

Defining the roles of ADAM proteases in trophoblast biology

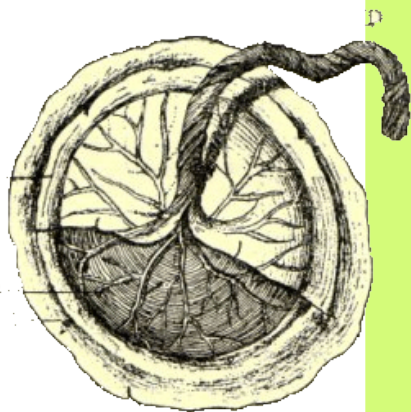
The University of Washington Placenta Symposium 2016

AG Beristain, PhD

The Child & Family Research Institute

The University of British Columbia

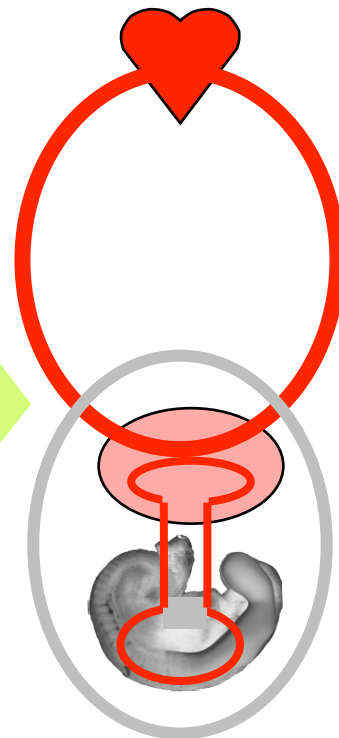
Vancouver, BC. Canada



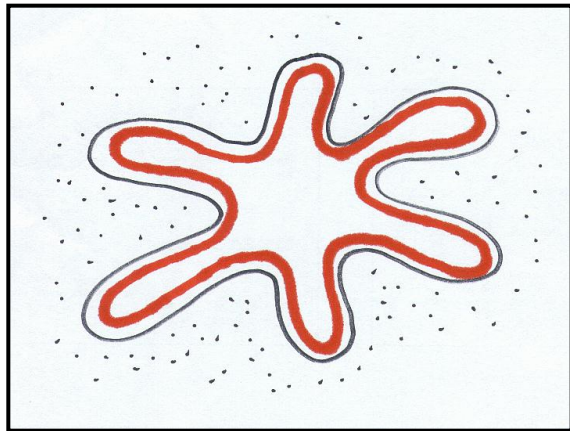
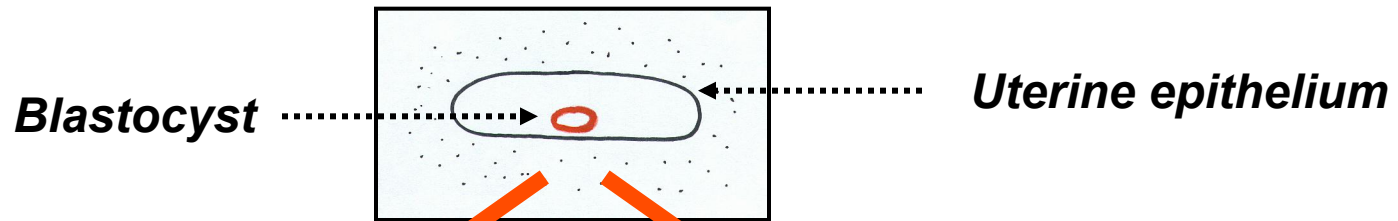
PLACENTA: *definition*

*An apposition of fetal and
maternal tissues for
physiological exchange*

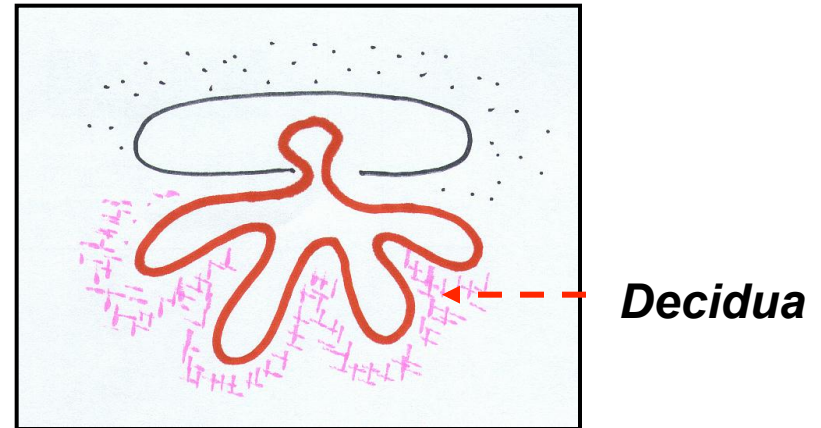
(Mossman, 1937)



Also Latin for “cake”



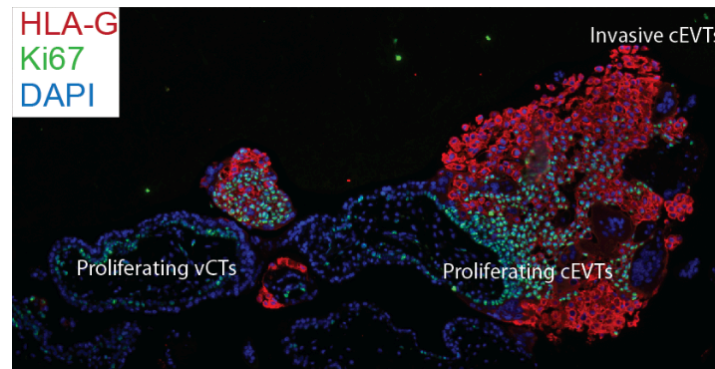
Noninvasive implantation
(Epitheliochorial placentation)



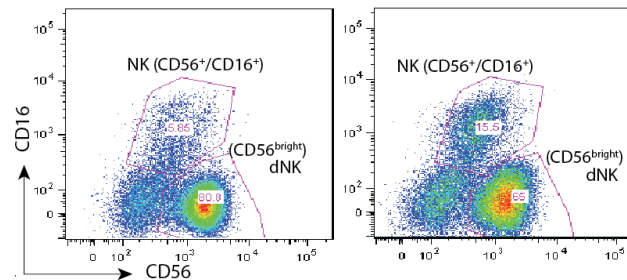
Invasive implantation
**(Endotheliochorial and
haemochorial placentation)**

Two main research streams in my lab...

- 1) Characterizing trophoblast (epithelial lineage) populations in developing placenta (fetal side)



- 2) Examine the importance of maternal immune cells in controlling trophoblast biology (mother's side)



