



GENDER-SPECIFIC PLACENTAL EXPRESSION OF CANNABINOID RECEPTOR ONE (CB1R) IN BABOON MODEL OF OBESITY

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Introduction

Maternal obesity is associated with offspring behavioral and cardiovascular developmental problems through poorly understood mechanisms. Endocannabinoids (eCB) are lipid derivatives, linking metabolism to mental and cardiovascular health (Figure 1). CB1R – a G-protein-coupled receptor- is the main target of endogenous (2-arachidonoylglycerol (2-AG) and N-arachidonylethanolamine (AEA)) and exogenous cannabinoids (e.g. marijuana). We previously reported a seemingly paradoxical decrease of CB1R ligands' concentrations (Figure 2), decreased mRNA, but not CB1R protein expression in placentas of obese baboons (increased food consumption model) (Placenta 2013 34(11):983) (Figure 3) and decreased placental protein expression of CB1R and CB2R in obese pregnant women carrying male fetuses (Figure 4.) (AJOG, 2014, 210(1):S94).

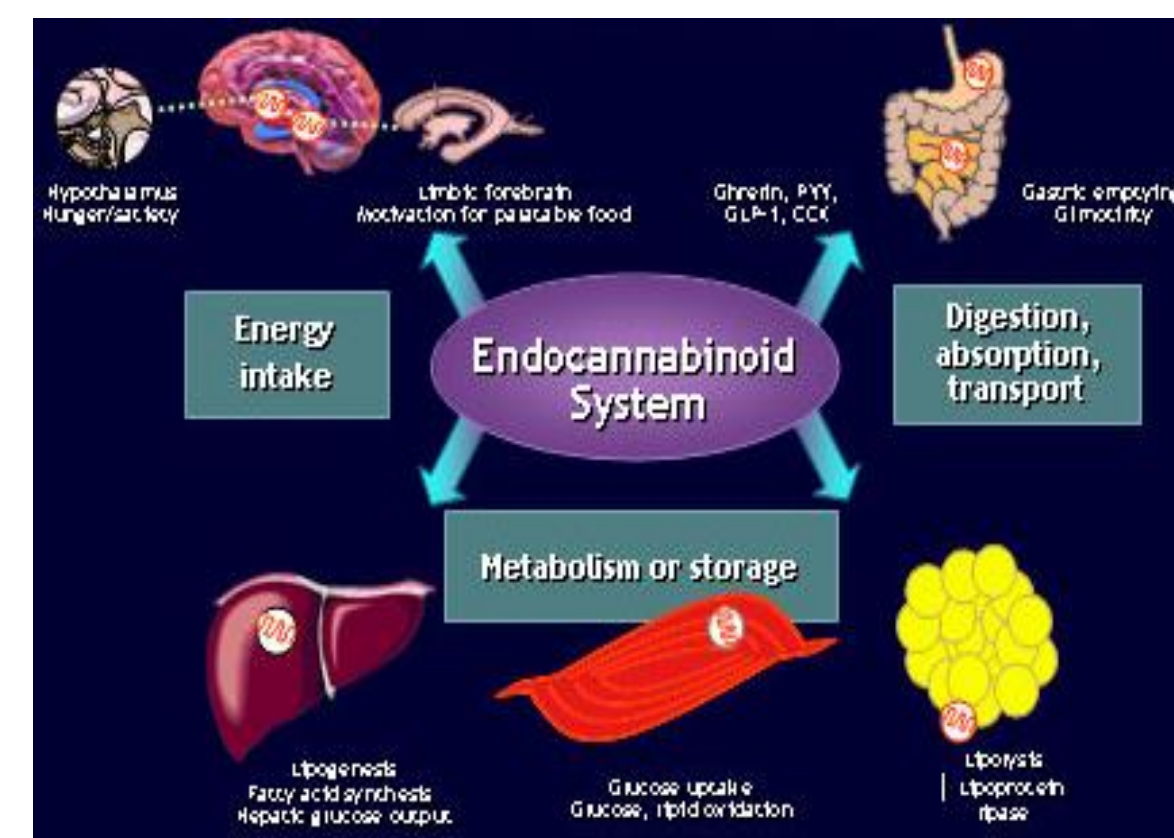


Figure 1. Endocannabinoid system and the effects on various systems of the body. (F. Xavier Pi-Sunyer, 2008)

