

Maternal effector T cells within decidua:

Lissauer, D.; Kilby, M. D.; Moss, P.

DOI:

[10.1016/j.placenta.2017.09.003](https://doi.org/10.1016/j.placenta.2017.09.003)

License:

Creative Commons: Attribution-NonCommercial-NoDerivs (CC BY-NC-ND)

Document Version

Peer reviewed version

Citation for published version (Harvard):

Lissauer, D, Kilby, MD & Moss, P 2017, 'Maternal effector T cells within decidua: The adaptive immune response to pregnancy?', *Placenta*, vol. 60, pp. 140-144. <https://doi.org/10.1016/j.placenta.2017.09.003>

[Link to publication on Research at Birmingham portal](#)

General rights

Unless a licence is specified above, all rights (including copyright and moral rights) in this document are retained by the authors and/or the copyright holders. The express permission of the copyright holder must be obtained for any use of this material other than for purposes permitted by law.

- Users may freely distribute the URL that is used to identify this publication.
- Users may download and/or print one copy of the publication from the University of Birmingham research portal for the purpose of private study or non-commercial research.
- User may use extracts from the document in line with the concept of 'fair dealing' under the Copyright, Designs and Patents Act 1988 (?)
- Users may not further distribute the material nor use it for the purposes of commercial gain.

Where a licence is displayed above, please note the terms and conditions of the licence govern your use of this document.

When citing, please reference the published version.

Take down policy

While the University of Birmingham exercises care and attention in making items available there are rare occasions when an item has been uploaded in error or has been deemed to be commercially or otherwise sensitive.

If you believe that this is the case for this document, please contact UBIRA@lists.bham.ac.uk providing details and we will remove access to the work immediately and investigate.

Accepted Manuscript

Maternal effector T cells within decidua: The adaptive immune response to pregnancy?

D. Lissauer, M.D. Kilby, P. Moss



PII: S0143-4004(17)31107-4

DOI: [10.1016/j.placenta.2017.09.003](https://doi.org/10.1016/j.placenta.2017.09.003)

Reference: YPLAC 3724

To appear in: *Placenta*

Received Date: 1 May 2017

Revised Date: 25 August 2017

Accepted Date: 7 September 2017

Please cite this article as: Lissauer D, Kilby MD, Moss P, Maternal effector T cells within decidua: The adaptive immune response to pregnancy?, *Placenta* (2017), doi: 10.1016/j.placenta.2017.09.003.

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

1 **Maternal effector T cells within decidua: the adaptive immune**
2 **response to pregnancy?**

3
4
5 **Lissauer D¹, Kilby, MD¹. and Moss, P^{2,3}**

6
7
8 ¹ Centre for Women's and Newborn's Health and Centre of Endocrinology, Diabetes and
9 Metabolism [CEDAM], College of Medical and Dental Sciences, University of Birmingham
10 and Birmingham Health Partners, UK

11 ² Institute of Immunology and Immunotherapy, College of Medical and Dental Sciences,
12 University of Birmingham and Birmingham Health Partners, UK

13 ³ Corresponding Author. p.moss@bham.ac.uk

14
15
16

17

18 **Abstract**

19 In human pregnancy the maternal immune system plays a critical role in the
20 regulation of many aspects of human reproduction including implantation,
21 placentation and defence against infection. Interest has been focussed on the role of
22 uterine natural killer cells (uNK) in the maternal decidua whereas effector CD4+ and
23 CD8+ T cells have received much less attention despite the observation that they
24 represent a major proportion of decidual leucocytes in the latter phase of pregnancy.
25 A range of recent studies have demonstrated that human decidual T cells are highly
26 differentiated, express a range of cytokines and cytotoxic markers, and demonstrate
27 a unique transcriptional profile characterised by high level expression of genes
28 involved in interferon-signalling. Moreover, subpopulations of effector T cells
29 demonstrate specificity for fetal tissue and are regulated through expression of
30 inhibitory checkpoint proteins and T regulatory cells. Nevertheless, many questions
31 remain to be answered, such as the potential role of maternal effector T cells in
32 either supporting successful pregnancy or potentially clearing fetal cells that have
33 entered the maternal circulation. In addition, there is an increasing interest in the role
34 of maternal effector T cells in the pathogenesis of disorders such as chronic villitis
35 miscarriage, stillbirth, fetal growth restriction and pre-eclampsia. Current debates in
36 relation to these questions will be discussed within this review.

37

38

**39 Maternal effector T cells comprise the major population of leucocytes within
40 decidua by the end of pregnancy**

41 The maintenance of a semi-allogeneic fetus within the mother represents a
42 considerable challenge to the maternal immune system during pregnancy [1]. A
43 wide range of mechanisms have been postulated as being important and have
44 evolved in order to limit maternal immune recognition of fetal tissue. One of the most
45 straightforward approaches might have been to exclude effector CD4+ and CD8+ T
46 cells from the decidual bed. However, effector T cells represent around 60% of the T
47 cell pool in the later stages of pregnancy [2] and whilst NK cells are relatively more
48 numerous in the early stages of pregnancy their numbers remain stable whilst those
49 of T cells show a gradual increase with advancing gestation [2,3]. As such this
50 temporal replacement of NK cells by T cells mirrors the kinetics of a peripheral
51 adaptive immune response and the uterine environment may perhaps be seen to
52 recapitulate short term lymphocyte dynamics over a 9 month period [4]. CD45RO+
53 effector T cells comprise around 60% of the decidual T cell repertoire at term whilst
54 representing only 30% of T cells within blood. This relative increase in 'antigen-
55 experienced' T cells could potentially indicate evidence of local activation although it
56 may also reflect selective recruitment of effector cells into decidual tissue. In this
57 regard it is important to compare the phenotypic and functional features of T cells in
58 the two compartments and this is shown in Table 1. This reveals a number of
59 differences between effector cells in decidua and blood, including the observation
60 that decidual effector cells are more highly differentiated than peripheral cells, with
61 over 40% of such cells demonstrating a CD27-CD28- phenotype compared to less
62 than 20% in blood [5].