Trophoblast team



Mahroo Aghababaei

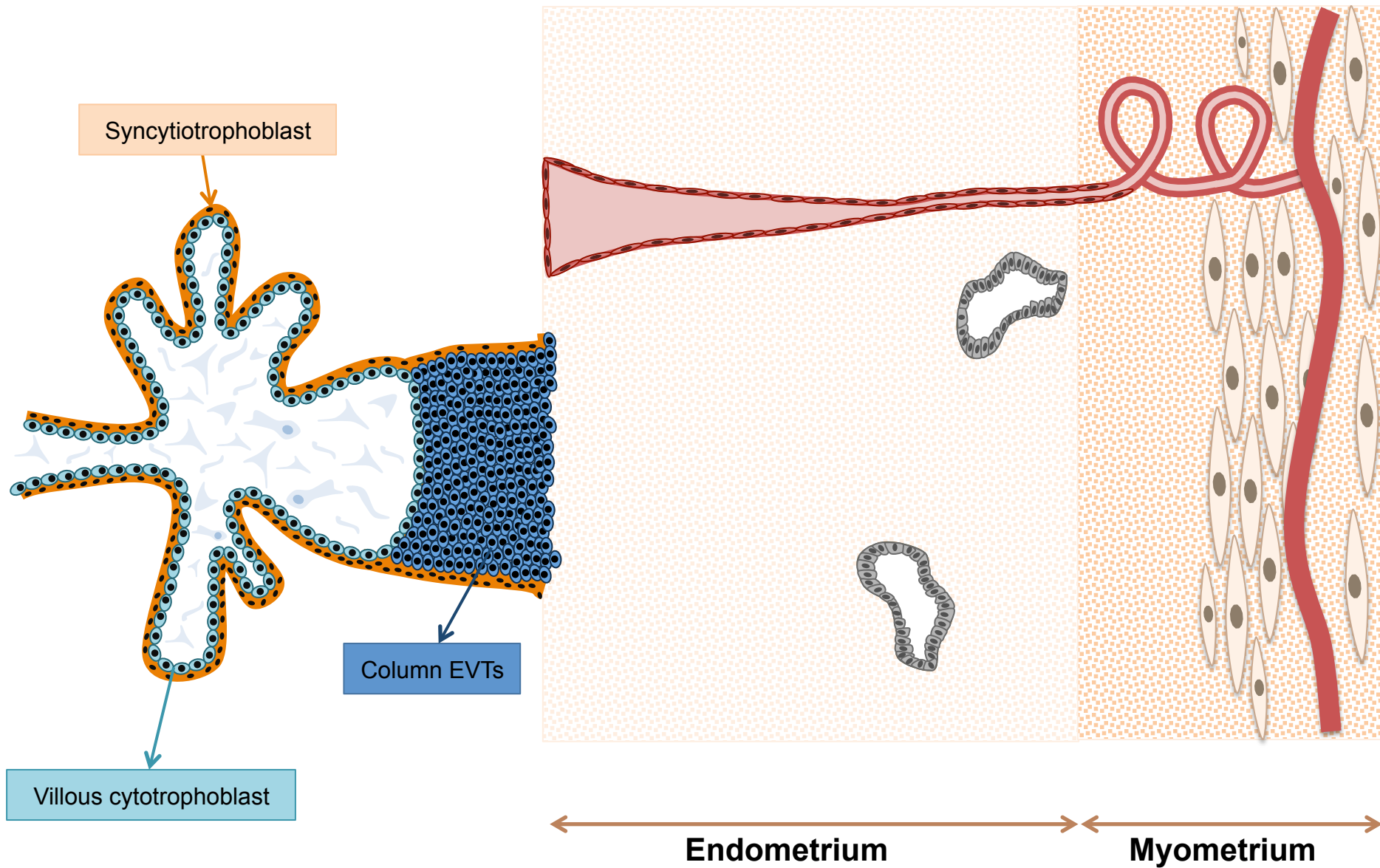


Lauren De Lucca

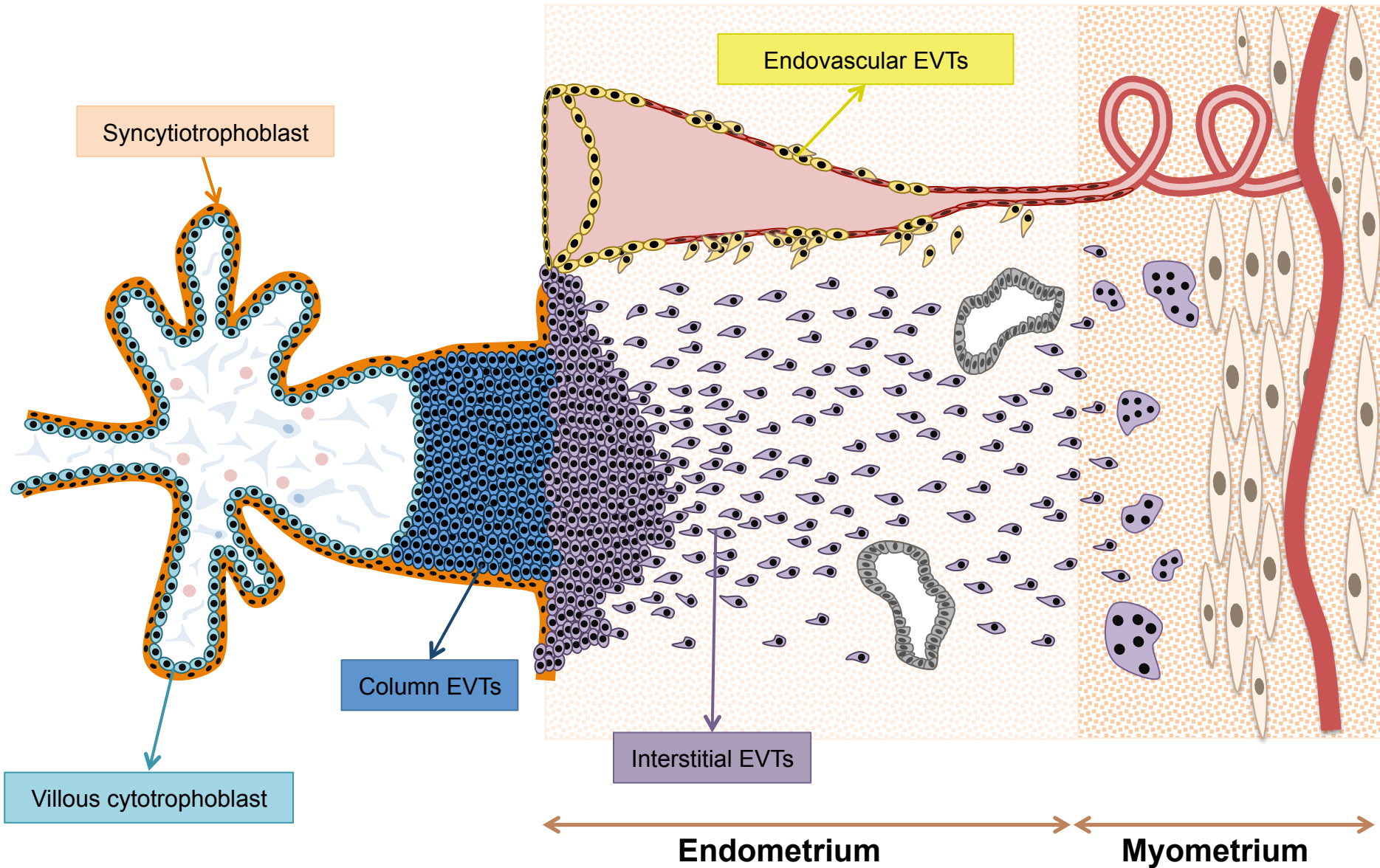


Hoa Le

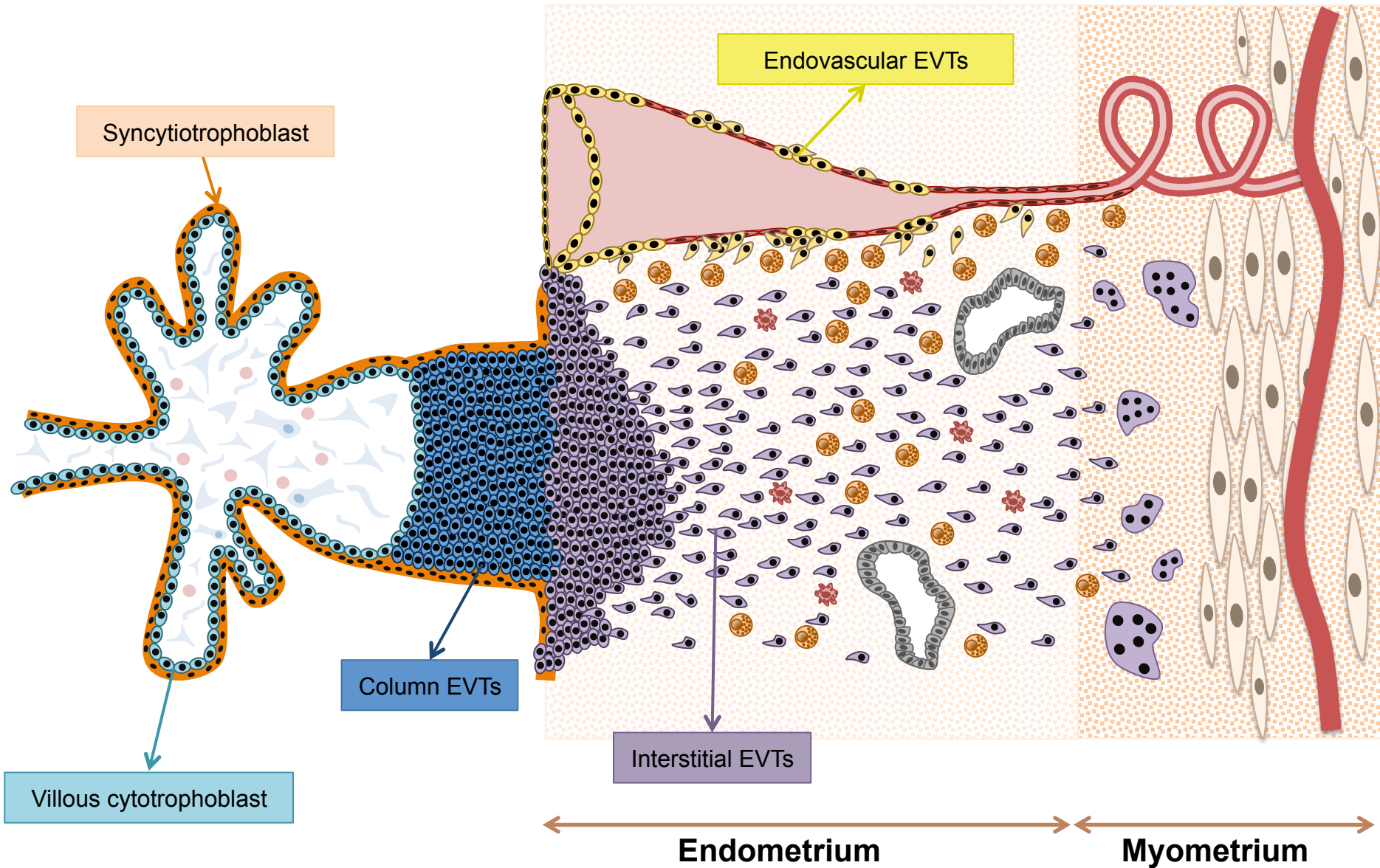
Establishment of the fetal-maternal interface



Establishment of the fetal-maternal interface



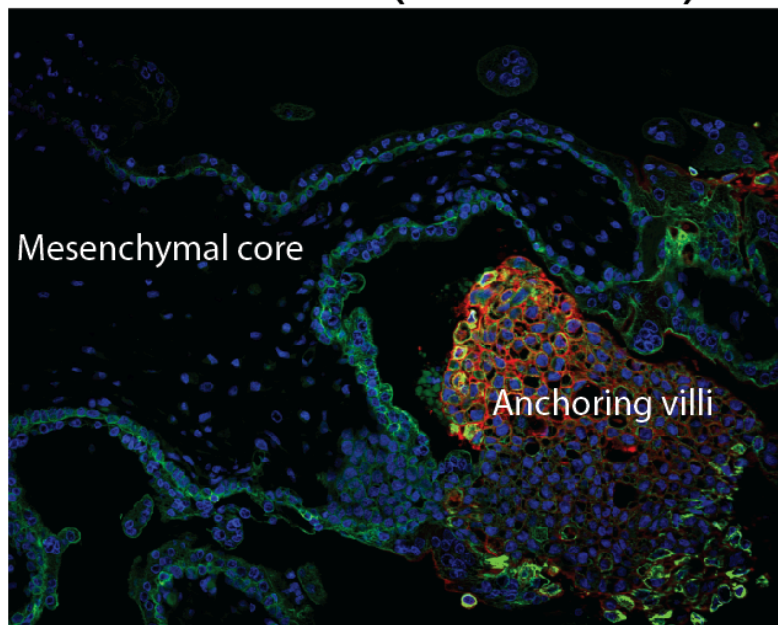
Establishment of the fetal-maternal interface



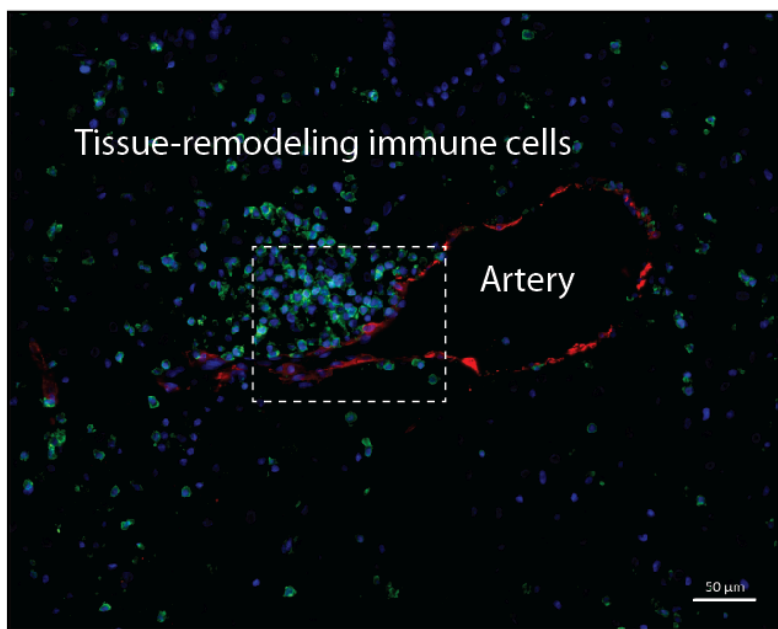
Placenta (10 wkGA)

Uterus (decidua)