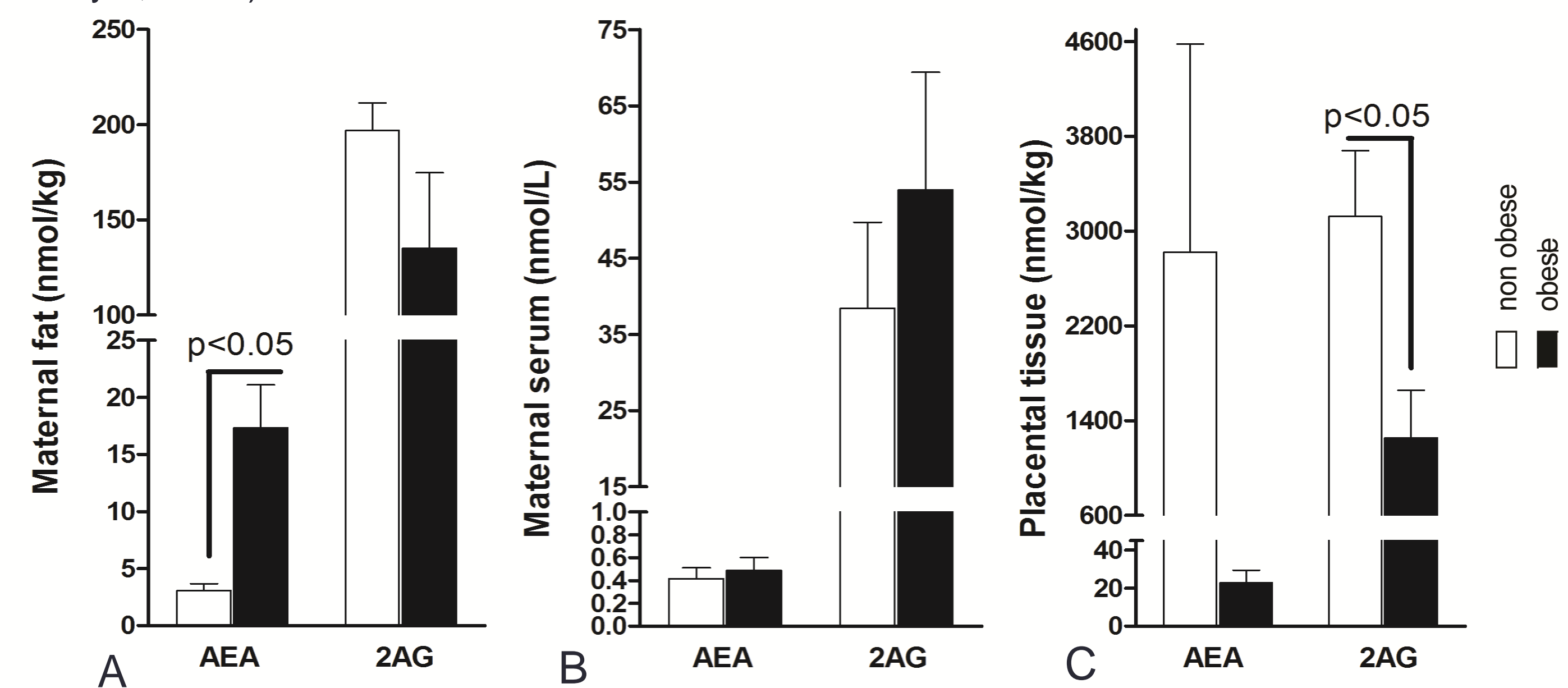


Figure 2. Concentrations of AEA and 2AG in placenta, maternal serum and maternal fat of obese (n=4) and non-obese (n=4) pregnant baboons (Placenta 2013 34(11):983).

