe, K. Arikawa, M. Homma,
880 Altered equilibrium between cortisol and cortisone in plasma in thyroid
881 dysfunction and inflammatory diseases, *Metabolism* 26 (1977) 989-997.
- 882 [95] Y. Kotelevtsev, M.C. Holmes, A. Burchell, P.M. Houston, D. Schmolli, P.
883 Jamieson, R. Best, R. Brown, C.R.W. Edwards, J.R. Seckl, J.J. Mullins, 11 β -
884 hydroxysteroid dehydrogenase type 1 knockout mice show attenuated
885 glucocorticoid inducible responses and resist hyperglycaemia on obesity or stress.,
886 *Proc Natl Acad Sci USA* 94 (1997) 14924-14929.
- 887 [96] N.M. Morton, M.C. Holmes, C. Fievet, B. Staels, A. Tailleux, J.J. Mullins, J.R.
888 Seckl, Improved lipid and lipoprotein profile, hepatic insulin sensitivity, and
889 glucose tolerance in 11 β -hydroxysteroid dehydrogenase type 1 null mice, *J Biol*
890 *Chem* 276 (2001) 41293-41300.
- 891 [97] P. Alberts, L. Engblom, N. Edling, M. Forsgren, G. Klingstrom, C. Larsson, Y.
892 Ronquist-Nii, B. Ohman, L. Abrahmsen, Selective inhibition of 11 β -
893 hydroxysteroid dehydrogenase type 1 decreases blood glucose concentrations in
894 hyperglycaemic mice, *Diabetologia* 45 (2002) 1528-1532.
- 895 [98] P. Alberts, C. Nilsson, G. Selen, L.O. Engblom, N.H. Edling, S. Norling, G.
896 Klingstrom, C. Larsson, M. Forsgren, M. Ashkzari, C.E. Nilsson, M. Fiedler, E.
897 Bergqvist, B. Ohman, E. Bjorkstrand, L.B. Abrahmsen, Selective inhibition of
898 11 β -hydroxysteroid dehydrogenase type 1 improves hepatic insulin sensitivity in
899 hyperglycemic mice strains, *Endocrinology* 144 (2003) 4755-4762.
- 900 [99] M. Berthiaume, M. Laplante, W. Festuccia, Y. Gelinas, S. Poulin, J. Lalonde, D.R.
901 Joannisse, R. Thieringer, Y. Deshaies, Depot-specific modulation of rat
902 intraabdominal adipose tissue lipid metabolism by pharmacological inhibition of
903 11 β -hydroxysteroid dehydrogenase type 1, *Endocrinology* 148 (2007) 2391-2397.
- 904 [100] M. Berthiaume, M. Laplante, W.T. Festuccia, K. Cianflone, L.P. Turcotte, D.R.
905 Joannisse, G. Olivecrona, R. Thieringer, Y. Deshaies, 11 β -HSD1 inhibition
906 improves triglyceridemia through reduced liver VLDL secretion and partitions
907 lipids toward oxidative tissues, *Am J Physiol Endocrinol Metab* 293 (2007) E1045-
908 1052.
- 909 [101] M. Berthiaume, M. Laplante, W.T. Festuccia, J.P. Berger, R. Thieringer, Y.
910 Deshaies, Preliminary report: pharmacologic 11 β -hydroxysteroid dehydrogenase
911 type 1 inhibition increases hepatic fat oxidation in vivo and expression of related
912 genes in rats fed an obesogenic diet, *Metabolism* 59 (2010) 114-117.
- 913 [102] J. Rosenstock, S. Banarer, V.A. Fonseca, S.E. Inzucchi, W. Sun, W. Yao, G.
914 Hollis, R. Flores, R. Levy, W.V. Williams, J.R. Seckl, R. Huber, The 11 β -
915 hydroxysteroid dehydrogenase type 1 inhibitor INCB13739 improves