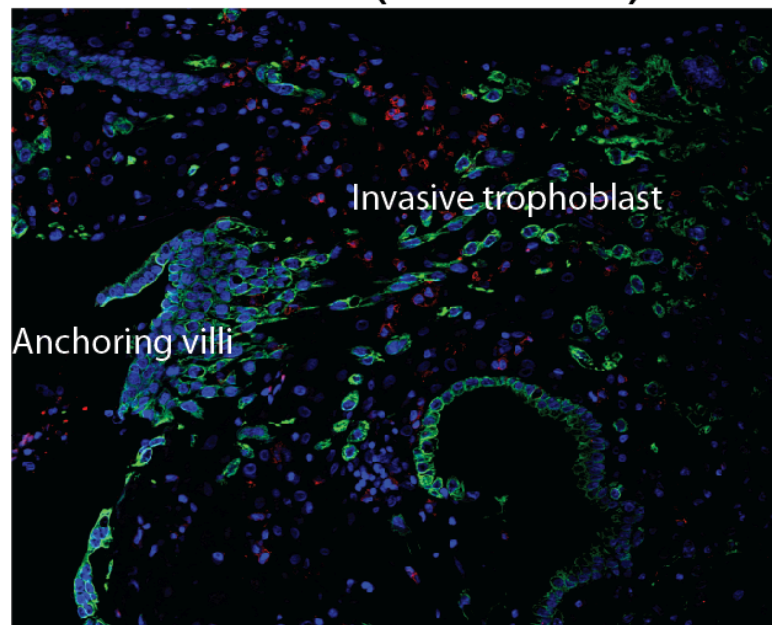
HLA-G
K7
DAPI



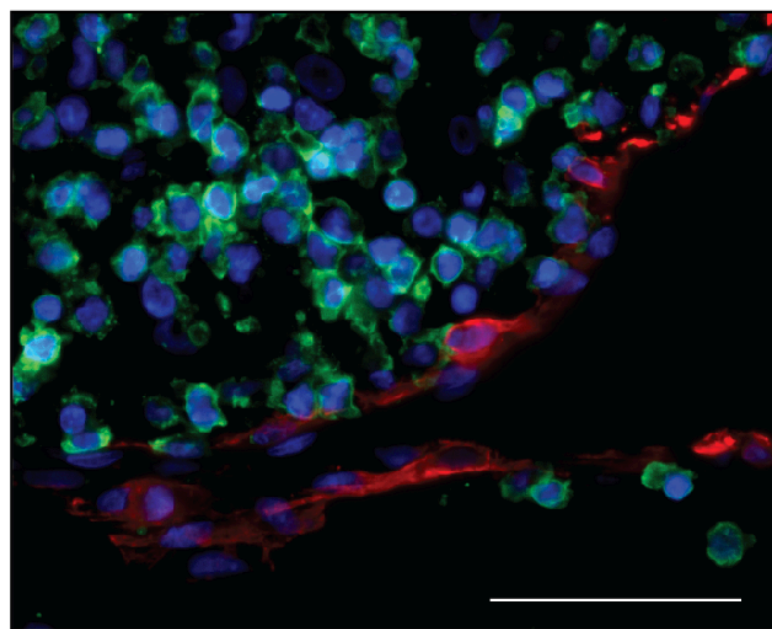
SMA
CD56
DAPI



DAPI
K7
CD56

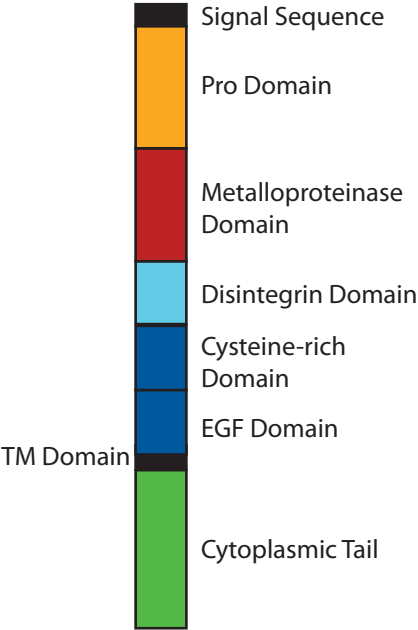


DAPI
CD56
SMA

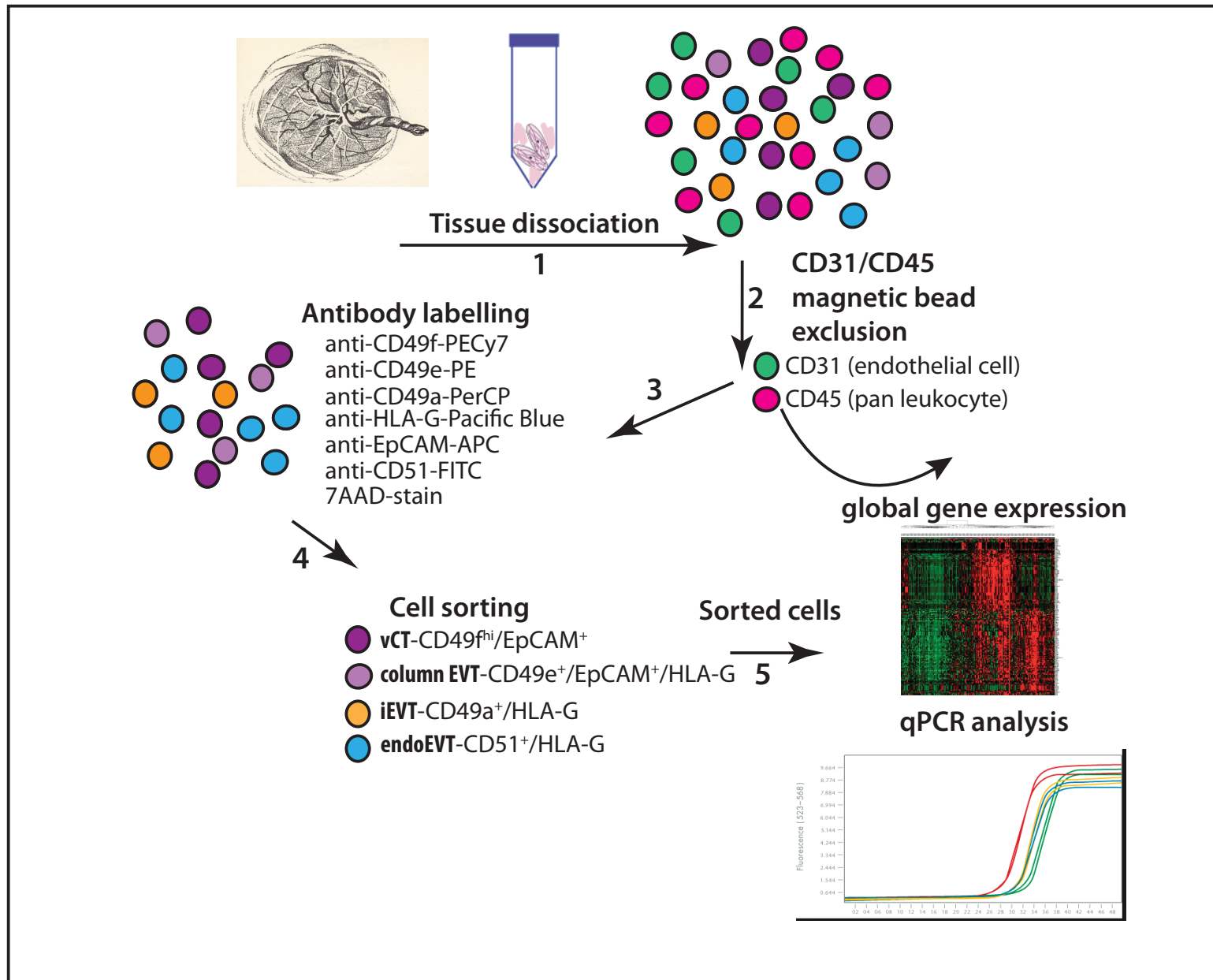


ADAM Family of Metalloproteinases

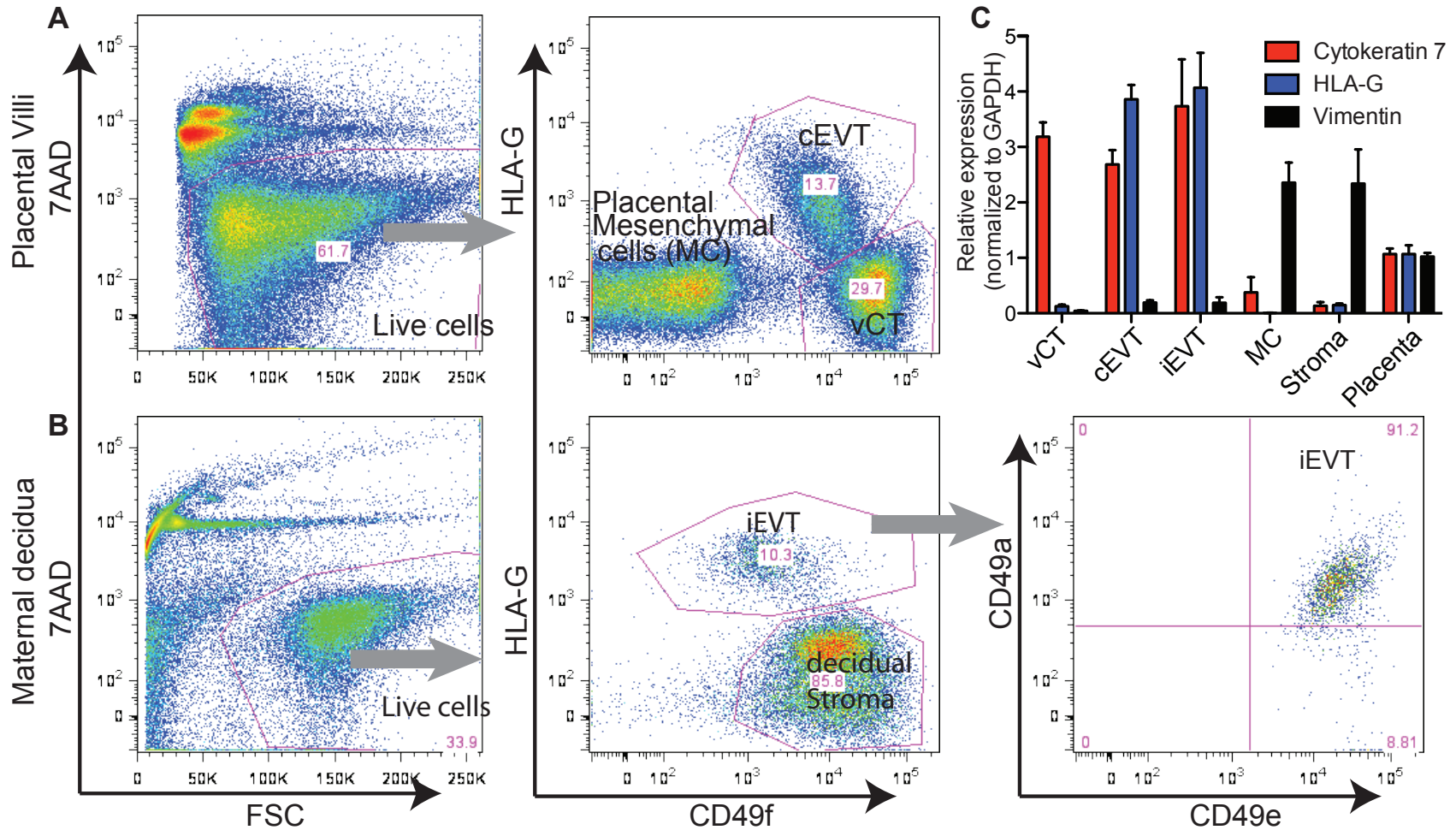
- Major proteinase family related to MMPs and snake venom disintegrins
- 21 human ADAMs have been described; of these only 12 contain conserved metalloproteinase Zn-binding active sites
- Play biological roles in growth factor availability, cell migration, integrin signaling, cell-cell fusion



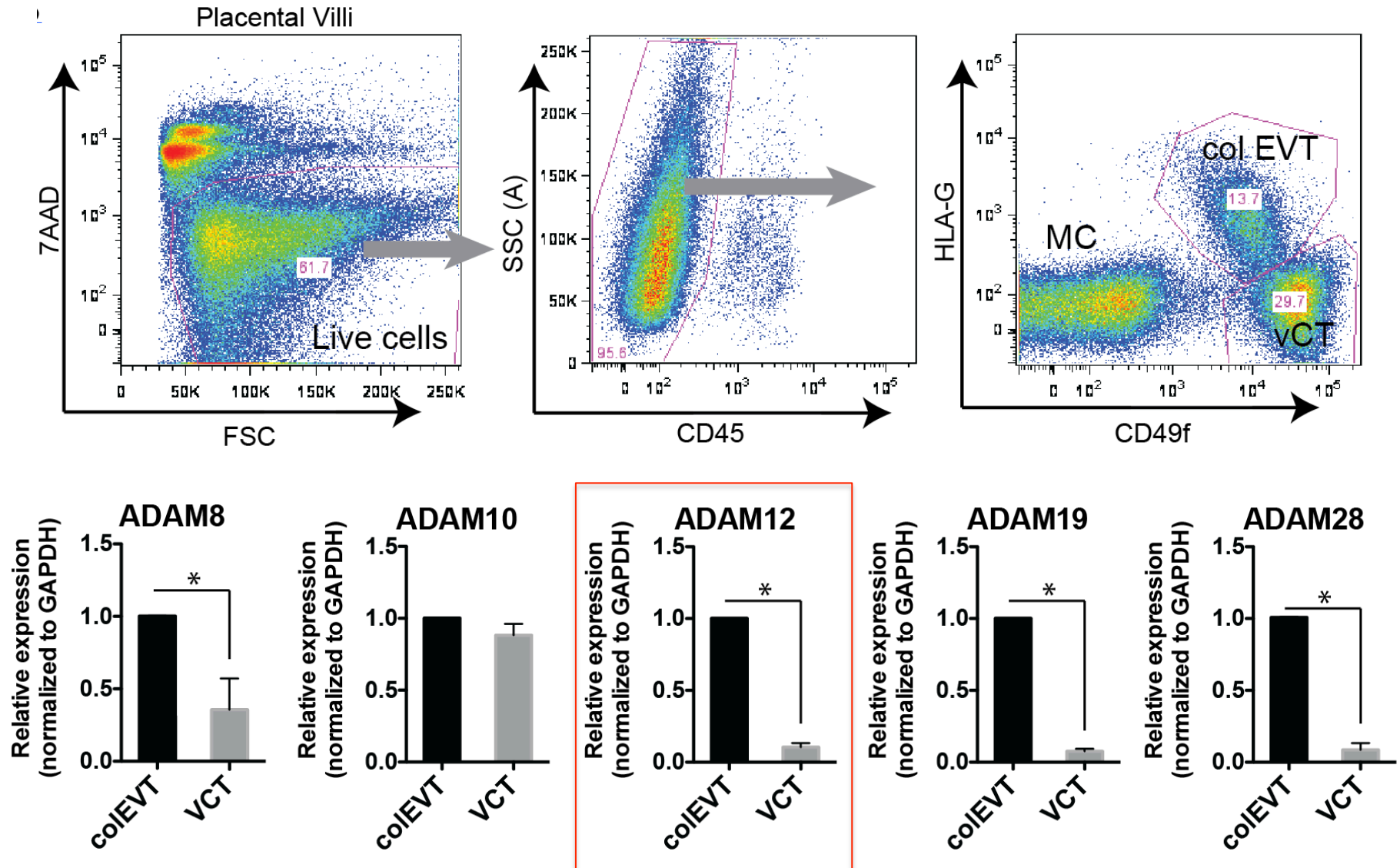
Characterizing trophoblast populations in the developing placenta



Characterizing trophoblast populations in developing placenta

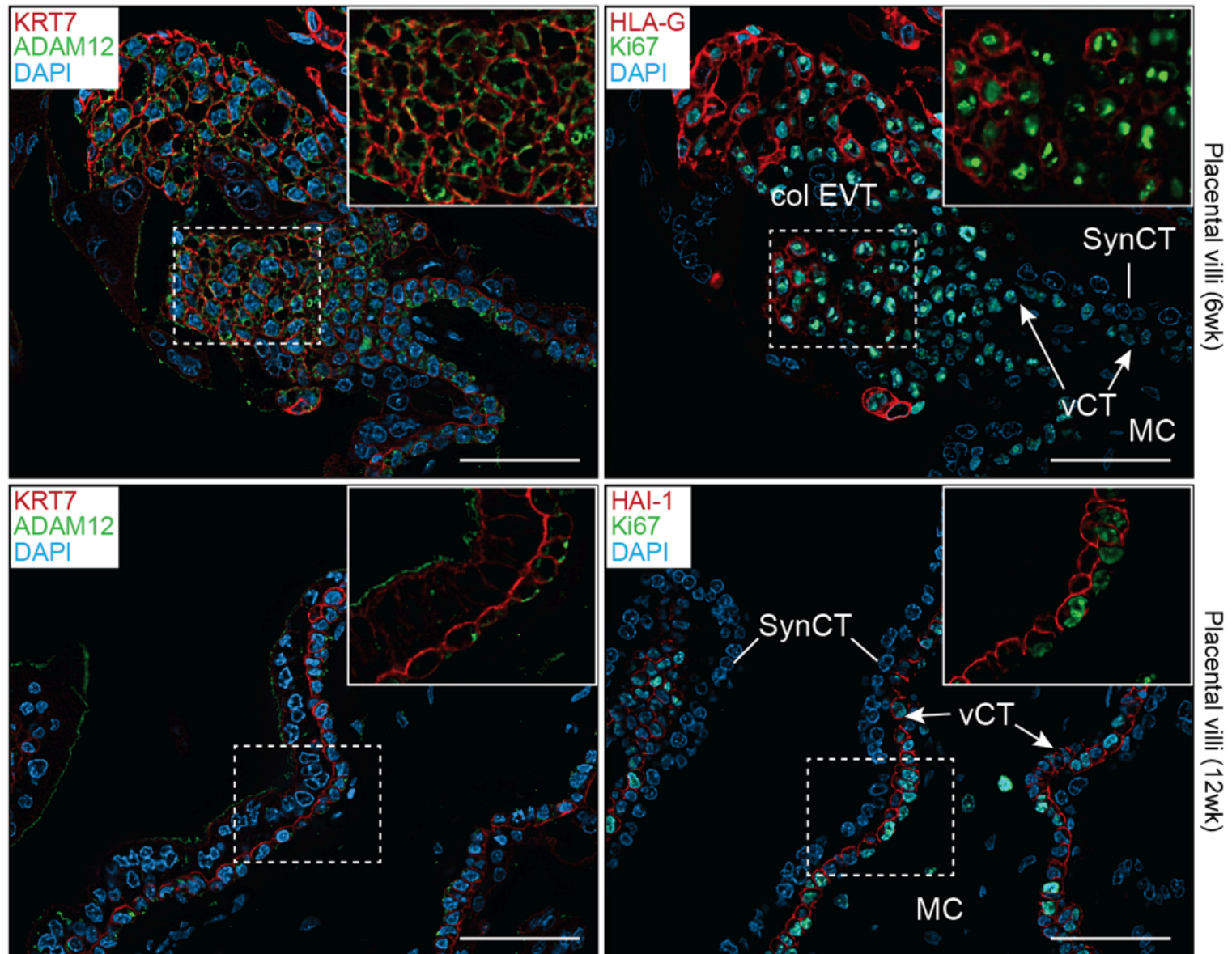


ADAM subtype expression in trophoblasts of placental villi

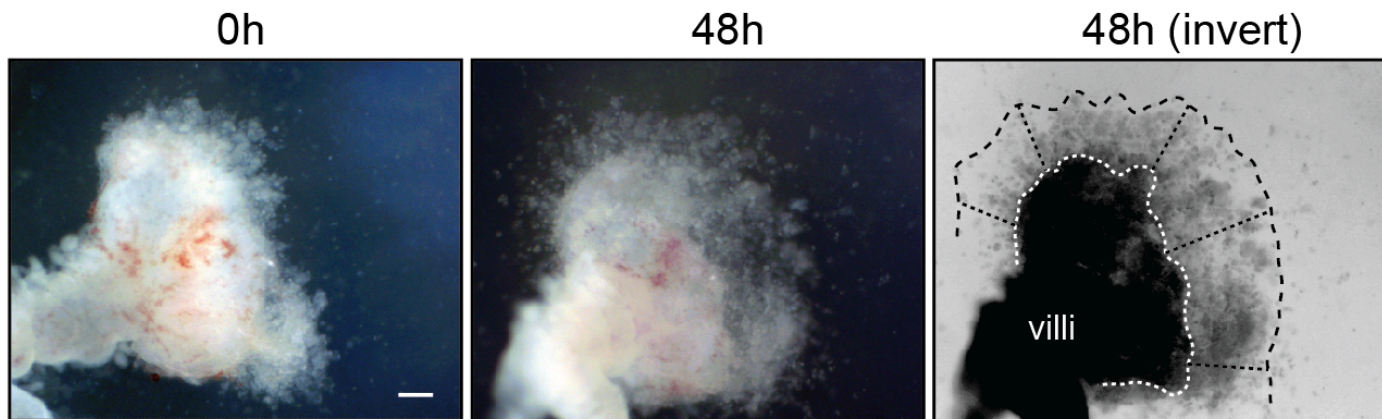
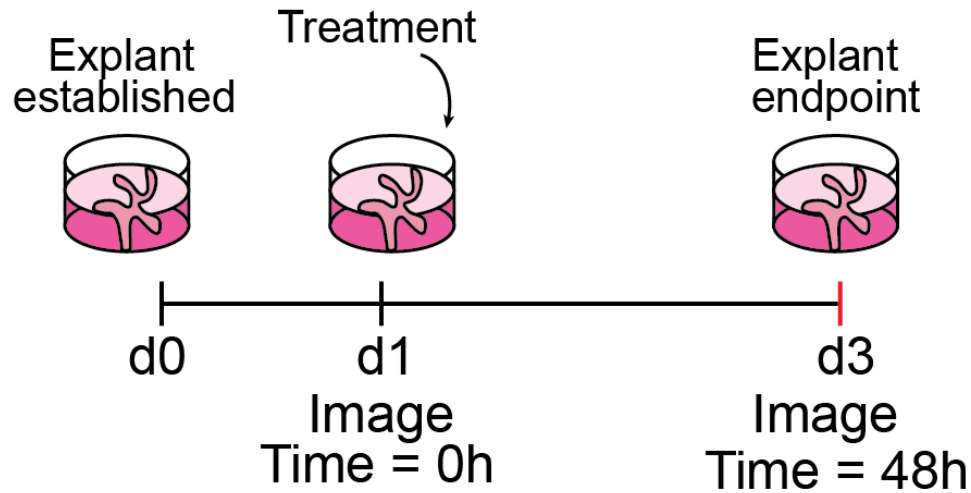