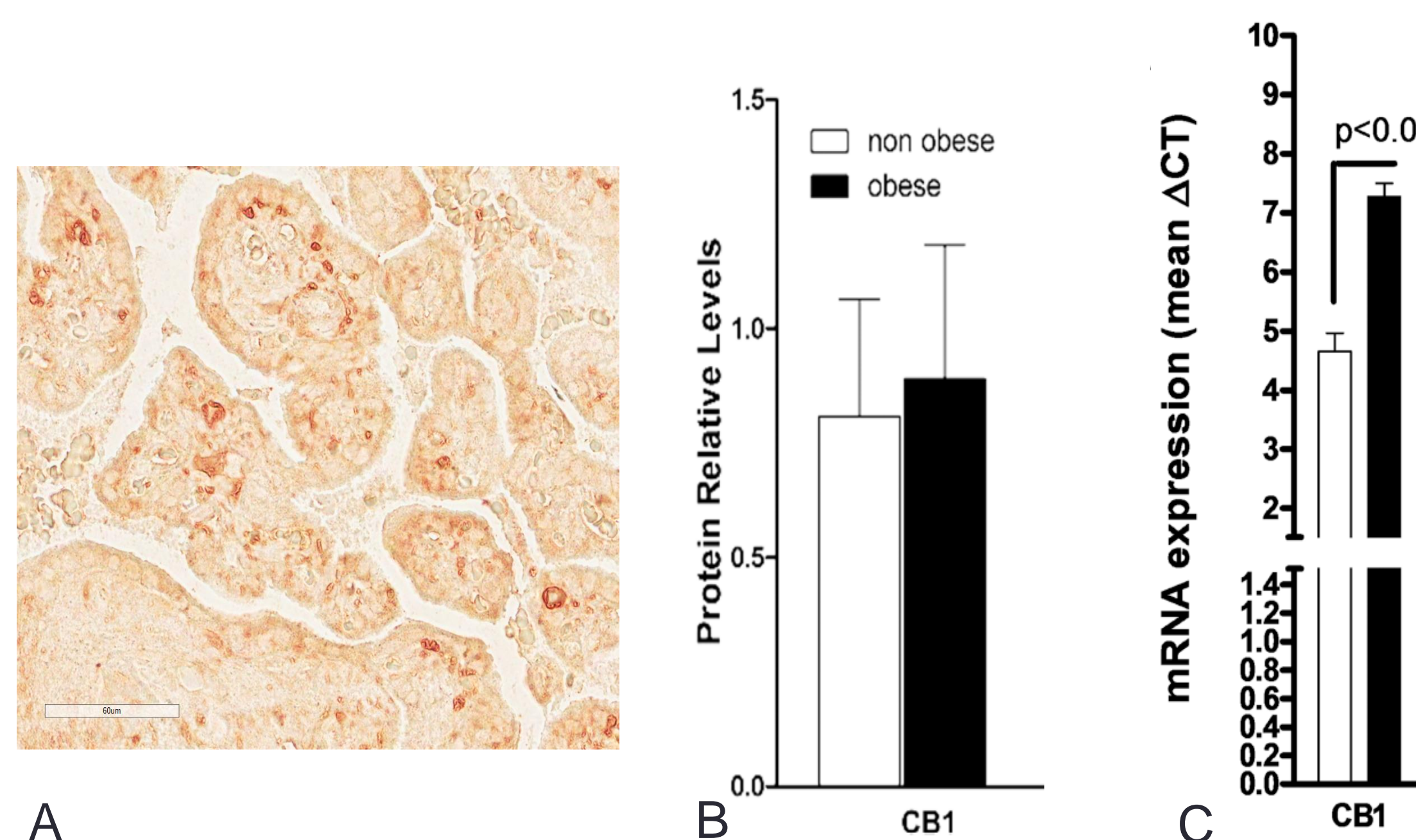


Figure 3. CB1R protein expression and quantifications (A,B) and mRNA expression (C) in placenta of naturally obese (n=4) and non-obese (n=4) baboons. (Placenta 2013 34(11):983).