- 916 hyperglycemia in patients with type 2 diabetes inadequately controlled by
917 metformin monotherapy, *Diabetes Care* 33 (2010) 1516-1522.
- 918 [103] P.U. Feig, S. Shah, A. Hermanowski Vosatka, D. Plotkin, M.S. Springer, S.
919 Donahue, C. Thach, E.J. Klein, E. Lai, K.D. Kaufman, Effects of an 11 β -
920 hydroxysteroid dehydrogenase type 1 inhibitor, MK-0916, in patients with type 2
921 diabetes mellitus and metabolic syndrome, *Diabetes Obes Metab* 13 (2011) 498-
922 504.
- 923 [104] S. Shah, A. Hermanowski-Vosatka, K. Gibson, R.A. Ruck, G. Jia, J. Zhang,
924 P.M.T. Hwang, N.W. Ryan, R.B. Langdon, P.U. Feig, Efficacy and safety of the
925 selective 11 β -HSD-1 inhibitors MK-0736 and MK-0916 in overweight and obese
926 patients with hypertension, *Journal of the American Society of Hypertension* 5
927 (2011) 166-176.
- 928 [105] M.M. Veniant, C. Hale, R. Komorowski, M.M. Chen, D.J. St Jean, C. Fotsch, M.
929 Wang, Time of the day for 11 β -HSD1 inhibition plays a role in improving glucose
930 homeostasis in DIO mice, *Diabetes Obes Metab* 11 (2009) 109-117.
- 931 [106] C.S. Wyrwoll, M.C. Holmes, J.R. Seckl, 11 β -Hydroxysteroid dehydrogenases
932 and the brain: From zero to hero, a decade of progress, *Frontiers in*
933 *Neuroendocrinology* 32 (2011) 265-286.
- 934 [107] J. Buren, S.-A. Bergstrom, E. Loh, I. Soderstrom, T. Olsson, C. Mattsson,
935 Hippocampal 11 β -hydroxysteroid dehydrogenase type 1 mRNA expression has a
936 diurnal variability which is lost in the obese Zucker rat, *Endocrinology* 148 (2007)
937 2716-2722.
- 938 [108] H.J. Harris, Y. Kotelevtsev, J.J. Mullins, J.R. Seckl, M.C. Holmes, Intracellular
939 regeneration of glucocorticoids by 11 β -hydroxysteroid dehydrogenase (11 β -HSD)-
940 1 plays a key role in regulation of the hypothalamic-pituitary-adrenal axis: analysis
941 of 11 β -HSD-1 deficient mice, *Endocrinology* 142 (2001) 114-120.
- 942 [109] P.H. Bisschop, M.J. Dekker, W. Osterthun, J. Kwakkel, J.J. Anink, A. Boelen,
943 U.A. Unmehopa, J.W. Koper, S.W. Lamberts, P.M. Stewart, D.F. Swaab, E. Fliers,
944 Expression of 11 β -hydroxysteroid dehydrogenase type 1 in the human
945 hypothalamus, *J Neuroendocrinol* (2013) Jan 3. doi: 10.1111/jne.12017. [Epub
946 ahead of print].
- 947 [110] G.G. Lavery, A.E. Zielinska, L.L. Gathercole, B. Hughes, N. Semjonous, P.
948 Guest, K. Saqib, M. Sherlock, G. Reynolds, S.A. Morgan, J.W. Tomlinson, E.A.
949 Walker, E.H. Rabbitt, P.M. Stewart, Lack of Significant Metabolic Abnormalities
950 in Mice with Liver-Specific Disruption of 11 β -Hydroxysteroid Dehydrogenase
951 Type 1, *Endocrinology* 153 (2012) 3236-3248.
- 952 [111] Z. Michailidou, M.D. Jensen, D.A. Dumesic, K.E. Chapman, J.R. Seckl, B.R.
953 Walker, N.M. Morton, Omental 11 β -hydroxysteroid dehydrogenase 1 correlates
954 with fat cell size independently of obesity, *Obesity* 15 (2007) 1155-1163.
- 955 [112] M.J. Lee, S.K. Fried, S.S. Mundt, Y. Wang, S. Sullivan, A. Stefanni, B.L.
956 Daugherty, A. Hermanowski-Vosatka, Depot-specific regulation of the conversion
957 of cortisone to cortisol in human adipose tissue, *Obesity (Silver Spring)* 16 (2008)
958 1178-1185.
- 959 [113] S. Cinti, G. Mitchell, G. Barbatelli, I. Murano, E. Ceresi, E. Faloia, S. Wang, M.
960 Fortier, A.S. Greenberg, M.S. Obin, Adipocyte death defines macrophage
961 localization and function in adipose tissue of obese mice and humans, *J Lipid Res*
962 46 (2005) 2347-2355.
- 963 [114] M. Jernas, J. Palming, K. Sjöholm, E. Jennische, P.A. Svensson, B.G.
964 Gabrielsson, M. Levin, A. Sjögren, M. Rudemo, T.C. Lystig, B. Carlsson, L.M.