ADAM12 serum levels decreased in PE, IUGR

ADAM12 expression in trophoblasts

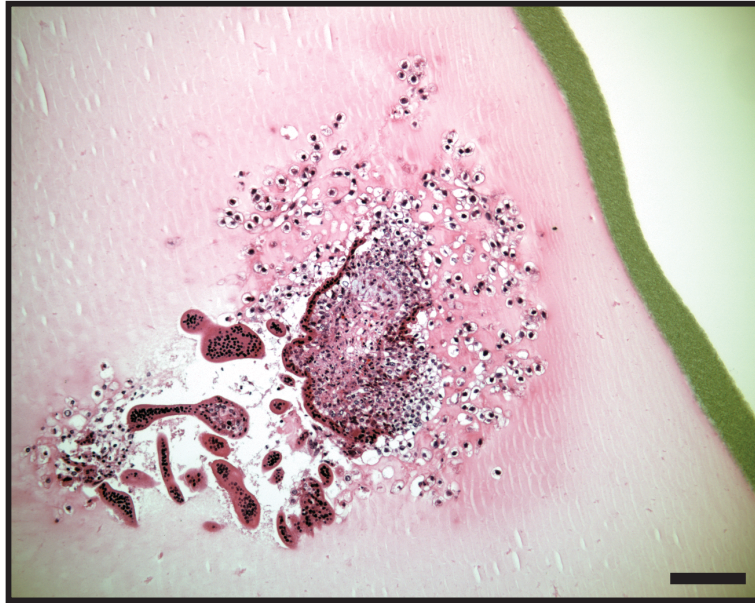


Ex vivo model to study early placenta development

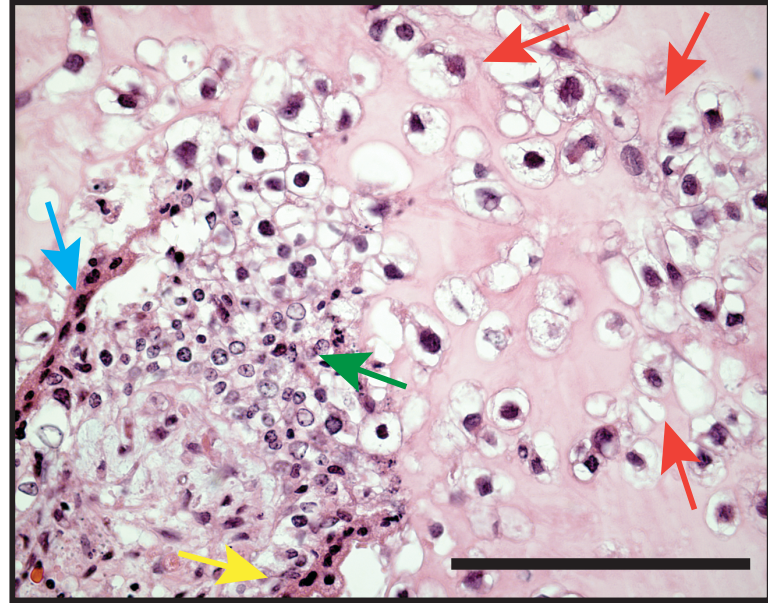


Explant model enables examination of multiple types of trophoblasts

H&E Matrigel Explant (96 h)



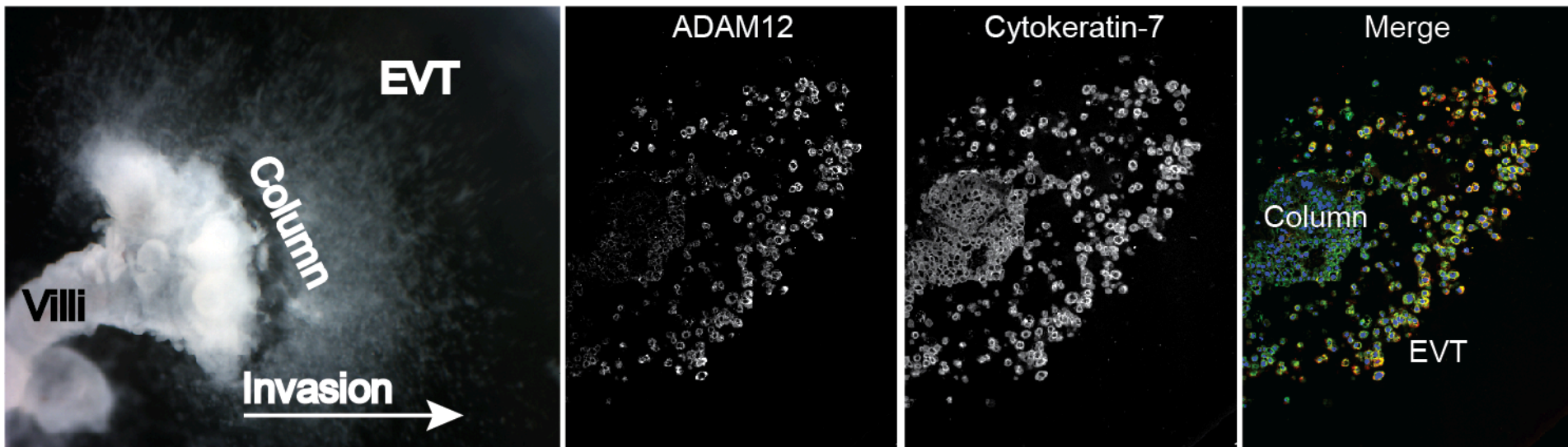
10x objective



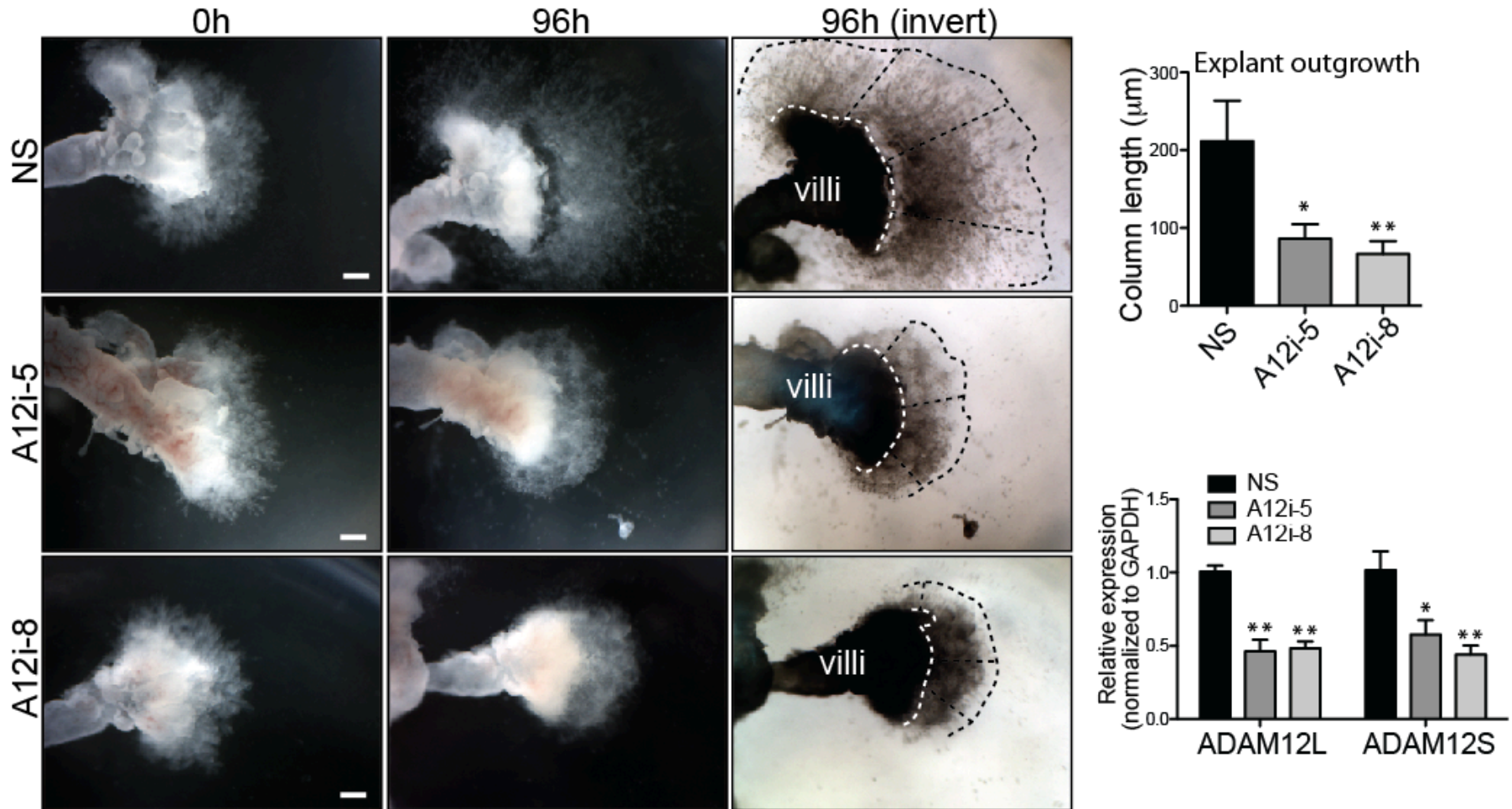
40x objective

- vCT
- synCT
- column EVT
- interstitial EVT

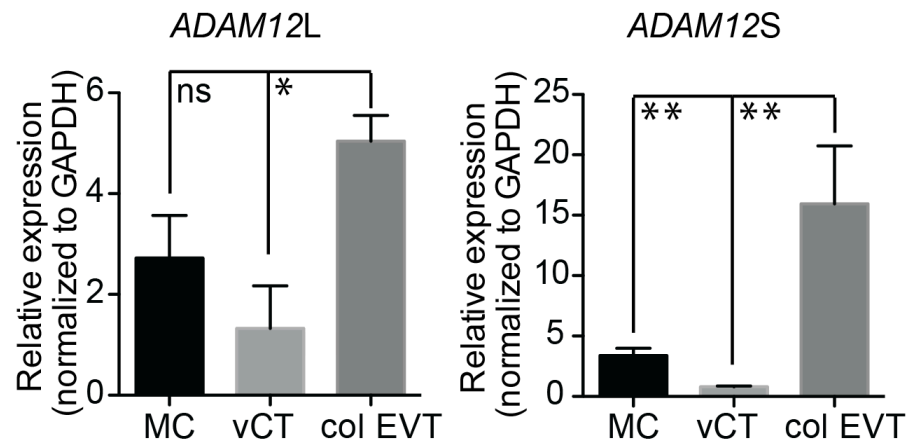
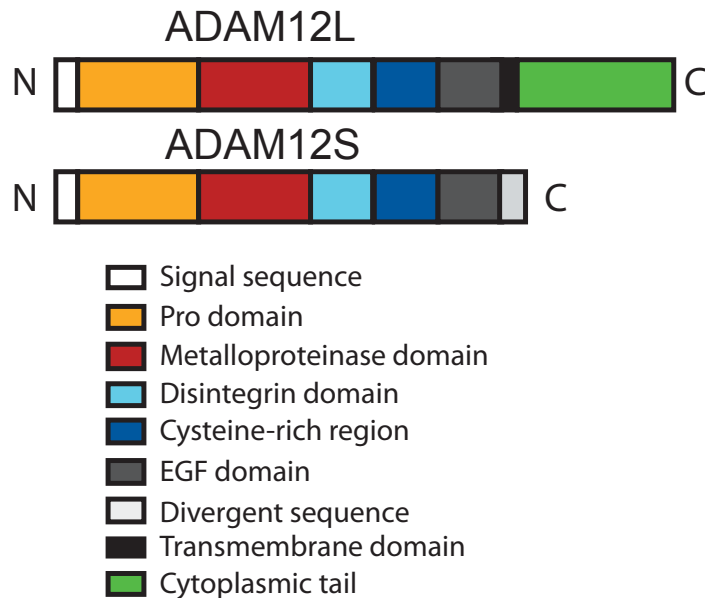
ADAM12 preferentially localizes to invasive trophoblasts