63 This pattern of local activation of decidual T cells might be taken to represent
64 recognition of fetal tissue. Fetal trophoblast cells downregulate the expression of
65 HLA-A and HLA-B, and observation that mismatch of polymorphic HLA-C alleles
66 between mother and fetus was associated with increased levels of T cell activation
67 provided some of the first support for this hypothesis [6].

68

**69 The CXCL10-CXCR3 axis is important in attracting effector T cells to decidual
70 tissue**

71 Chemokines are important regulators of leukocyte migration and are therefore likely
72 to play a major role in reproductive biology. CXCR3 is a receptor for inflammatory
73 chemokines and studies within pregnant mice have shown that it undergoes
74 epigenetic silencing on T cells within decidua [7], although this may be overcome in
75 the setting of local inflammation [8]. In contrast our findings reveal CXCR3 to be
76 expressed on 17% of human decidual CD4+ T cells, one of many differences found
77 between the immunological environment between mice and humans [5]. CXCL10, an
78 important ligand for CXCR3, is strongly expressed and identifies the CXCL10-
79 CXCR3 axis as an important mediator of effector cell migration into decidual tissue.

80

81 **Maternal effector T cells within decidua can recognize fetal tissue**

82 It is now clear that an adaptive immune response against fetal tissue develops in
83 most, if not all, human pregnancies. This is indicated most clearly in the development
84 of alloreactive HLA-specific antibodies and the development of sensitive assays has
85 revealed that these are found in most mothers following a single pregnancy [9].
86 Moreover, the presence of IgG isotypes reveals that antigen-specific T cell 'help' has
87 also been established. The potential importance of these antibodies in relation to
88 fetal health is uncertain and such responses may simply reflect an epiphenomenon
89 that is of no consequence to pregnancy outcome. However some studies do reveal a
90 weak clinical association and HLA-C specific antibodies have been shown to be
91 more common in women with recurrent fetal loss (miscarriage) [10].

92 Given this observation, it is perhaps not surprising that maternal cellular adaptive
93 immune responses against fetal tissue are also generated during human pregnancy.
94 Culture and expansion of maternal T cells *in vitro* first identified cells that were able
95 to recognise paternal cells [11] and these observations have been substantiated by
96 techniques that directly visualise T cells with alloreactive potential. Indeed our own
97 work [12, 13], and that of others [14-16], has identified HY-specific CD8+ T cells in
98 the maternal circulation following male pregnancies. These CD8 T cells are present
99 for many years following pregnancy and can be reactivated *in vitro* to generate highly
100 cytotoxic T cells that lyse male cells. HY-specific CD8+ cells were detectable in 32%
101 of women following a single male pregnancy and this proportion rose to 50% of
102 those with 2 or more male pregnancies, indicating that alloreactive cellular immunity
103 is boosted by recurrent episodes of fetal microchimerism. Until recently, it has not
104 been possible to directly identify T cells with fetal specificity within decidual tissue but

105 we have also utilized HLA-peptide multimer technology to identify HY-specific CD8 T
106 cells in decidual tissue [5]. Indeed, the frequency of such cells is greatly increased
107 compared to peripheral blood and indicates that cytotoxic cells with specificity for
108 fetal tissue are localised in direct anatomical contact.

109

110 **Effector T cells within decidua display a novel profile of functional activity**

111 Tilbergs et al studied CD8+ effector T cells within decidual tissue and demonstrated
112 a unique Th1 pattern of high level IFN γ expression together with low levels of
113 perforin and granzyme [17]. Our own studies of decidual CD4 and CD8+ T cells
114 confirmed IFN γ expression in many cells but also revealed expression of IL-4 in a
115 minority population. In particular whilst IFN γ expression was observed in 60% of
116 CD8+ T cells, 1.2% of cells also expressed IL-4, a value which, whilst relatively
117 modest, was higher than expression within 0.7% of CD8+ cells within maternal
118 peripheral blood. Comparable values for CD4+ T cells were 25% and 5%
119 respectively and IL-4 expression was markedly higher than on maternal T cells from
120 peripheral blood [5]. Interestingly, IL-4 expression can be induced from peripheral T
121 cells following incubation with progesterone and this 'T_{prog}' phenotype may therefore
122 partially reflect the effect of the local hormonal microenvironment [18, 19].