- 965 Carlsson, M. Lonn, Separation of human adipocytes by size: hypertrophic fat cells
966 display distinct gene expression, *FASEB J* 20 (2006) 1540-1542.
- 967 [115] H. Masuzaki, J. Paterson, H. Shinyama, N.M. Morton, J.J. Mullins, J.R. Seckl,
968 J.S. Flier, A transgenic model of visceral obesity and the metabolic syndrome,
969 *Science* 294 (2001) 2166-2170.
- 970 [116] H. Masuzaki, H. Yamamoto, C.J. Kenyon, J.K. Elmquist, N.M. Morton, J.M.
971 Paterson, H. Shinyama, M.G. Sharp, S. Fleming, J.J. Mullins, J.R. Seckl, J.S. Flier,
972 Transgenic amplification of glucocorticoid action in adipose tissue causes high
973 blood pressure in mice, *J Clin Invest* 112 (2003) 83-90.
- 974 [117] E.E. Kershaw, N.M. Morton, H. Dhillon, L. Ramage, J.R. Seckl, J.S. Flier,
975 Adipocyte-specific glucocorticoid inactivation protects against diet-induced
976 obesity, *Diabetes* 54 (2005) 1023-1031.
- 977 [118] S.A. Hawley, M.D. Fullerton, F.A. Ross, J.D. Schertzer, C. Chevtzoff, K.J.
978 Walker, M.W. Peggie, D. Zibrova, K.A. Green, K.J. Mustard, B.E. Kemp, K.
979 Sakamoto, G.R. Steinberg, D.G. Hardie, The Ancient Drug Salicylate Directly
980 Activates AMP-Activated Protein Kinase, *Science* 336 (2012) 918-922.
- 981 [119] N. Hosogai, A. Fukuhara, K. Oshima, Y. Miyata, S. Tanaka, K. Segawa, S.
982 Furukawa, Y. Tochino, R. Komuro, M. Matsuda, I. Shimomura, Adipose tissue
983 hypoxia in obesity and its impact on adipocytokine dysregulation, *Diabetes* 56
984 (2007) 901-911.
- 985 [120] I. Stuart Wood, F.t.P.r. de Heredia, B. Wang, P. Trayhurn, Cellular hypoxia and
986 adipose tissue dysfunction in obesity, *Proceedings of the Nutrition Society* 68
987 (2009) 370.
- 988 [121] N. Halberg, T. Khan, M.E. Trujillo, I. Wernstedt-Asterholm, A.D. Attie, S.
989 Sherwani, Z.V. Wang, S. Landskroner-Eiger, S. Dineen, U.J. Magalang, R.A.
990 Brekken, P.E. Scherer, Hypoxia-inducible factor 1alpha induces fibrosis and
991 insulin resistance in white adipose tissue, *Mol Cell Biol* 29 (2009) 4467-4483.
- 992 [122] A. Hermanowski-Vosatka, J.M. Balkovec, K. Cheng, H.Y. Chen, M. Hernandez,
993 G.C. Koo, C.B. Le Grand, Z. Li, J.M. Metzger, S.S. Mundt, H. Noonan, C.N.
994 Nunes, S.H. Olson, B. Pikounis, N. Ren, N. Robertson, J.M. Schaeffer, K. Shah,
995 M.S. Springer, A.M. Strack, M. Strowski, K. Wu, T. Wu, J. Xiao, B.B. Zhang,
996 S.D. Wright, R. Thieringer, 11 β -HSD1 inhibition ameliorates metabolic syndrome
997 and prevents progression of atherosclerosis in mice, *J Exp Med* 202 (2005) 517-
998 527.
- 999 [123] M.J. Luo, R. Thieringer, M.S. Springer, S.D. Wright, A. Hermanowski-Vosatka,
1000 A. Plump, J.M. Balkovec, K. Cheng, G.J. Ding, D.W. Kawka, G.C. Koo, C.B. Le
1001 Grand, Q. Luo, M.M. Maletic, L. Malkowitz, K. Shah, I. Singer, S.T. Waddell,
1002 K.K. Wu, J. Yuan, J. Zhu, S. Stepaniants, X. Yang, P.Y. Lum, I.M. Wang, 11 β -
1003 HSD1 inhibition reduces atherosclerosis in mice by altering pro-inflammatory gene
1004 expression in the vasculature, *Physiol Genomics* 45 (2013) 47-57.
- 1005 [124] T.M.J. Kipari, T.-Y. Man, P.W.F. Hadoke, J.S. Savill, K.E. Chapman, J.R. Seckl,
1006 11 β -hydroxysteroid dehydrogenase type 1 deficiency in bone marrow-derived cells
1007 reduces atherosclerosis, *FASEB J* in press (2013).
- 1008 [125] G.A. Deuchar, D. McLean, P.W.F. Hadoke, D.G. Brownstein, D.J. Webb, J.J.
1009 Mullins, K. Chapman, J.R. Seckl, Y.V. Kotelevtsev, 11 β -Hydroxysteroid
1010 Dehydrogenase Type 2 Deficiency Accelerates Atherogenesis and Causes
1011 Proinflammatory Changes in the Endothelium in Apoe $^{-/-}$ Mice, *Endocrinology* 152
1012 (2011) 236-246.