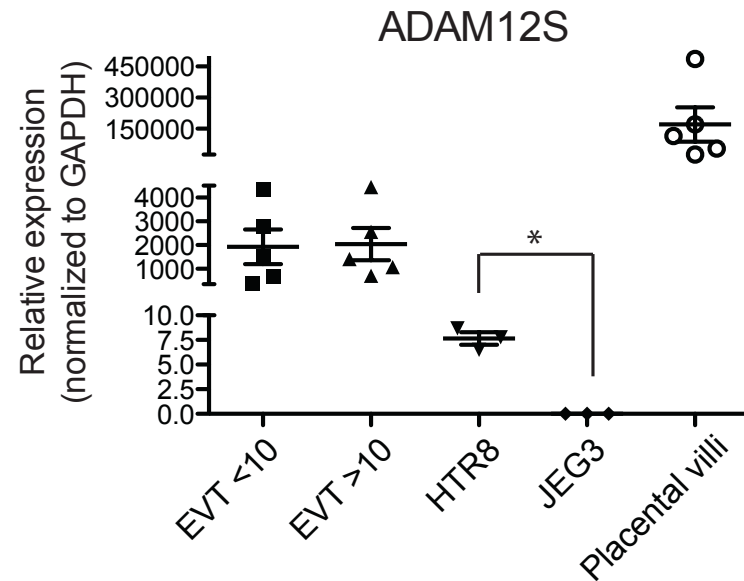
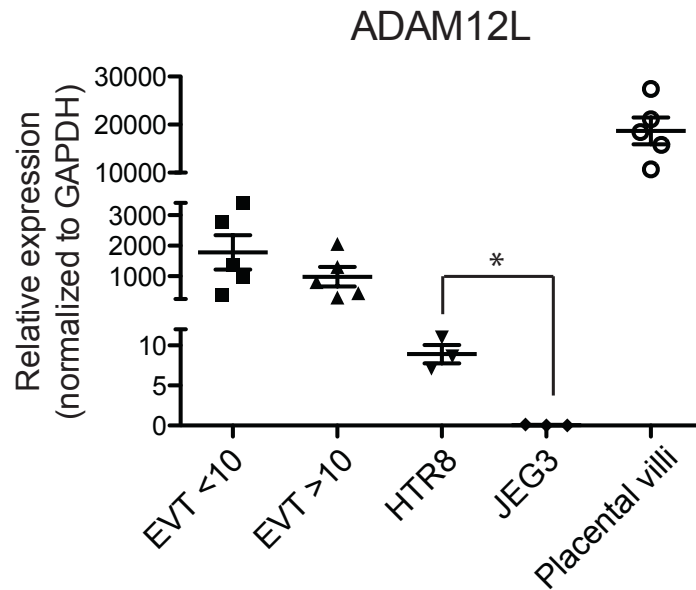
ADAM12 blocks column outgrowth



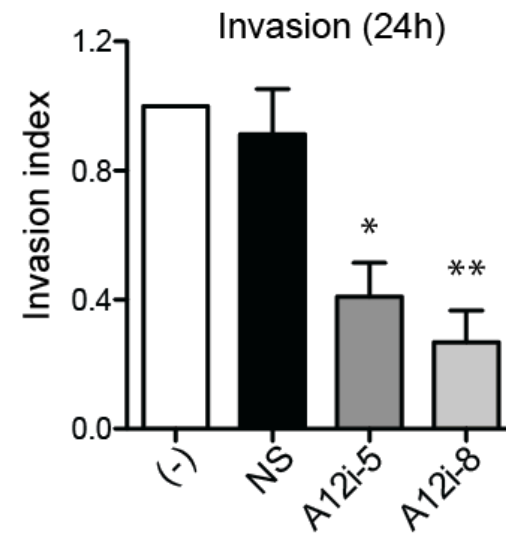
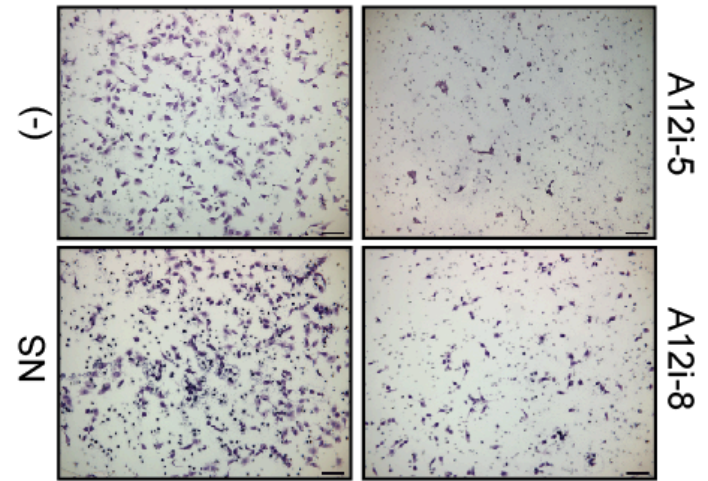
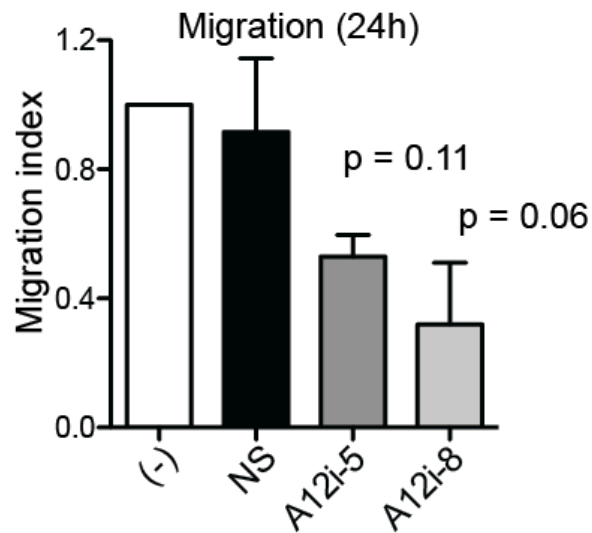
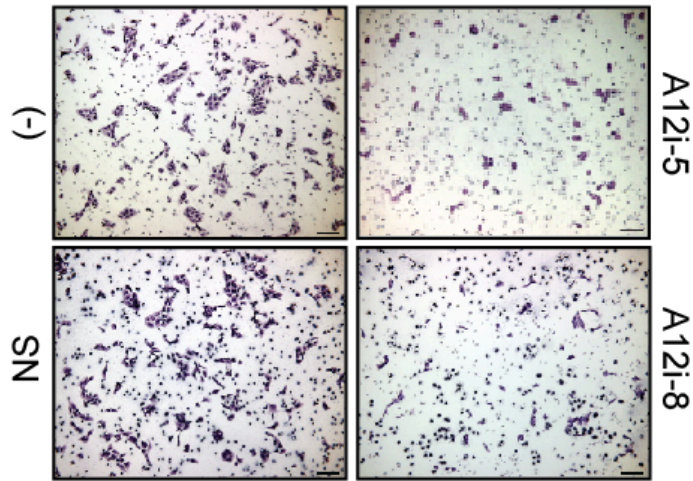
Two splice variants of ADAM12 in humans (only one in mice)



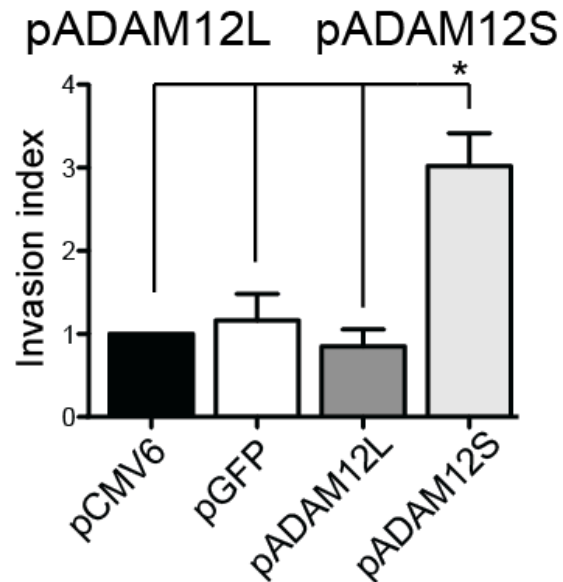
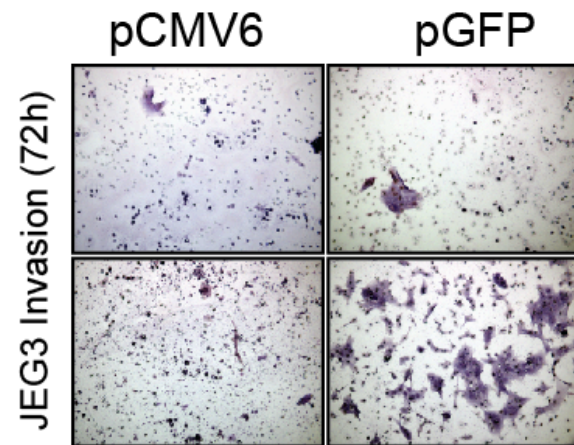
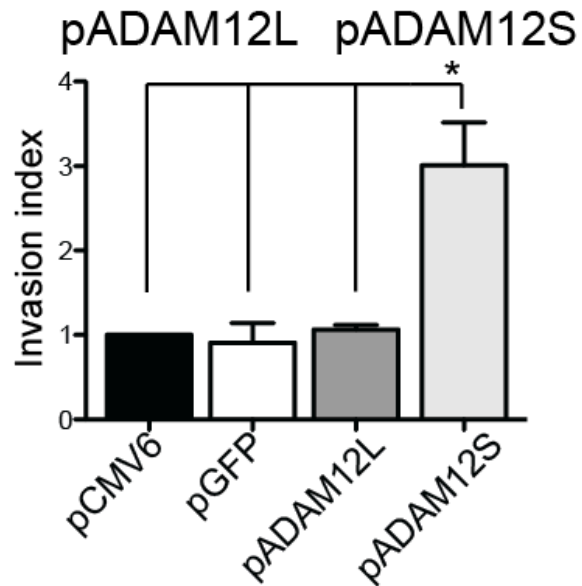
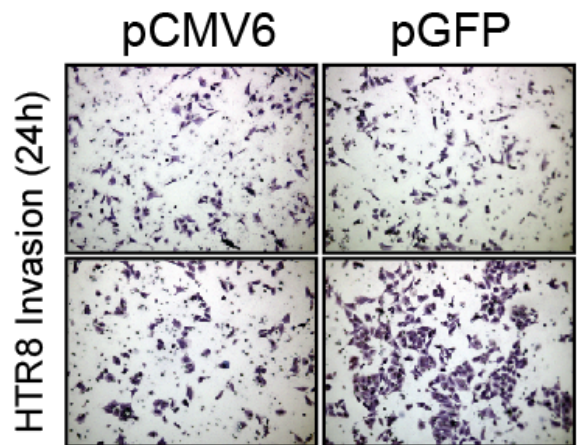
Primary cultures & cell lines



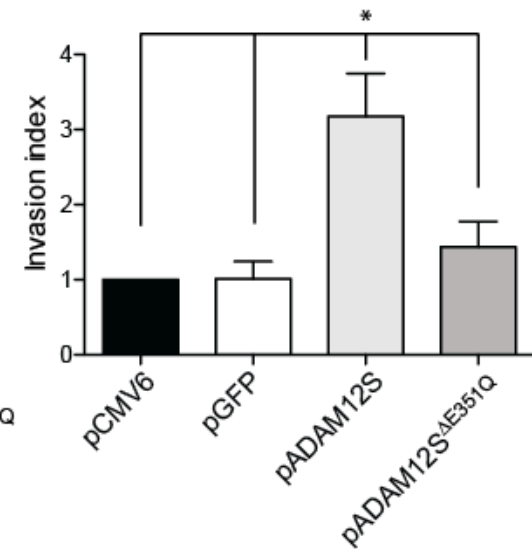
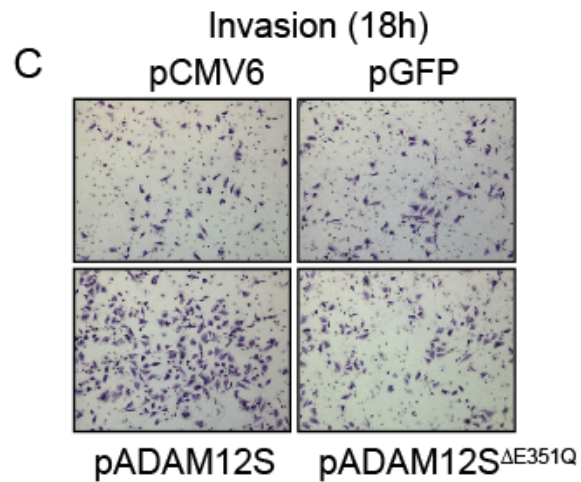
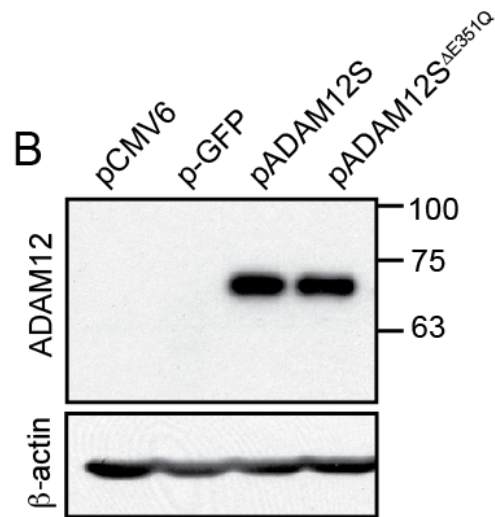
Loss of ADAM12 impairs motility