123 We recently completed a comparative transcriptional analysis of effector CD4 and
124 CD8+ T cells from decidua and maternal peripheral blood. A wide range of genes
125 were differentially expressed in decidual T cells with a striking upregulation of those
126 which encode proteins involved in the signalling response to interferon [5]. This
127 profile is highly unusual within effector T cells and suggests that decidual tissue is
128 characterized by high levels of local interferon production. This subject has been
129 relatively poorly studied although immunohistochemical expression of type 1
130 interferon has been observed within cells of the monocytic lineage [20,21]. It is
131 interesting to speculate on what may drive interferon production but it is well
132 established that endogenous retroviruses play an important role in the generation of
133 syncytiotrophoblast [22-26]. Notably, the *Syncytin-1* protein encoded from the *env*
134 gene of *ERVW-1* has an essential role in formation of the syncytiotrophoblast and is
135 released into the periphery via placental microvesicles which are themselves able to
136 illicit a T cell response [27].

137 **Effector T cells within decidua demonstrate specificity for pathogens**

138 Pregnancy is associated with altered regulation of immune responses that can
139 potentially increase susceptibility to some infectious diseases. The increased
140 mortality rate of pregnant women following avian influenza infection is one such
141 example. As such, it is not surprising that T cells within decidual tissue exhibit
142 specificity for local pathogens [28]. T cells with specificity for cytomegalovirus and
143 Epstein-Barr virus are preferentially recruited into decidua and mediate pathogen
144 surveillance of maternal cells [29]. Importantly, these populations recognise peptides
145 restricted by HLA-A and HLA-B alleles which are themselves not presented on fetal
146 trophoblast. Recent investigations have also shown that effector cells can recognise
147 peptides restricted through HLA-C alleles, potentially indicating efficacy in control of
148 infected fetal tissue [30].

149

150 **The function of effector T cells is modulated through intrinsic and extrinsic** 151 **regulation**

152 The finding of large numbers of functional effector T cells within decidual tissue
153 raises the question of how such cells are regulated in order to limit potential
154 immunopathology or fetal damage. In this regard extrinsic regulation mediated
155 through T regulatory cells and cell-intrinsic expression of inhibitory checkpoint
156 proteins are both emerging as important control mechanisms.

157 T regulatory cells are increased within decidual tissue at term pregnancy and can
158 comprise over 20% of all CD4⁺ T cells [31-33]. Indeed, murine experiments have
159 shown that depletion of T regulatory cells can trigger fetal rejection [34] and it has
160 been suggested that the evolution of the FoxP3⁺ T regulatory cell was a key event in
161 the development of eutherian reproduction [35]. In line with previous reports [33], we
162 also find that decidual T cells proliferate in response to cord blood lymphocytes and
163 that this is increased following depletion of T regulatory cells [5]. These observations
164 indicate that such decidual populations do have specificity for maternal antigens and
165 that this is at least partly regulated through the action of autologous T regulatory
166 populations.

167 Checkpoint proteins such as PD-1 and TIM-3 are now considered to be amongst the
168 most important molecules within clinical medicine [36, 37]. This is due to the
169 dramatic efficacy of antibody-mediated blockade of PD-1 function in the treatment of
170 solid tumours [38]. PD-1 expression on T cells is often taken to represent an

171 'exhausted' state and is believed to reflect cells that have undergone repeated
172 stimulation within antigen. However, despite the considerable success of checkpoint
173 blockade in cancer therapy there is less understanding as to the physiological role of
174 checkpoint proteins in human T cell physiology. Interestingly, a high level of
175 checkpoint protein expression is observed on T cells within decidua [39]. Wang et al
176 reported large populations of Tim-3+PD-1+ CD8+ T cells within decidua during early
177 human pregnancy and showed that incubation of CD8 T cells with trophoblast led to
178 further checkpoint upregulation [40,41]. Interestingly, PD-L1 is expressed on
179 syncytiotrophoblast, as well as intermediate trophoblastic cells located in the chorion
180 laeve and implantation site [42], and some studies have indicated that PD-1
181 blockade in pregnant mice may result in fetal loss [43]. Of note, these observations
182 provide further evidence that the study of reproductive immunology will be of huge
183 importance in understanding tumour immunology and lend support to the concept of
184 cancer as a 'somatic pregnancy' [44].

185

186 **Fetal-specific maternal T cells may play a role in limiting fetal microchimerism**

187 It is now generally accepted that pregnancy leads to a state of microchimerism within
188 the mother in which significant amounts of fetal tissue and cells are released into the
189 maternal circulation [45]. Moreover, these fetal cells have been shown to survive for
190 long periods within the mother and have even been implicated in a range of clinical
191 disorders such as thyroiditis. Whilst such fetal cells may provide potential benefit to
192 the mother, such as potentially supporting repair of maternal tissue, it would seem
193 reasonable that such chimerism would need to be controlled by the maternal
194 immune system. In this regard, the humoral and cellular maternal response against
195 fetal tissue may have an important role in the suppression of fetal chimerism.
196 Indeed, some support for this hypothesis comes from the observation that the
197 degree of chimerism has been reported to fall with repeated pregnancies, whilst the
198 magnitude of fetal-specific immunity appears to increase [46]. These observations
199 could provide important insights into the regulation of chimerism in disorders such as
200 transplantation.