- 1013 [126] R.S. Hardy, E.H. Rabbitt, A. Filer, P. Emery, M. Hewison, P.M. Stewart, N.
 1014 Gittoes, C.D. Buckley, K. Raza, M.S. Cooper, Local and systemic glucocorticoid
 1015 metabolism in inflammatory arthritis, *Ann Rheum Dis* 67 (2008) 1204-1210.
- 1016 [127] M. Schmidt, C. Weidler, H. Naumann, S. Anders, J. Scholmerich, R.H. Straub,
 1017 Reduced capacity for the reactivation of glucocorticoids in rheumatoid arthritis
 1018 synovial cells: Possible role of the sympathetic nervous system?, *Arthritis Rheum*
 1019 52 (2005) 1711-1720.
- 1020 [128] R.S. Hardy, A. Filer, M.S. Cooper, G. Parsonage, K. Raza, D.L. Hardie, E.H.
 1021 Rabbitt, P.M. Stewart, C.D. Buckley, M. Hewison, Differential expression,
 1022 function and response to inflammatory stimuli of 11 β -hydroxysteroid
 1023 dehydrogenase type 1 in human fibroblasts: a mechanism for tissue-specific
 1024 regulation of inflammation, *Arthritis Res Ther* 8 (2006) R108.
- 1025 [129] N. Olsen, T. Sokka, C.L. Seehorn, B. Kraft, K. Maas, J. Moore, T.M. Aune, A
 1026 gene expression signature for recent onset rheumatoid arthritis in peripheral blood
 1027 mononuclear cells, *Ann Rheum Dis* 63 (2004) 1387-1392.
- 1028 [130] C.S. Haas, C.J. Creighton, X. Pi, I. Maine, A.E. Koch, G.K. Haines, S. Ling,
 1029 A.M. Chinnaiyan, J. Holoshitz, Identification of genes modulated in rheumatoid
 1030 arthritis using complementary DNA microarray analysis of lymphoblastoid B cell
 1031 lines from disease-discordant monozygotic twins, *Arthritis Rheum* 54 (2006) 2047-
 1032 2060.
- 1033 [131] S. Suzuki, H. Tsubochi, H. Ishibashi, T. Suzuki, T. Kondo, H. Sasano, Increased
 1034 expression of 11 β -hydroxysteroid dehydrogenase type 2 in the lungs of patients
 1035 with acute respiratory distress syndrome, *Pathol Int* 53 (2003) 751-756.
- 1036 [132] M.J. Young, Mechanisms of mineralocorticoid receptor-mediated cardiac fibrosis
 1037 and vascular inflammation, *Curr Opin Nephrol Hypertens* 17 (2008) 174-180.
- 1038 [133] J.M. Luther, P. Luo, Z. Wang, S.E. Cohen, H.S. Kim, A.B. Fogo, N.J. Brown,
 1039 Aldosterone deficiency and mineralocorticoid receptor antagonism prevent
 1040 angiotensin II-induced cardiac, renal, and vascular injury, *Kidney Int* 82 (2012)
 1041 643-651.
- 1042 [134] E.R. de Kloet, J.M. Reul, W. Sutanto, Corticosteroids and the brain, *J Steroid*
 1043 *Biochem Mol Biol* 37 (1990) 387-394.
- 1044 [135] J.W. Funder, Mineralocorticoid receptor activation and oxidative stress,
 1045 *Hypertension* 50 (2007) 840-841.
- 1046 [136] C. Guo, V. Ricchiuti, B.Q. Lian, T.M. Yao, P. Coutinho, J.R. Romero, J. Li, G.H.
 1047 Williams, G.K. Adler, Mineralocorticoid receptor blockade reverses obesity-
 1048 related changes in expression of adiponectin, peroxisome proliferator-activated
 1049 receptor-gamma, and proinflammatory adipokines, *Circulation* 117 (2008) 2253-
 1050 2261.
- 1051 [137] R.A. Frieler, H. Meng, S.Z. Duan, S. Berger, G. Schutz, Y. He, G. Xi, M.M.
 1052 Wang, R.M. Mortensen, Myeloid-Specific Deletion of the Mineralocorticoid
 1053 Receptor Reduces Infarct Volume and Alters Inflammation During Cerebral
 1054 Ischemia, *Stroke* 42 (2011) 179-185.