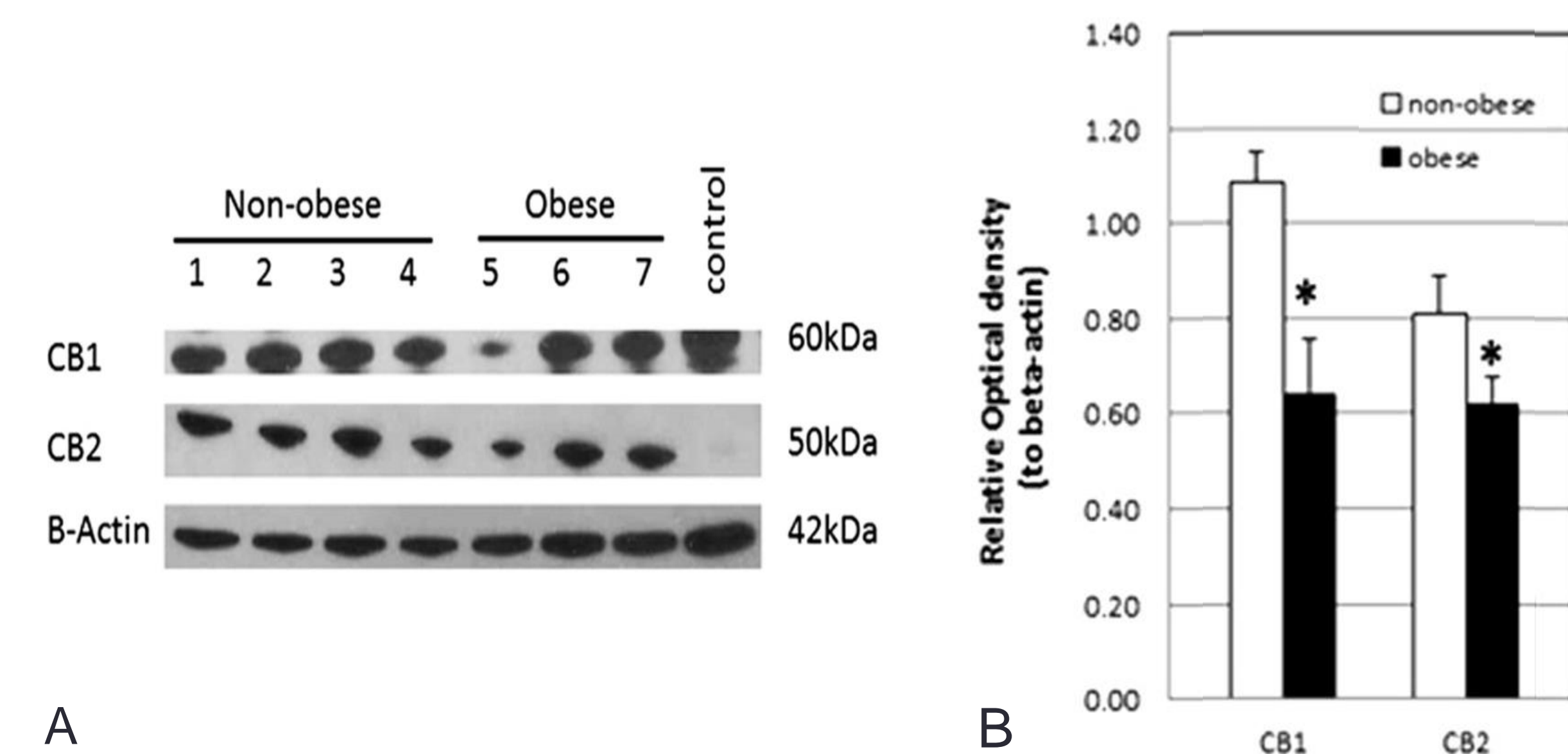


Figure 4. Placental protein expression of CB1R and CB2R in obese (n=3) and non-obese (n=4) pregnant women carrying male fetuses (A). Quantification and analysis by western blot (B) (AJOG, 2014, 210(1):S94).

Objectives

To explore the role of dietary fat composition (higher fat/higher carbohydrate diet) vs. overeating pattern in placental CB1R expression.

Methods

Baboons (*Papio spp*) were fed a diet of 45% fat (HFD; n=4) while controls (CTR; n=4) ate 12% fat from at least 9 months prior to conception through pregnancy until 0.9 gestation (Placenta 2009 30(9):760). Immunohistochemistry (IHC) was performed on placental tissue of male fetuses from mothers of HFD and control groups (Figure 5,6) using the CB1R monoclonal primary antibody (Immunogenes; Budakeszi, Hungary), the secondary antibody is included in the Vectastain ABC kit (Vector laboratories; Burlingame, CA); methodology was performed according to manufacturers instructions. The slides were scanned using the NanoZoomer SQ (Hamamatsu; Middlesex, NJ), quantification was performed using ImageScope™ v11.1.2.752 by Aperio (Leica Biosystems; Buffalo Grove, IL).