201

202 **Maternal T cell responses against fetal tissue may be associated with obstetric** 203 **complications**

204 The observation that maternal T cells demonstrate antigenic specificity for fetal
205 tissue raises the obvious question as to whether these may be implicated in the
206 pathogenesis of pregnancy complications. Relatively little evidence exists to
207 implicate maternal T cell responses in the development of pre-eclampsia and this
208 may reflect the fact that the cardinal feature of impaired trophoblast invasion of
209 maternal spiral arteries is determined early within pregnancy and at a time when
210 natural killer cells dominate the cellular infiltrate. More recently, Leavey et al have
211 suggested that T cell mediated pathology may indeed be related to a subset of
212 mothers with preeclampsia who demonstrate a form of disorder characterized by
213 poor fetal outcome and growth restriction but relatively less impact on maternal
214 health [47].

215 Despite this, fetal-specific immunity is strongly implicated in the development of
216 chronic villitis ('villitis of uncertain/unknown aetiology'; VUE) which is a relatively
217 common cause of fetal growth restriction and pre-term delivery [48-51]. VUE is
218 characterized by an inflammatory cell infiltrate of placental macrophages and T cells
219 within the villi which develops in the absence of infection. A specific feature is non-
220 uniform involvement of villi and ultimately this can lead to "obliterative fetal
221 vasculopathy". Interestingly, this maternal infiltrate typically shows a CD8:CD4 ratio
222 around 3:1 and T cells comprise almost half of the cellular infiltrate. It has been
223 suggested that such villitis may represent relatively uncontrolled maternal immune
224 recognition of fetal tissue and this is supported by the finding that antibodies against
225 fetal tissue are commonly observed in this disorder and associate with the deposition
226 of C4 complement components. However caution is needed in this interpretation as
227 some degree of VUE can be observed in around 10% of pregnancies and as such
228 this process might even be regarded as a 'normal variant'.

229

230 **Maternal effector T cells represent a fascinating topic for future investigation**

231 Within a relatively short period of time conventional wisdom has moved from the
232 belief that the placenta represents an effective barrier between the mother and fetus
233 to an understanding that cells traffic between circulations and immune cells play an
234 important role in supporting pregnancy. Decidual NK cells have been shown to
235 possess a range of unique functional properties that represent novel
236 immunotherapeutic opportunities [52] (Figure 1). Investigation into the physiological
237 and potential immunopathological role of maternal alloreactive T cells has followed

238 more slowly. Despite this, it is now clear that maternal effector CD45RO+ T cells
239 represent the majority of T cells within decidual tissue in the latter stages of
240 pregnancy and display a range of novel properties. It remains possible that the
241 primary role of these cells lies in the control of local infection with no major
242 significance in relation to reproductive outcome. Nevertheless, further investigation
243 into the specificity and unique properties of these remarkable cells may uncover
244 novel insights into the physiology of human placentation, the pathogenesis of
245 reproductive disorders and offer clues towards a range of additional disorders such
246 as cancer and transplantation biology.
247

248

249

250 **Acknowledgement**

251 Presented at the 2016 Global Pregnancy Collaboration (CoLab) conference in
252 Oxford, England. The Global Pregnancy Collaboration is part of the Pre-eclampsia-
253 Eclampsia Monitoring, Prevention & Treatment (PRE-EMPT) initiative funded by the
254 University of British Columbia, a grantee of the Bill & Melinda Gates Foundation.

255

256

257 **References**

258

259 1. Moffett, A., and C. Loke, Immunology of placentation in eutherian mammals, *Nat.*
260 *Rev. Immunol.* 6: (2006) 584–594.

261 2. Stallmach T, Hebisch G, Orban P and Lu X.

262 Aberrant positioning of trophoblast and lymphocytes in the fetomaternal
263 interface with pre-eclampsia. *Virchows Arch.* (1999) 434(3):207-11.

264 3. Sykes, L., D. A. MacIntyre, X. J. Yap, T. G. Teoh, P. R. Bennett, L. Sykes, D. A.
265 MacIntyre, X. J. Yap, T. G. Teoh, and P. R. Bennett, The Th1:Th2 Dichotomy of
266 Pregnancy and Preterm Labour. *Mediators Inflamm.* (2012): 1–12.

267 4. Williams, P. J., R. F. Searle, S. C. Robson, B. A. Innes, and J. N. Bulmer.
268 Decidual leucocyte populations in early to late gestation normal human pregnancy. *J.*
269 *Reprod. Immunol.* (2009) 82: 24–31.

270 5. Powell R, Lissauer D, Tamblyn J, Beggs A, Cox P, Moss P and Kilby MD. Decidual
271 T cells exhibit a highly differentiated phenotype and demonstrate potential fetal-
272 specificity and a strong transcriptional response to interferon γ . *J. Immunol.* In Press.

273 6. Tilburgs, T., S. A. Scherjon, B. J. van der Mast, G. W. Haasnoot, M. Versteeg-V D
274 Voort-Maarschalk, D. L. Roelen, J. J. van Rood, and F. H. J. Claas. Fetal-maternal
275 HLA-C mismatch is associated with decidual T cell activation and induction of
276 functional T regulatory cells. *J. Reprod. Immunol.* (2009) 82: 148–57.

277 7. Nancy, P., and A. Erlebacher, Epigenetic repression of chemokine expression at
278 the maternal-fetal interface as a mechanism of fetomaternal tolerance. *Médecine*
279 *Sci. M/S* 28: (2012) 1037–9.