1055 FIGURE LEGENDS

1056 **Figure 1. 11 β -HSD1 is induced upon macrophage differentiation**

1057 Expression of 11 β -HSD1 is negligible in human monocytes, but is induced on
 1058 differentiation into macrophages. Polarisation of macrophages to an M1 phenotype
 1059 further induces 11 β -HSD1 whereas polarisation to an M2 phenotype has no further
 1060 effect on expression. Differentiation of monocytes into macrophages in the presence of
 1061 IL-4 and/or IL-13 further induces 11 β -HSD1 (see text for details).
 1062

1063

1064 **Figure 2. Macrophage polarisation is associated with a switch in energy metabolism**

1065 M1 macrophages show a predominantly glycolytic metabolism. High levels of glucose-
1066 6-phosphate (G6P) may ensure a ready supply of NADPH cofactor to 11! -HSD1,
1067 driving high conversion of cortisone (E) to cortisol (F). M2 polarised macrophages are
1068 oxidative, with lower levels of glycolysis and lower levels of 11! -HSD1 converting E
1069 to F. Whether changes in energy metabolism drive changes in macrophage 11! -HSD1
1070 expression is currently unknown (see text for details).

1071

1072 **Figure 3. Effects of 11!-HSD1 deficiency/inhibition on acute and chronic**
1073 **inflammation**

1074 Deficiency or inhibition of 11! -HSD1 worsens or exacerbates acute inflammation, but
1075 may also promote its successful resolution. During chronic metabolic inflammation
1076 (obesity, atherosclerosis, diabetes), 11! -HSD1 deficiency/inhibition is beneficial,
1077 reducing inflammatory cell recruitment to sites of inflammation and promoting insulin
1078 sensitisation. However, during chronic non-resolving inflammation, the pro-angiogenic,
1079 pro-fibrotic phenotype of 11! -HSD1 deficiency/inhibition may worsen tissue damage
1080 (see text for details).