ADAM12S drives invasion



ADAM12S requires intrinsic proteolytic activity



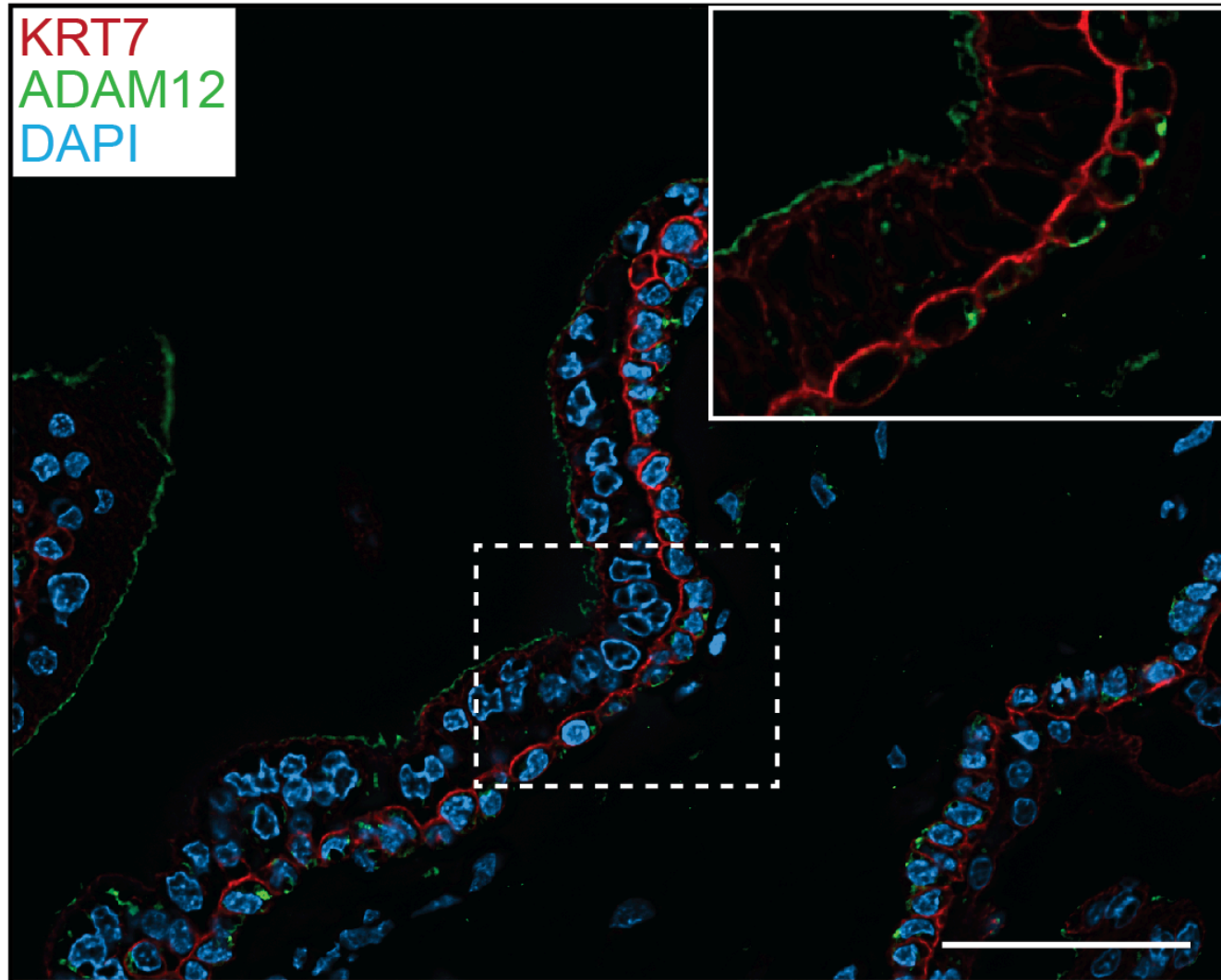
So far...

- ADAM12 localizes to invasive trophoblasts
- ADAM12 controls trophoblast invasion and column outgrowth

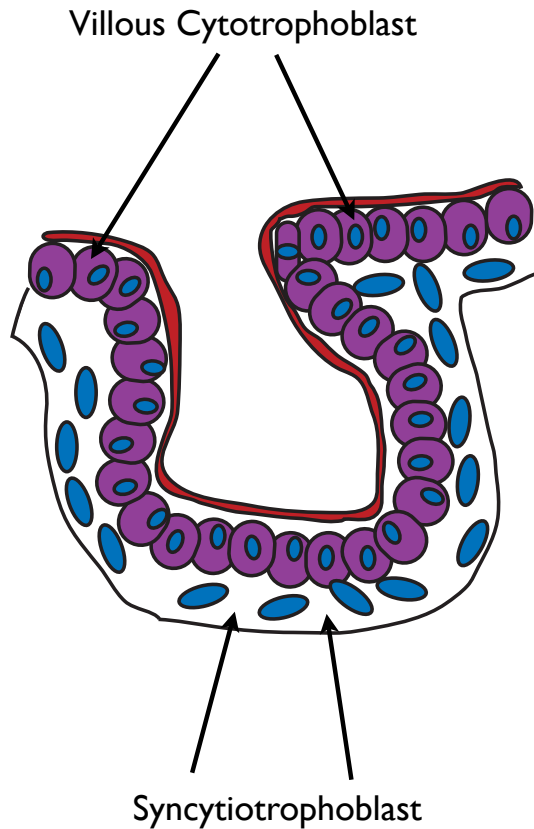
But...

-ADAM12 also localizes to other trophoblast populations

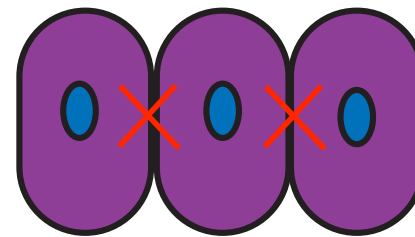
ADAM12 localizes sporadically to progenitor cells and apical layer



In vitro syncytialization (cell-cell fusion)

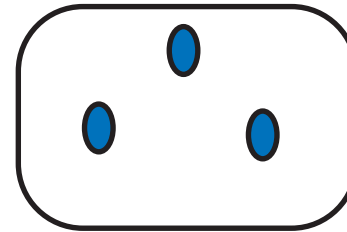


Trophoblast cell-cell fusion

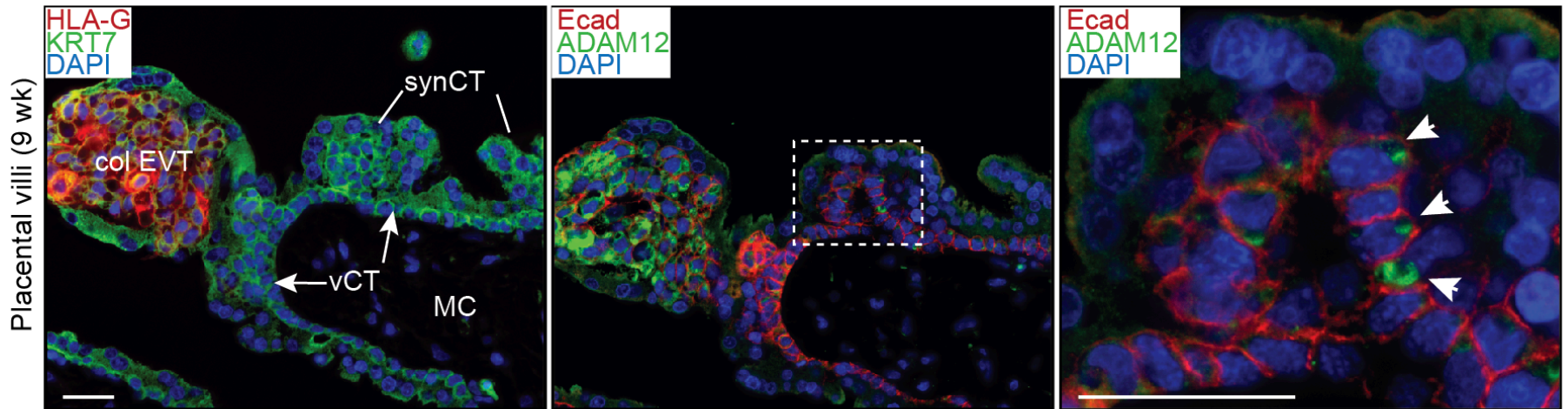


PKA
Gcm1
Cx43

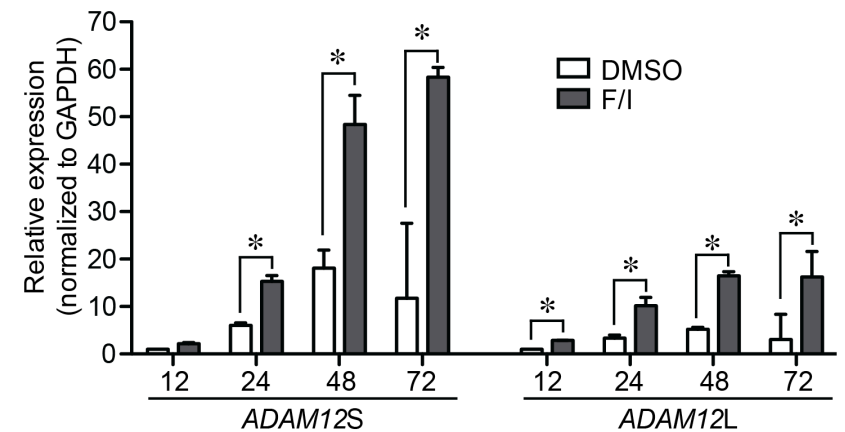
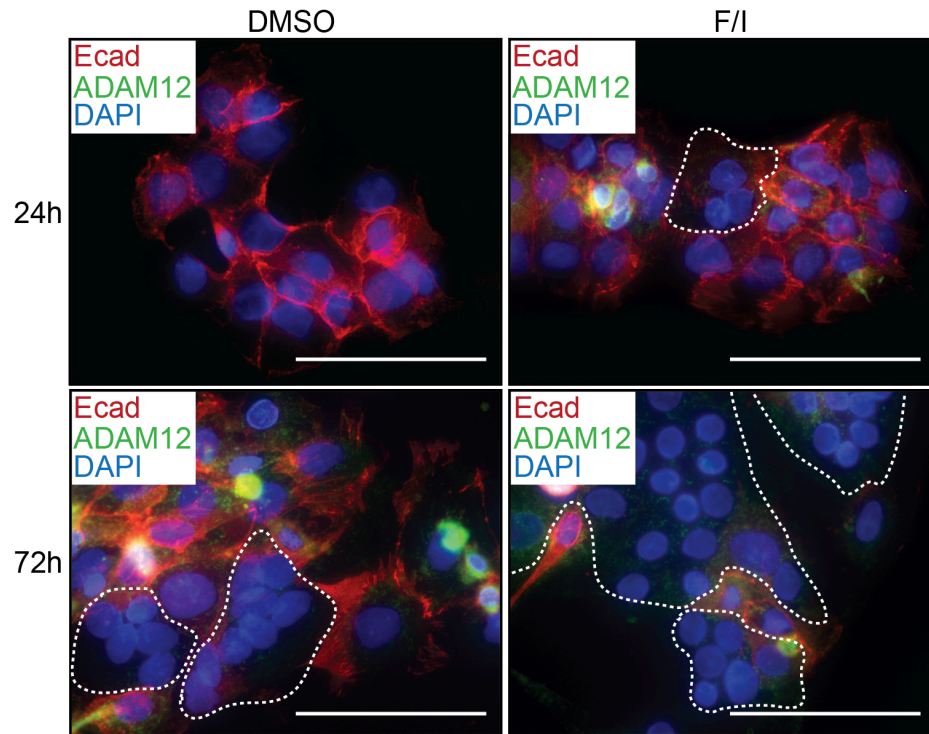
loss of intercellular adhesive structures
increase in fusogenic proteins



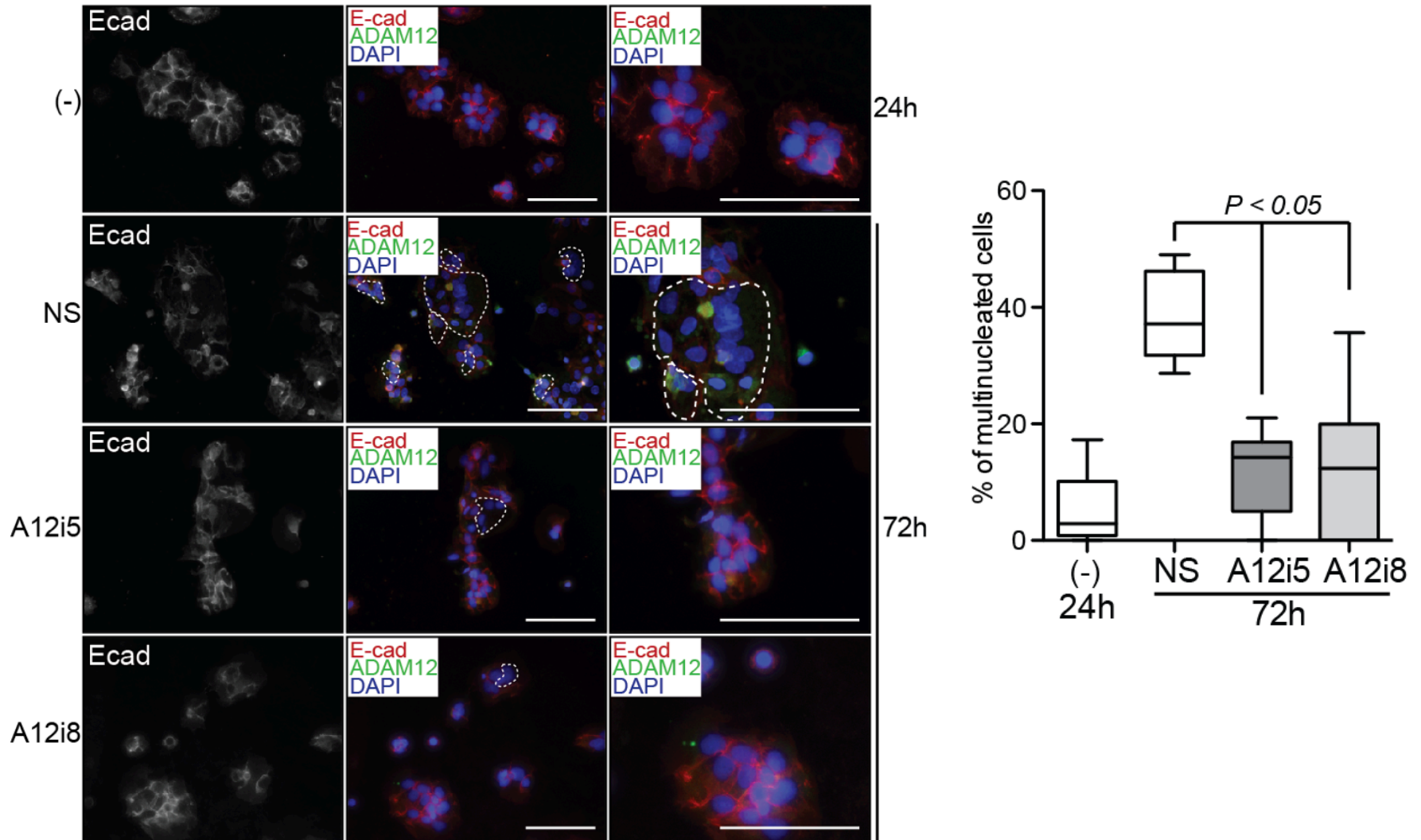
ADAM12 localizes to fusing trophoblast *in vivo*