280 8. Chaturvedi V, Ertelt JM, Jiang TT, Kinder JM, Xin L, Owens KJ, Jones HN, Way
281 SS. CXCR3 blockade protects against *Listeria monocytogenes* infection-induced
282 fetal wastage. *J Clin Invest.* (2015) 125(4):1713-25.

283 9. Masson E, Vidal C, Deschamps M, Bongain S, Thevenin C, Dupont I, Rietmulher
284 D, Pouthier F, Mongaillard G, Chabod J, Ferrand C, Tiberghien P, Rebibou JM,

- 285 Incidence and risk factors of anti-HLA immunization after pregnancy. *Hum*
286 *Immunol.* (2013) Aug;74(8):946-51.
287
- 288 10. Meuleman T, van Beelen E, Kaaja RJ, van Lith JM, Claas FH, Bloemenkamp
289 KW, HLA-C antibodies in women with recurrent miscarriage suggests that antibody
290 mediated rejection is one of the mechanisms leading to recurrent miscarriage. *J*
291 *Reprod Immunol.* (2016) 116:28-34.
- 292 11. van Kampen CA, Versteeg-van der Voort Maarschalk MF, Langerak-Langerak J,
293 van Beelen E, Roelen DL, Claas FH, Pregnancy can induce long-persisting primed
294 CTLs specific for inherited paternal HLA antigens. *Hum*
295 *Immunol.* (2001) Mar;62(3):201-7
- 296 12. Lissauer, D., K. Piper, O. Goodyear, M. D. Kilby, and P. A. H. Moss, Fetal-
297 specific CD8+ cytotoxic T cell responses develop during normal human pregnancy
298 and exhibit broad functional capacity. *J. Immunol.* (2012) 189: 1072–80.
- 299 13. Piper, K. P., A. McLarnon, J. Arrazi, C. Horlock, J. Ainsworth, M. D. Kilby, W. L.
300 Martin, and P. Moss, Functional HY-specific CD8+ T cells are found in a high
301 proportion of women following pregnancy with a male fetus. *Biol. Reprod.* (2007) 76:
302 96–101.
- 303 14. Verdijk, R. M., A. Kloosterman, J. Pool, M. van de Keur, A. M. I. H. Naipal, A. G.
304 S. van Halteren, A. Brand, T. Mutis, and E. Goulmy, Pregnancy induces minor
305 histocompatibility antigen-specific cytotoxic T cells: implications for stem cell
306 transplantation and immunotherapy. *Blood* (2004) 103: 1961–4.
- 307 15. Verdijk, R. M., A. Kloosterman, J. Pool, M. van de Keur, A. M. I. H. Naipal, A. G.
308 S. van Halteren, A. Brand, T. Mutis, and E. Goulmy, Pregnancy induces minor
309 histocompatibility antigen-specific cytotoxic T cells: implications for stem cell
310 transplantation and immunotherapy. *Blood* (2004) 103.
- 311 16. van Halteren, A. G. S., E. Jankowska-Gan, A. Joosten, E. Blokland, J. Pool, A.
312 Brand, W. J. Burlingham, and E. Goulmy. Naturally acquired tolerance and
313 sensitization to minor histocompatibility antigens in healthy family members. *Blood*
314 (2009) 114.

- 315 17. Tilburgs, T., D. Schonkeren, M. Eikmans, N. M. Nagtzaam, G. Datema, G. M.
316 Swings, F. Prins, J. M. van Lith, B. J. van der Mast, D. L. Roelen, S. A. Scherjon,
317 and F. H. Claas. Human decidual tissue contains differentiated CD8+ effector-
318 memory T cells with unique properties. *J. Immunol.* (2010) 185: 4470–7.
- 319 18. Lissauer, D., S. A. Eldershaw, C. F. Inman, A. Coomarasamy, P. A. H. Moss,
320 and M. D. Kilby, Progesterone promotes maternal-fetal tolerance by reducing human
321 maternal T-cell polyfunctionality and inducing a specific cytokine profile. *Eur. J.*
322 *Immunol.* (2015) 45: 2858–72.
- 323 19. Raghupathy, R., E. Al Mutawa, M. Makhseed, F. Azizieh, and J. Szekeres-
324 Bartho, Modulation of cytokine production by dydrogesterone in lymphocytes from
325 women with recurrent miscarriage. (2005) *BJOG* 112: 1096–101.
- 326 20. Chard, T., P. H. Craig, M. Menabawey, and C. Lee, Alpha interferon in human
327 pregnancy. *Br. J. Obstet. Gynaecol.* (1986) 93: 1145–9.
- 328 21. Khan, N. U., A. Gibson, and A. K. Foulis, The distribution of immunoreactive
329 interferon-alpha in formalin-fixed paraffin-embedded normal human foetal and infant
330 tissues. *Immunology* (1990) 71: 230–5.
- 331 22. Bannert, N., and R. Kurth, Retroelements and the human genome: new
332 perspectives on an old relation. *Proc. Natl. Acad. Sci. U. S. A.* (2004) 14572–9.
- 333 23. Tristem, M, Identification and characterization of novel human endogenous
334 retrovirus families by phylogenetic screening of the human genome mapping project
335 database. (2000) *J. Virol.* 74: 3715–30.
- 336 24. Blond, J. L., F. Besème, L. Duret, O. Bouton, F. Bedin, H. Perron, B. Mandrand,
337 and F. Mallet, Molecular characterization and placental expression of HERV-W, a
338 new human endogenous retrovirus family. *J. Virol.* (1999) 73: 1175–85.
- 339 25. McCoy, J. M., S. Mi, X. Lee, X. Li, G. M. Veldman, H. Finnerty, L. Racie, E.
340 LaVallie, X.-Y. Tang, P. Edouard, S. Howes, and J. C. Keith, Syncytin is a captive
341 retroviral envelope protein involved in human placentalmorphogenesis. *Nature*
342 (2000) 403: 785–789.
- 343 26. Kämmerer, U., A. Germeyer, S. Stengel, M. Kapp, and J. Denner, Human