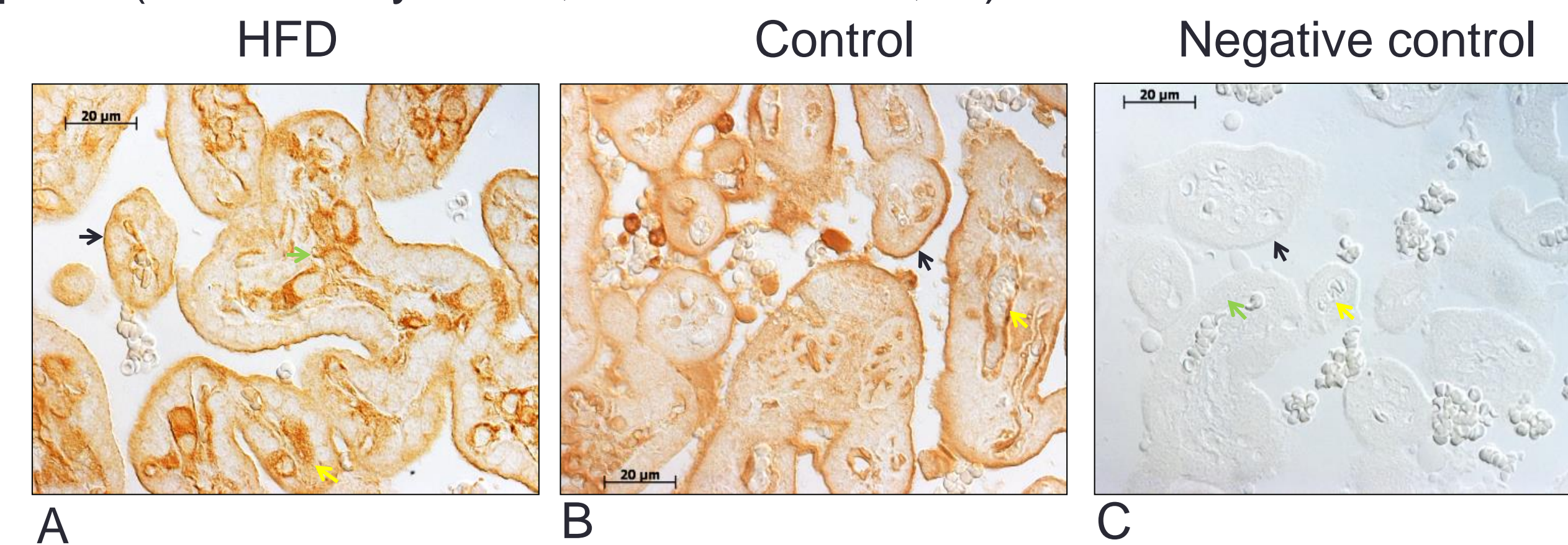


Figure 5. CB1R proteins expression in placenta of male fetuses in HFD (A) and control (B,C). Black arrow shows the syncytiotrophoblast, green arrow shows cytotrophoblast (A), and yellow arrow shows endothelium (A, B, and C).