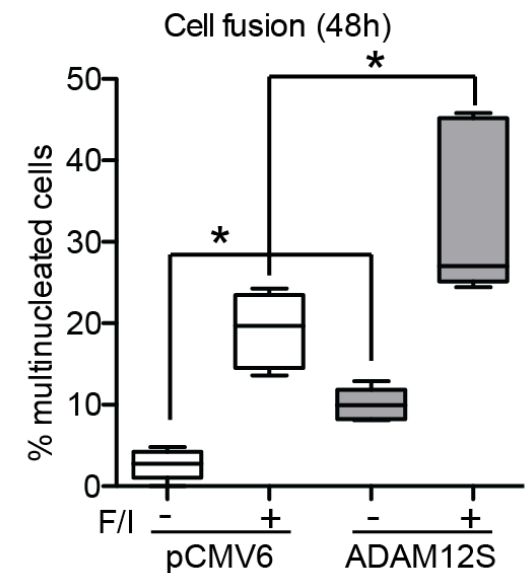
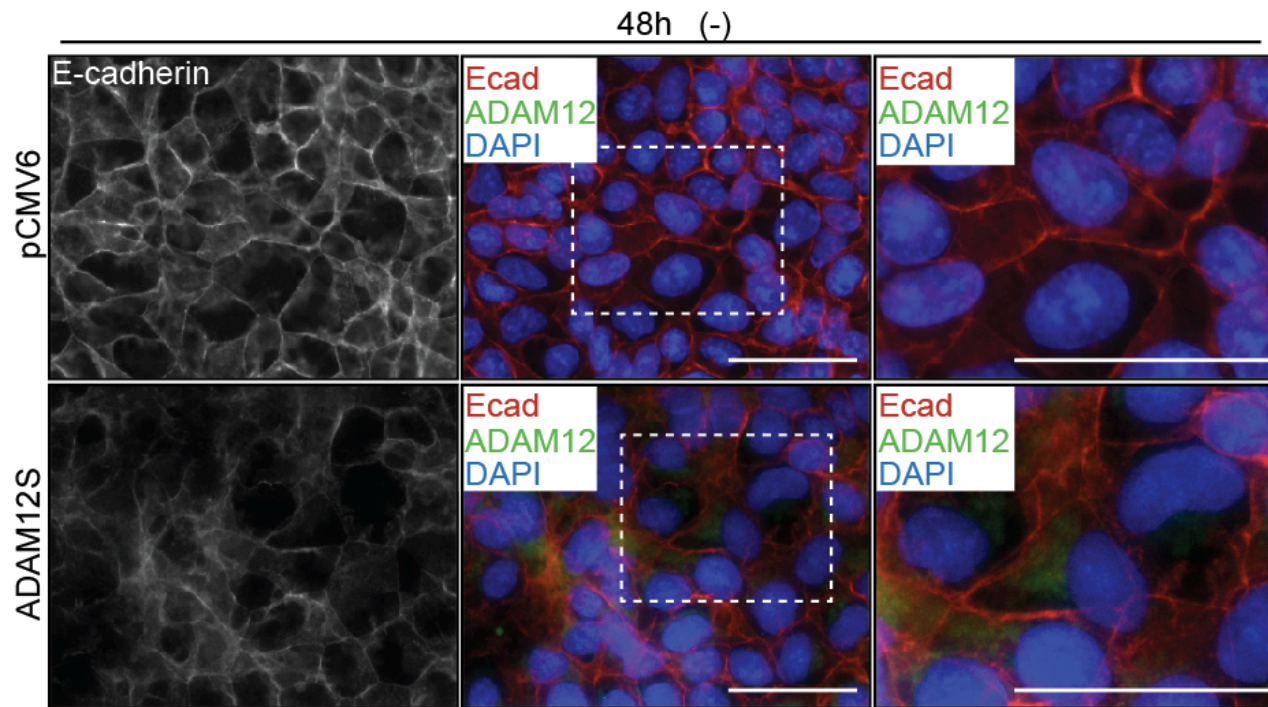
ADAM12 levels increase during cell fusion



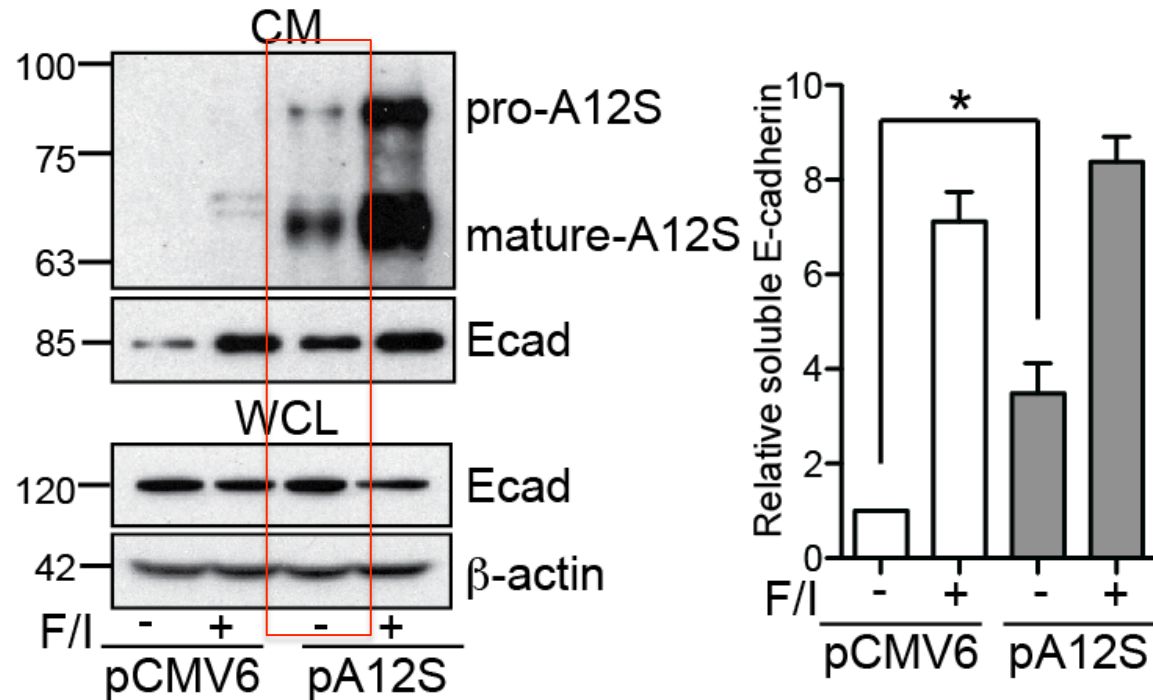
Loss of ADAM12 impairs cell fusion



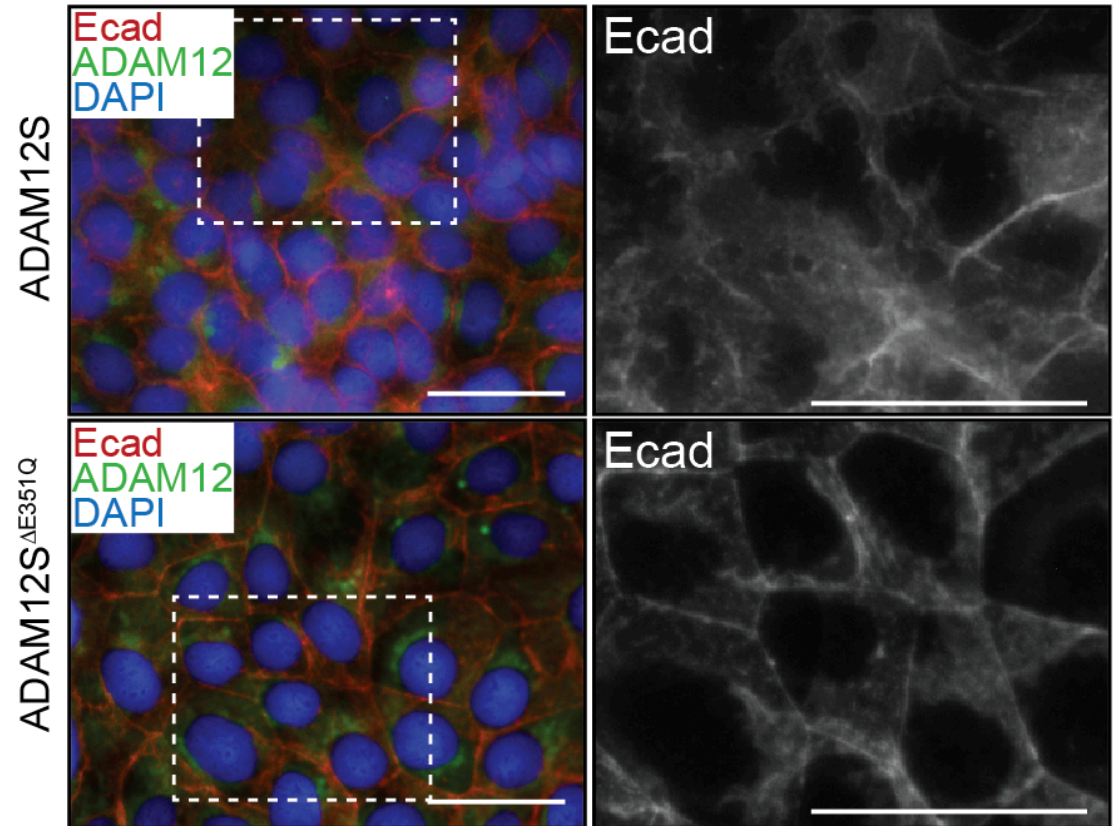
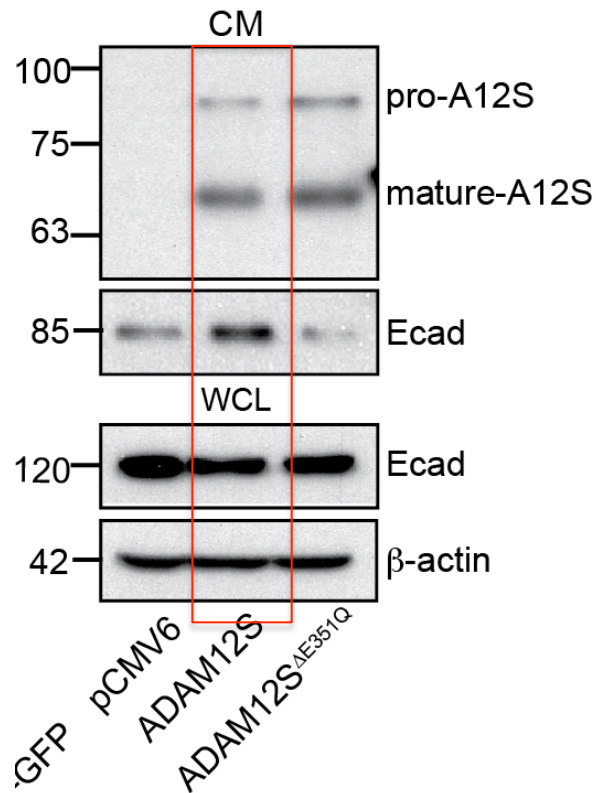
Ectopic ADAM12S potentiates fusion



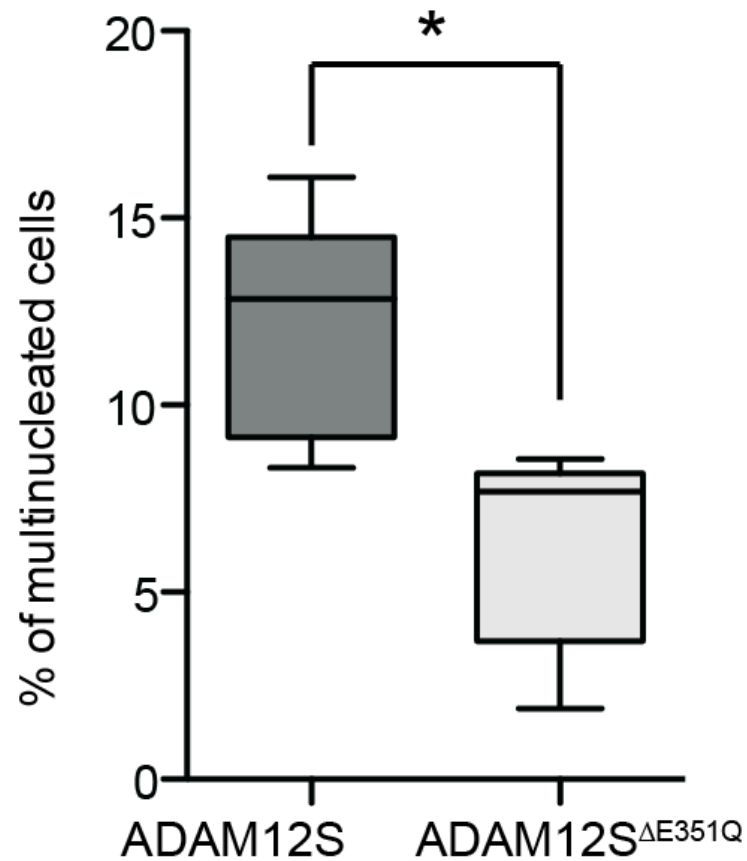
Ectopic ADAM12 generates soluble E-cadherin