- 344 endogenous retrovirus K (HERV-K) is expressed in villous and extravillous
345 cytotrophoblast cells of the human placenta. *J. Reprod. Immunol.* (2011) 91: 1–8.
- 346 27. Holder, B. S., C. L. Tower, K. Forbes, M. J. Mulla, J. D. Aplin, and V. M.
347 Abrahams, Immune cell activation by trophoblast-derived microvesicles is mediated
348 by syncytin 1. *Immunology* (2012) 136: 184–191.
- 349 28. Crespo ÂC, van der Zwan A, Ramalho-Santos J, Strominger JL, Tilburgs T,
350 Cytotoxic potential of decidual NK cells and CD8+ T cells awakened by infections. *J*
351 *Reprod Immunol.* (2017) Feb;119:85-90.
- 352 29. van Egmond A, van der Keur C, Swings GM, Scherjon SA, Claas FH, The
353 possible role of virus-specific CD8(+) memory T cells in decidual tissue. *J Reprod*
354 *Immunol.* (2016) Feb;113:1-8.
- 355 30. Crespo ÂC, Strominger JL, Tilburgs T. Expression of KIR2DS1 by decidual
356 natural killer cells increases their ability to control placental HCMV infection. *Proc*
357 *Natl Acad Sci U S A.* (2017) 113(52):15072-15077.
358
- 359 31. Rowe, J. H., J. M. Ertelt, L. Xin, and S. S. Way, Pregnancy imprints regulatory
360 memory that sustains anergy to fetal antigen. *Nature* (2012) 490: 102–6.
- 361 32. Aluvihare, V. R., M. Kallikourdis, and A. G. Betz, Regulatory T cells mediate
362 maternal tolerance to the fetus. *Nat. Immunol.* (2004) 5: 266–71.
- 363 33. Tilburgs, T., D. L. Roelen, B. J. van der Mast, G. M. de Groot-Swings, C.
364 Kleijburg, S. A. Scherjon, and F. H. Claas. Evidence for a selective migration of
365 fetus-specific CD4+CD25bright regulatory T cells from the peripheral blood to the
366 decidua in human pregnancy. *J. Immunol.* (2008) 180: 5737–45.
- 367 34. Chen T, Darrasse-Jèze G, Bergot AS, Courau T, Churlaud G, Valdivia
368 K, Strominger JL, Ruocco MG, Chaouat G and Klatzmann D. Self-specific
369 memory regulatory T cells protect embryos at implantation in mice. *J*
370 *Immunol.* (2013) 191(5):2273-81.
- 371 35. Samstein, R. M., A. Arvey, S. Z. Josefowicz, X. Peng, A. Reynolds, R.
372 Sandstrom, S. Neph, P. Sabo, J. M. Kim, W. Liao, M. O. Li, C. Leslie, J. A.
373 Stamatoyannopoulos, and A. Y. Rudensky, Foxp3 exploits a pre-existent enhancer
374 landscape for regulatory T cell lineage specification. *Cell* (2012) 151: 153–66.