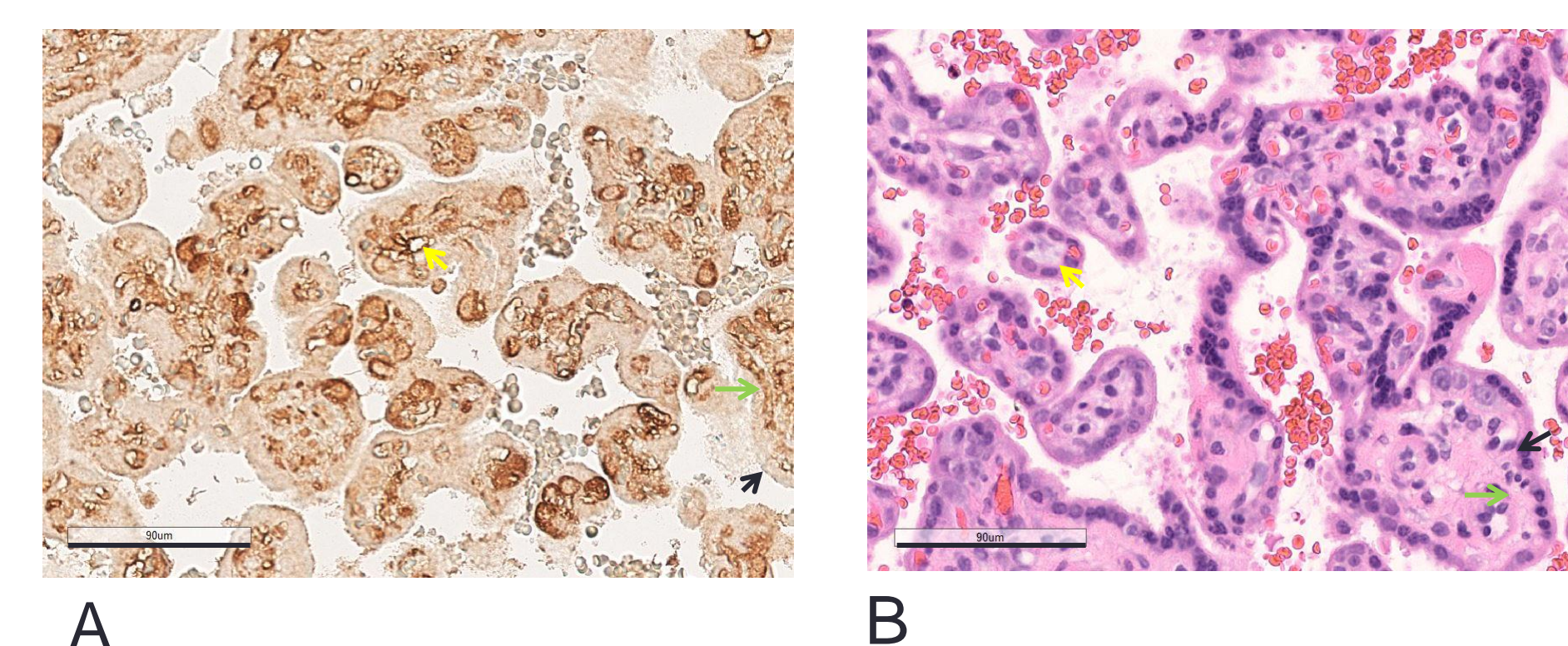
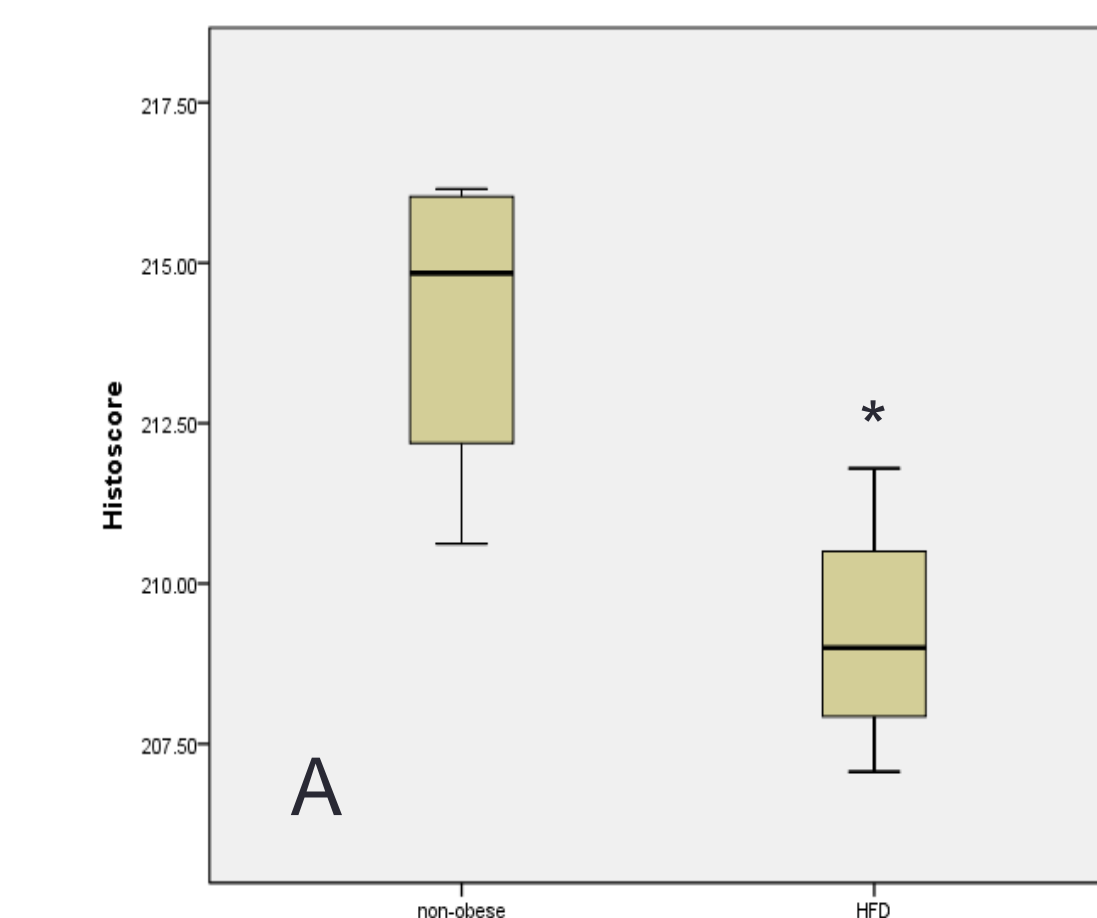


Figure 6. CB1R protein expression (A) and H&E staining (B) in placenta of male fetuses, control group. Black arrow is syncytiotrophoblast, green arrow is cytotrophoblast and yellow arrow endothelium.