Loss of ADAM12 proteolytic function prevents E-cadherin shedding



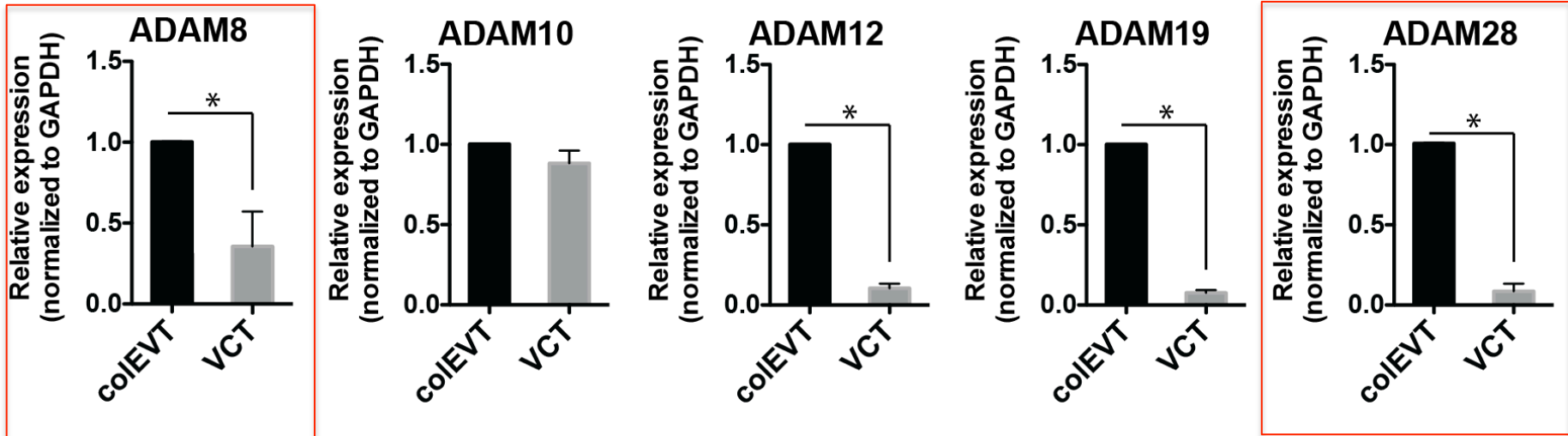
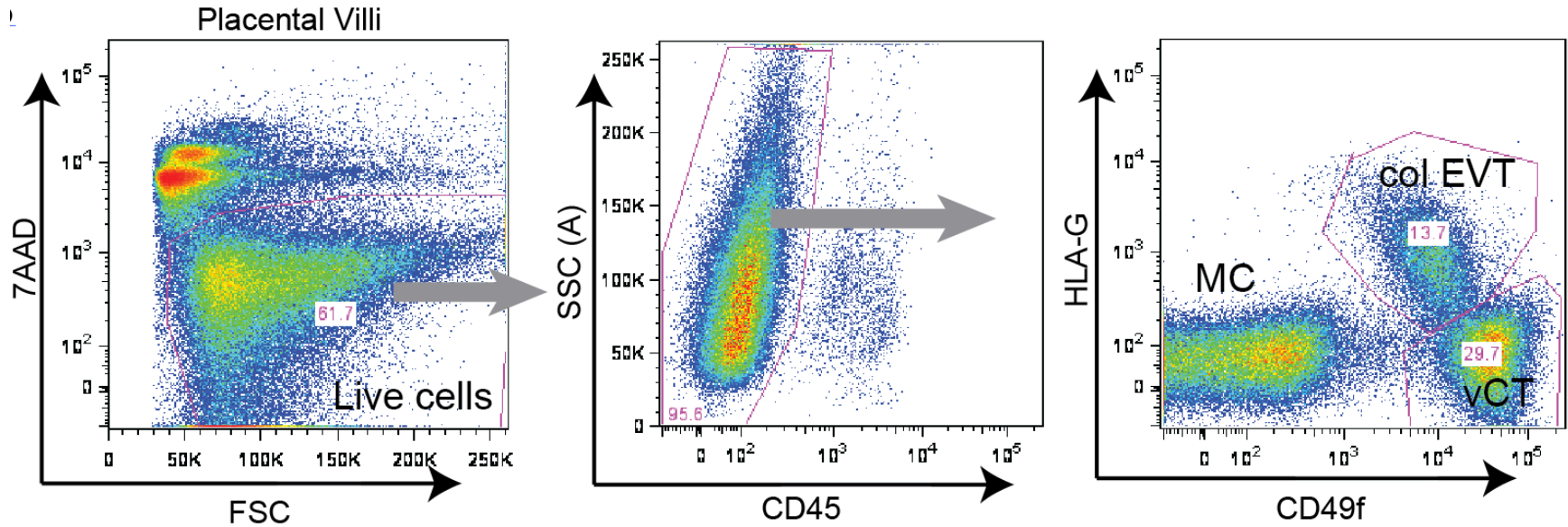
Catalytic inactivation of ADAM12 impairs trophoblast fusion



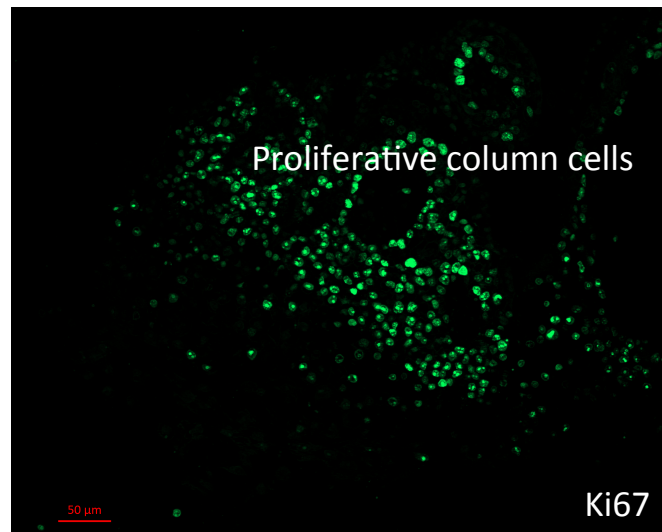
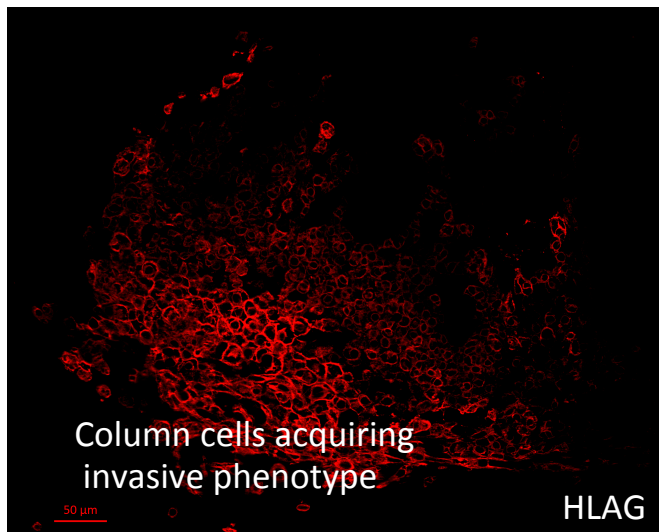
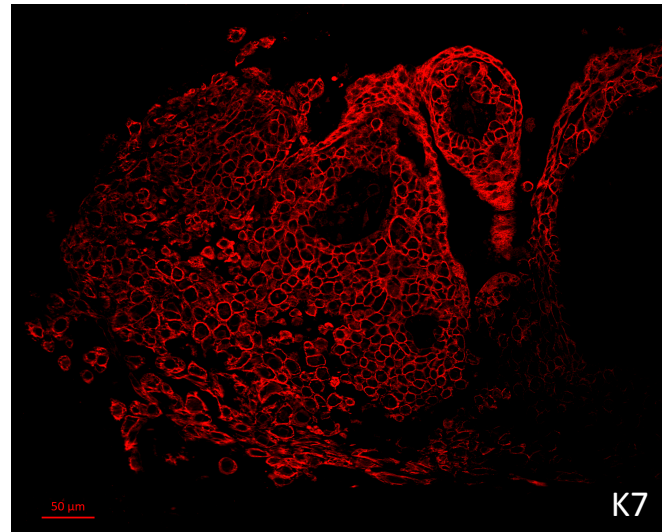
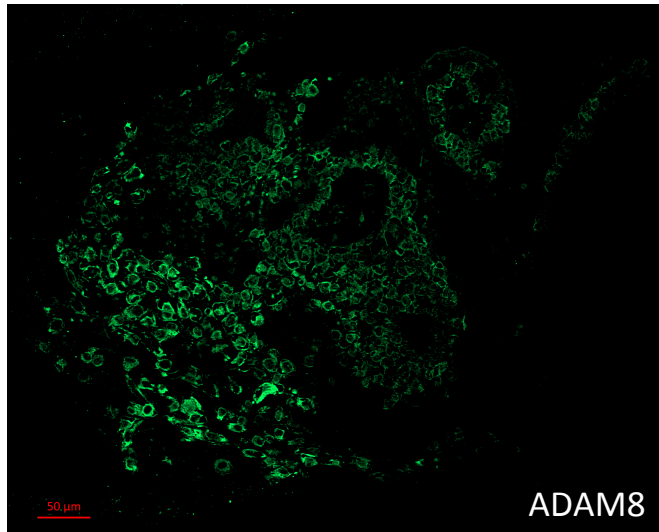
Summary

- ADAM12 is expressed in trophoblast populations of the human placenta
- ADAM12 localizes specifically to invasive EVT subsets and to fusing cytotrophoblasts
- ADAM12S promotes trophoblast column outgrowth (motility?)
- ADAM12S cleaves the ectodomain of E-cadherin; potentiates cell fusion

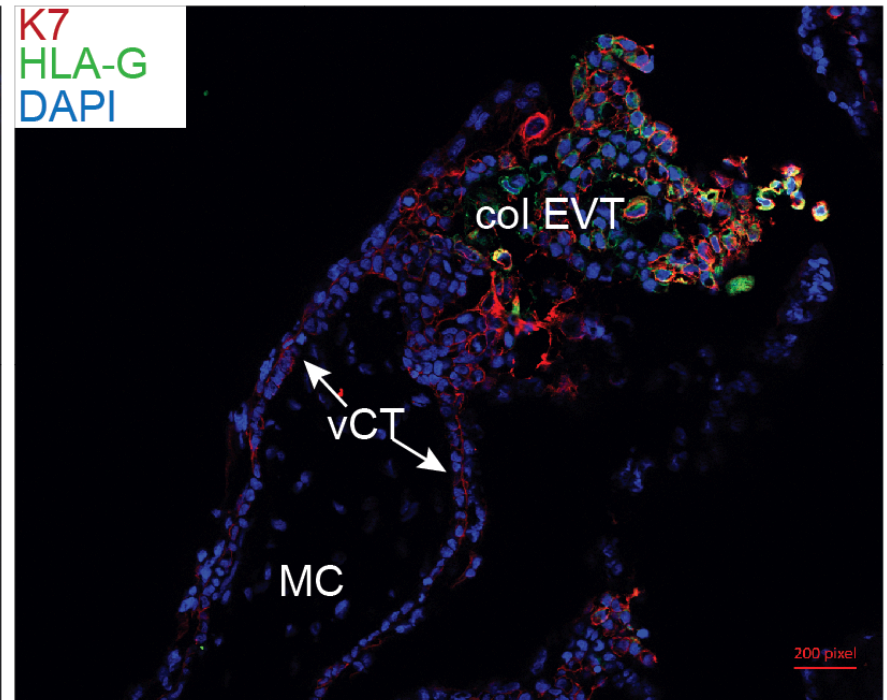
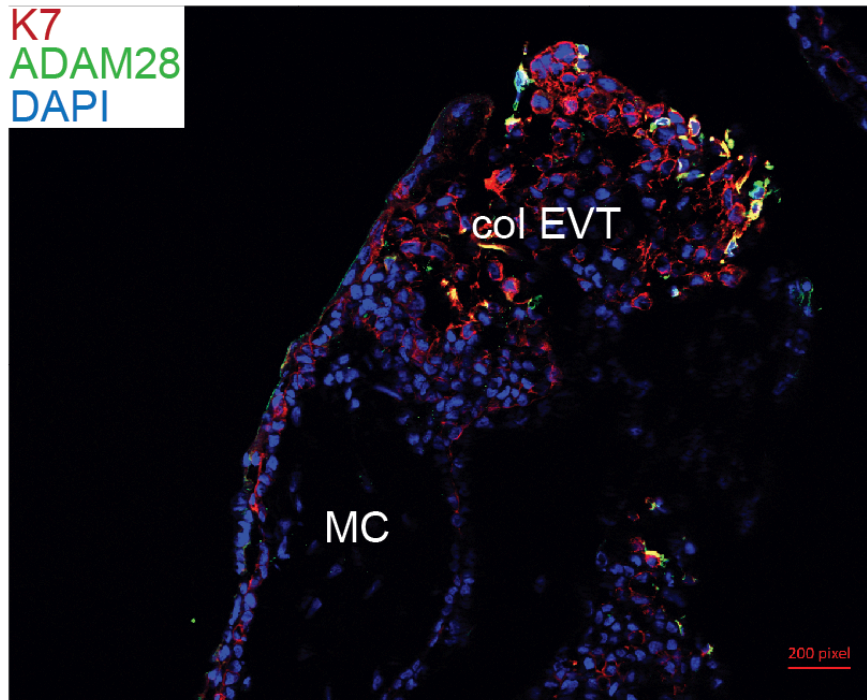
ADAM subtype expression in trophoblast of placental villi



ADAM8 localization in placental explant



ADAM28 localization within first trimester placental villi



Acknowledgements

BC Women's Hospital's CARE Program

Sofie Perdu - Research technician

Yoon Kim – Graduate student

Lauren De Luca – Graduate student

Kathy Chan – CFRI summer student

Dr. Barbara Castellana – Postdoc

Dr. Hoa Le – Research associate

Mahroo Aghababaei – graduated!



a place of mind

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