- 375 36. Liang, S. C., Y. E. Latchman, J. E. Buhlmann, M. F. Tomczak, B. H. Horwitz, G.
376 J. Freeman, and A. H. Sharpe, Regulation of PD-1, PD-L1, and PD-L2 expression
377 during normal and autoimmune responses. *Eur. J. Immunol.* (2003) 33: 2706–16.
- 378 37. Tanaka, K., M. J. Albin, X. Yuan, K. Yamaura, A. Habicht, T. Murayama, M.
379 Grimm, A. M. Waaga, T. Ueno, R. F. Padera, H. Yagita, M. Azuma, T. Shin, B. R.
380 Blazar, D. M. Rothstein, M. H. Sayegh, and N. Najafian, PDL1 is required for
381 peripheral transplantation tolerance and protection from chronic allograft rejection. *J.*
382 *Immunol.* (2007) 179: 5204–10.
- 383 38. Topalian, S. L., F. S. Hodi, J. R. Brahmer, S. N. Gettinger, D. C. Smith, D. F.
384 McDermott, J. D. Powderly, R. D. Carvajal, J. A. Sosman, M. B. Atkins, P. D.
385 Leming, D. R. Spigel, S. J. Antonia, L. Horn, C. G. Drake, D. M. Pardoll, L. Chen, W.
386 H. Sharfman, R. A. Anders, J. M. Taube, T. L. McMiller, H. Xu, A. J. Korman, M.
387 Jure-Kunkel, S. Agrawal, D. McDonald, G. D. Kollia, A. Gupta, J. M. Wigginton, and
388 M. Sznol. 2012. Safety, activity, and immune correlates of anti-PD-1 antibody in
389 cancer. *N. Engl. J. Med.* (2012) 366: 2443–54.
- 390 39. Guleria, I., A. Khosroshahi, M. J. Ansari, A. Habicht, M. Azuma, H. Yagita, R. J.
391 Noelle, A. Coyle, A. L. Mellor, S. J. Khoury, and M. H. Sayegh, A critical role for the
392 programmed death ligand 1 in fetomaternal tolerance. *J. Exp. Med.* (2005) 202.
- 393 40. Wang, S., X. Zhu, Y. Xu, D. Zhang, Y. Li, Y. Tao, H. Piao, D. Li, and M. Du.
394 Programmed cell death-1 (PD-1) and T-cell immunoglobulin mucin-3 (Tim-3)
395 regulate CD4+ T cells to induce Type 2 helper T cell (Th2) bias at the maternal-fetal
396 interface. *Hum. Reprod.* (2016) 31: 700–11.
- 397 41. Chabtini, L., B. Mfarrej, M. Mounayar, B. Zhu, I. Batal, P. J. Dakle, B. D. Smith,
398 O. Boenisch, N. Najafian, H. Akiba, H. Yagita, and I. Guleria, TIM-3 regulates innate
399 immune cells to induce fetomaternal tolerance. *J. Immunol.* (2013) 190: 88–96.
- 400 42. Veras, E., R. J. Kurman, T.-L. Wang, and I.-M. Shih, PD-L1 Expression in
401 Human Placentas and Gestational Trophoblastic Diseases. *Int. J. Gynecol. Pathol.*
402 (2017). Mar;36(2):146-153
- 403 43. Wang, S.-C., Y.-H. Li, H.-L. Piao, X.-W. Hong, D. Zhang, Y.-Y. Xu, Y. Tao, Y.
404 Wang, M.-M. Yuan, D.-J. Li, and M.-R. Du, PD-1 and Tim-3 pathways are associated

405 with regulatory CD8+ T-cell function in decidua and maintenance of normal
406 pregnancy. (2015) *Cell Death Dis.* 6: e1738.

407 44. Old, LJ. Cancer is a somatic cell pregnancy. *Cancer Immun.* (2007) Nov 6;7:19.

408 45. Bianchi DW. Progress in the genetic analysis of fetal cells circulating in maternal
409 blood. *Curr Opin Obstet Gynecol.* (1997) 9(2):121-5

410 46. Gammill HS, Guthrie KA, Aydelotte TM, Adams Waldorf KM, Nelson JL, Effect of
411 parity on fetal and maternal microchimerism: interaction of grafts within a host?
412 *Blood.* (2010) 116(15):2706-12.

413 47. Leavey, K., S. J. Benton, D. Gynspan, J. C. Kingdom, S. A. Bainbridge, and B.
414 J. Cox, Unsupervised Placental Gene Expression Profiling Identifies Clinically
415 Relevant Subclasses of Human Preeclampsia. *Hypertens.* (2016) 68: 137–47.

416 48. Kapur, P., D. Rakheja, A. M. Gomez, J. Sheffield, P. Sanchez, and B. B. Rogers,
417 Characterization of Inflammation in Syphilitic Villitis and in Villitis of Unknown
418 Etiology. *Pediatr. Dev. Pathol.* (2004) 7: 453–458.

419 49. Redline, R. W., and P. Patterson, Villitis of unknown etiology is associated with
420 major infiltration of fetal tissue by maternal inflammatory cells. *Am. J. Pathol.* (1993)
421 143: 473–9.

422 50. Redline, R. W, Villitis of unknown etiology: noninfectious chronic villitis in the
423 placenta. *Hum. Pathol.* (2007) 38: 1439–1446.

424 51. Tamblyn, J. A., D. M. Lissauer, R. Powell, P. Cox, and M. D. Kilby. The
425 immunological basis of villitis of unknown etiology – Review. *Placenta* (2013) 34:
426 846–855.

427 52. Faas MM, de Vos P. Uterine NK cells and macrophages in pregnancy. *Placenta.*
428 (2017) Aug;56:44-52.

429

430

431

432

ACCEPTED MANUSCRIPT

Figure 1. Representation of maternal effector T cells within decidua and their potential functions. These are likely to include (1) recognition of fetal antigen *or* virally-infected EVT through peptides on HLA-C alleles, (2) potential access to maternal blood vessels to limit systemic fetal chimerism and (3) potential to breach syncytiotrophoblast and damage fetal blood vessels

	Effector T cells within decidua	Effector T cells within maternal peripheral blood
Percentage of total T cell repertoire	~60%	~30%
Degree of differentiation	more differentiated (~40% CD27-CD28-)	less differentiated (~20% CD27-CD28-)
Pattern of cytokine production following stimulation with PMA/Ionomycin mitogen	CD4+ : IFN γ , 25%; Il-4, 5% CD8+ IFN γ , 60%; Il-4, 1.2%	CD4+ : IFN γ , 17%; Il-4, 2% CD8+ IFN γ , 41%; Il-4, 0.7%
Expression of checkpoint proteins	CD4+ 43% PD-1+ CD8+ 68% PD-1+	CD4+ 20% PD-1+ CD8+ 25% PD-1+
Frequency of fetal-specific T cells	Use of HLA-peptide multimers reveals increased numbers compared to blood	Rare – but potential role in controlling fetal chimerism
Differentially expressed genes	Increase in genes which mediate interferon signaling response	

Table 1. Comparison of the features of CD45RO+ effector T cells within decidua and maternal peripheral blood at term. Data from Powell et al, (Submitted).