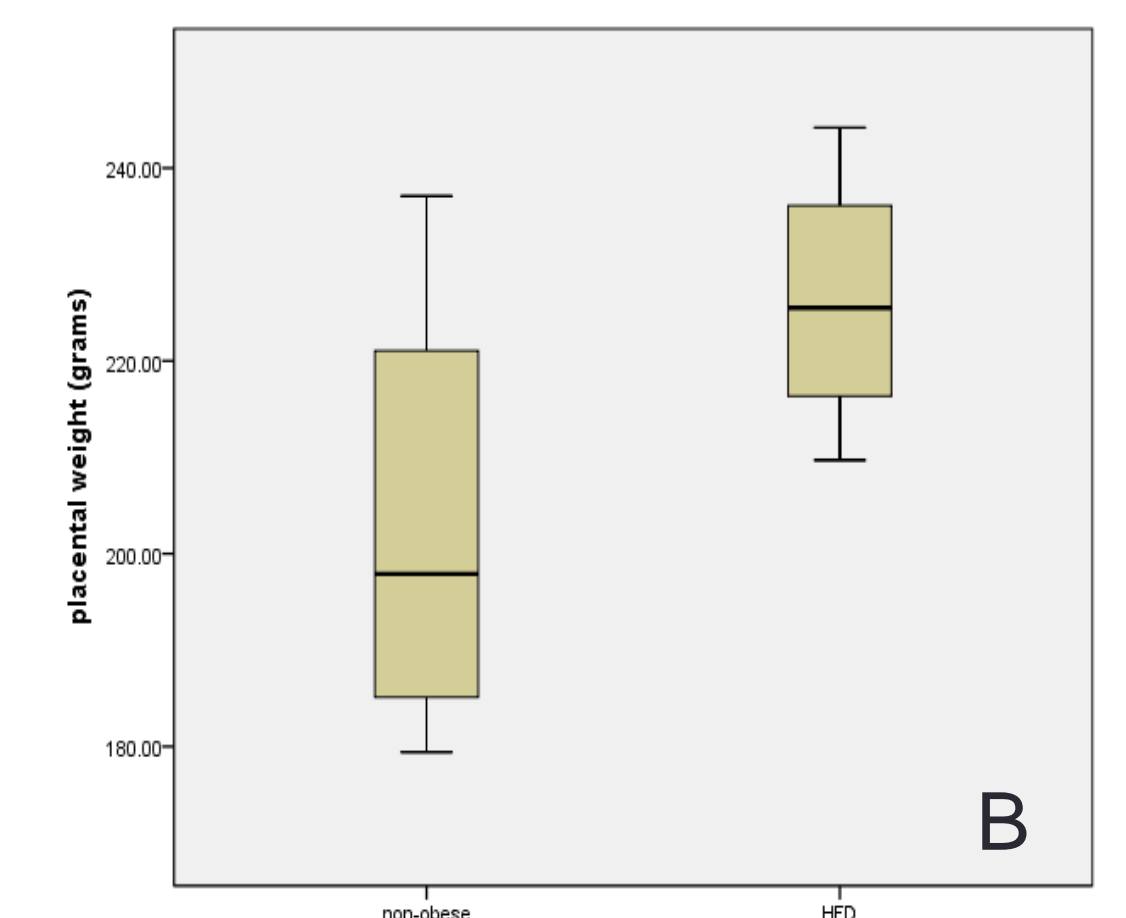
Results

CB1R is expressed in fetal endothelium, cytotrophoblast and syncytiotrophoblast (Figures 5, 6). The expression of CB1R protein was decreased in the villous tree of HFD animals (214.11 ± 1.28 vs 209.22 ± 0.98 ; CTR vs. HFD, respectively, $p=0.05$, Mann-Whitney U test). The placental (203.00 ± 12.49 g vs. 226.23 ± 7.13 g) and fetal (810 ± 17.76 g vs 849.59 ± 24.47 g) weights did not differ between the groups (Figure 7).

Placental CB1R expression in HFD (n=4) and non-obese (n=4) dams, carrying male fetus



Weight of placenta in HFD (n=4) and non-obese (n=4) dams, carrying male fetuses.