1081

Figure 1

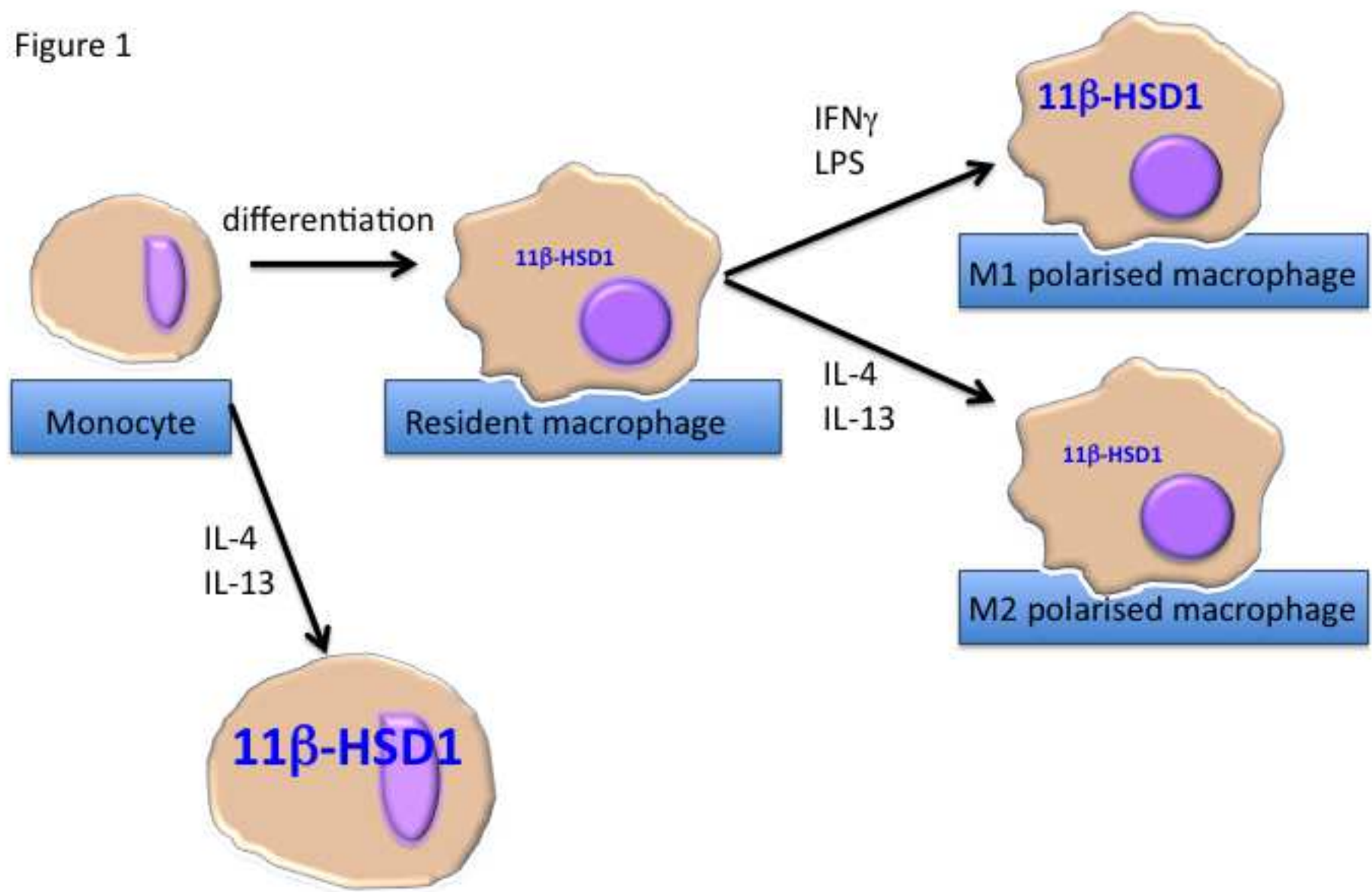