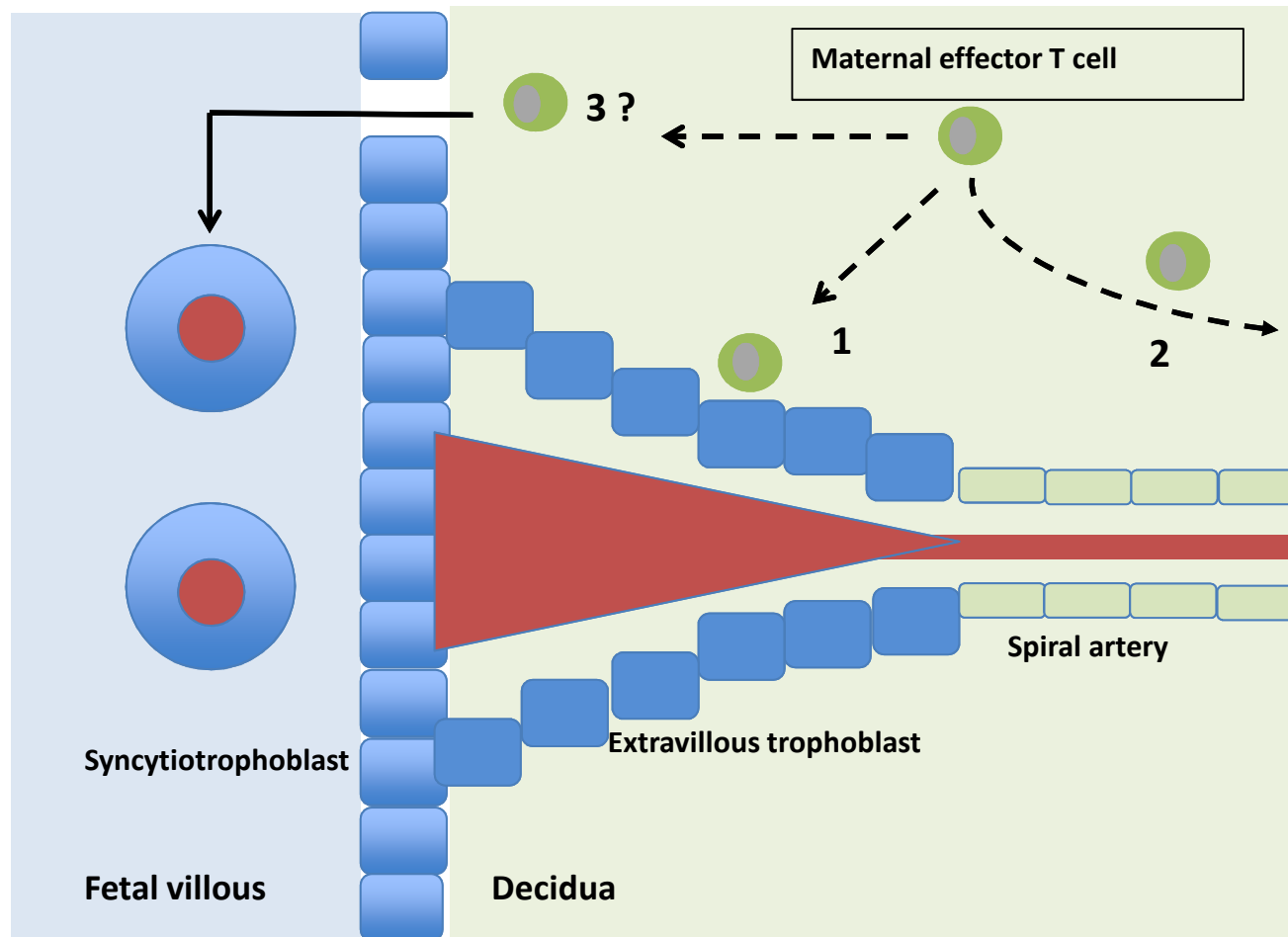


Figure 1. Representation of maternal effector T cells within decidua and their potential function. These are likely to include (1) recognition of fetal antigen *or* virally infected EVT through peptides on HLA-C alleles, (2) potential access to maternal blood vessels to limit systemic fetal chimerism and (3) potential to breach damaged syncytiotrophoblast and damage fetal blood vessels

Highlights

Maternal effector T cells within decidua: the adaptive immune response to pregnancy?

Lissauer D¹, Kilby, MD¹. and Moss, P^{2,3}

Highlights

CD45RO+ effector T cells comprise the majority of CD4+ and CD8+ T cells in decidua and are more highly differentiated than T cells in blood

Human effector T cells express CXCR3 which may guide cells to decidua

These cells include populations that can produce IFN γ or Il-4

Microarray shows that decidual T cells demonstrate a transcriptional response to interferon signalling

T cells proliferate to cord blood indicating a response to fetal antigen and this is increased when T regulatory cells are removed.