Weight of male fetuses delivered by control (n=4) and HFD (n=4) dams.

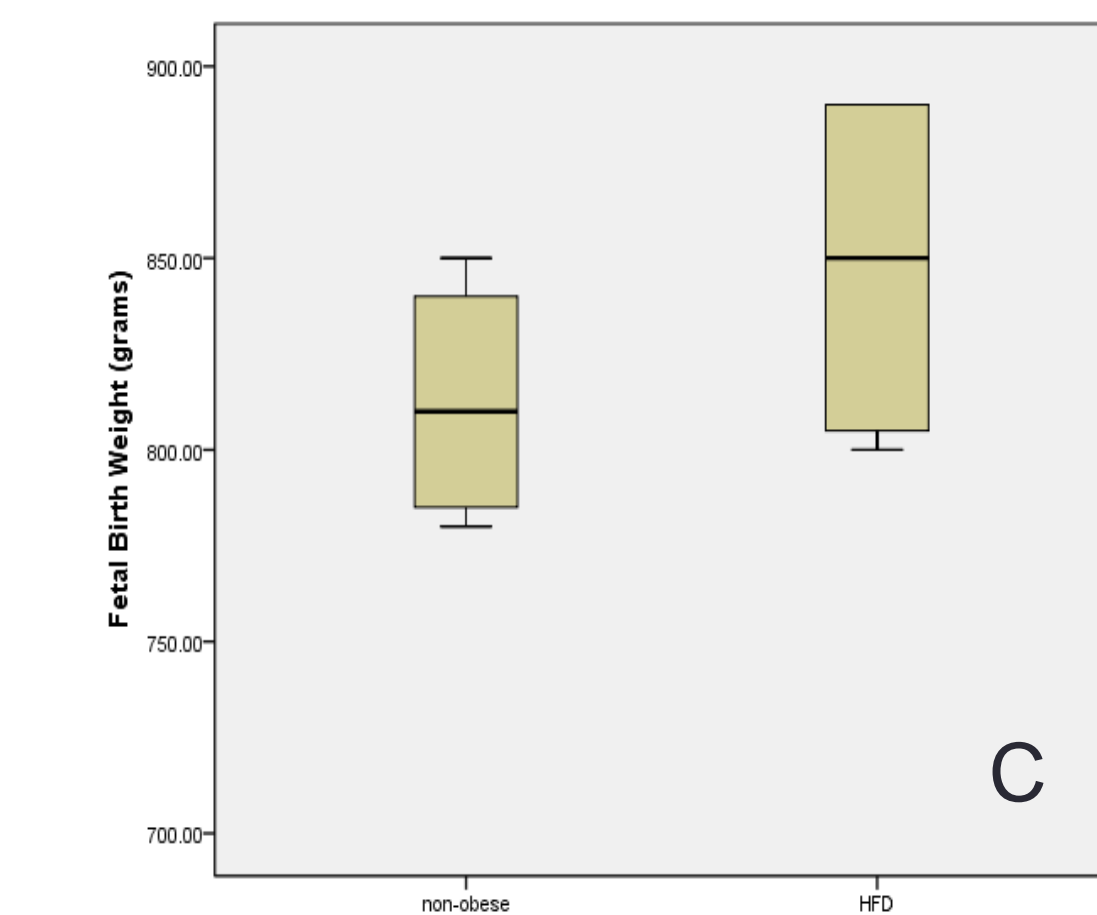


Figure 7. Placental CB1R protein expression in HFD (n=4) and non-obese (n=4) dams (A); placental (B) and fetal weights (C).

Conclusion

Availability of the cytoplasmic pool for placental CB1R is regulated by a high fat diet, but not increased food intake in this baboon model of maternal obesity. Decreased placental CBR1 availability might alter eCBs regulated fetal vascular reactivity

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Acknowledgement

Authors thank personnel of the Texas Biomedical Institute and Center for Pregnancy and Newborn Research (UTHSC—San Antonio) for their help. This study was partially supported by Texas Biomedical Research Institute Grant C06 RRO13556 and NIH grant HD21350 to Dr. Peter Nathanielsz (UTHSC—San Antonio), UTHSCSA ERC New Investigator Award to N.S.-L., NIH NCRR grant P51 RR013986 to the Southwest National Primate Research. This work was supported by the TTUHC start-up funds to N.S.-L., Dr. Carrillo's travel was supported by the NIH young investigator award. Also supported NIH R24OD010916 and P51 RR013986.