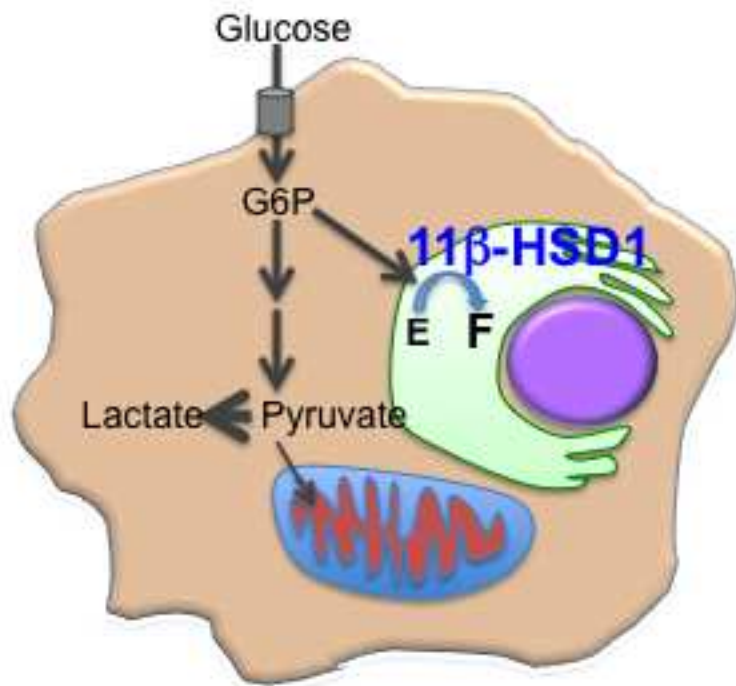
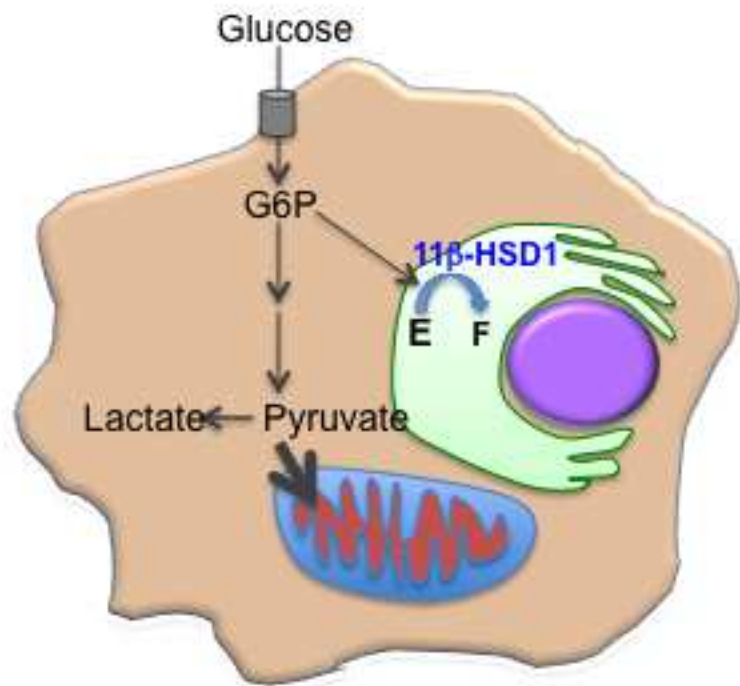


Figure 2



M1 polarised macrophage

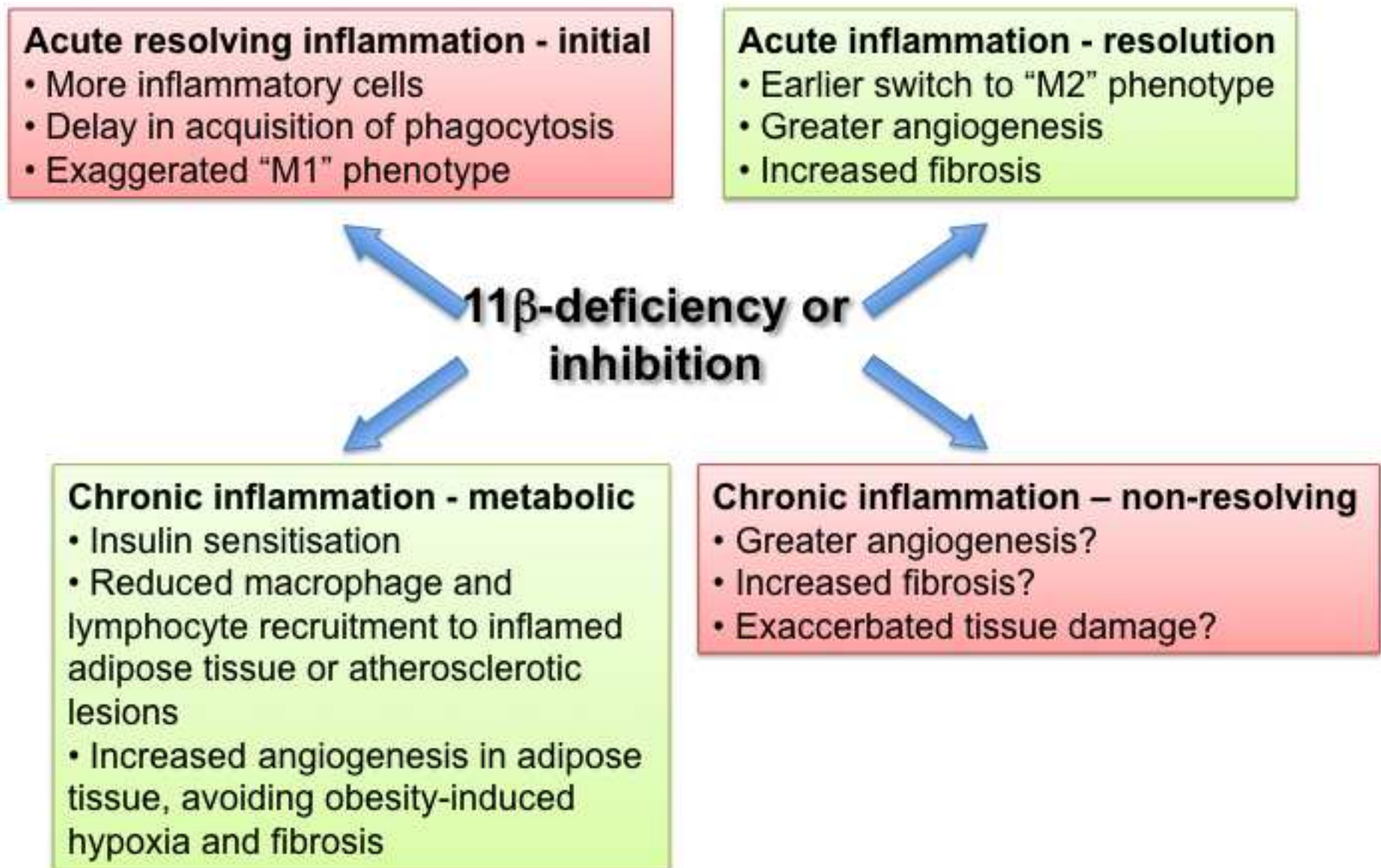
- ↑ Glycolysis
- ↓ O₂ consumption
- ↑ 11β-HSD1



M2 polarised macrophage

- ↓ Glycolysis
- ↑ O₂ consumption
- ↓ 11β-HSD1

Figure 3



Highlights

- !! 11 β -HSD1 converts inert glucocorticoids into active forms, amplifying glucocorticoid action
- !! 11 β -HSD1 is markedly induced by pro-inflammatory cytokines
- !! 11 β -HSD1 deficiency/inhibition worsens acute inflammation
- !! 11 β -HSD1 inhibition reduces inflammation in obesity or atherosclerosis
- !! an increased angiogenic response may underlie some of the benefits

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