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A case of adrenal vein sampling in primary aldosteronism with homolateral suppression

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1 **Supplemental File**

2 **A CASE OF ADRENAL VEIN SAMPLING IN PRIMARY ALDOSTERONISM WITH**
3 **“HOMOLATERAL SUPPRESSION”**

4
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24
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1 **Immunohistochemistry**

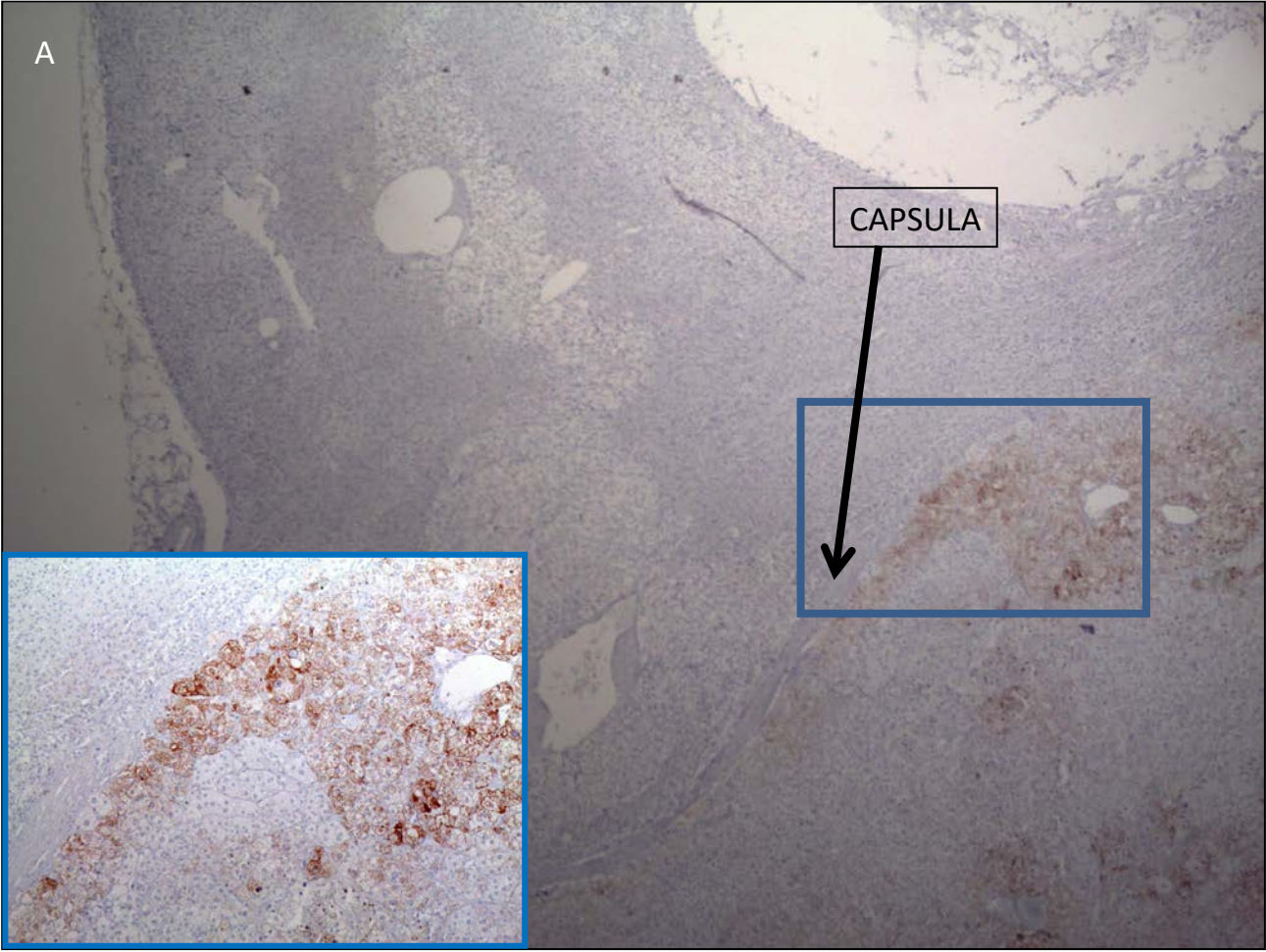
2 Sections of 10 µm thickness from paraffin-embedded adrenal tissue of the patient with APA were
3 quenched for endogenous peroxidase using H₂O₂ and subsequently incubated with antibodies
4 against human CYP11B1 (11β-hydroxylase, 1:1000 diluted) or human CYP11B2 (aldosterone
5 synthase, 1:100 diluted) for 1 h. After rinsing, the EnVision reagent (Dako, Carpinteria, CA)
6 coupled with peroxidase-labelled polymer was incubated as secondary antibody for 30 min. The
7 proteins were visualized using 3,3'-diaminobenzidine tetrahydrochloride counterstained with
8 haematoxylin. Both antibodies were kindly provided by prof. Celso Gomez-Sanchez (University of
9 Mississippi, Jackson, MS).

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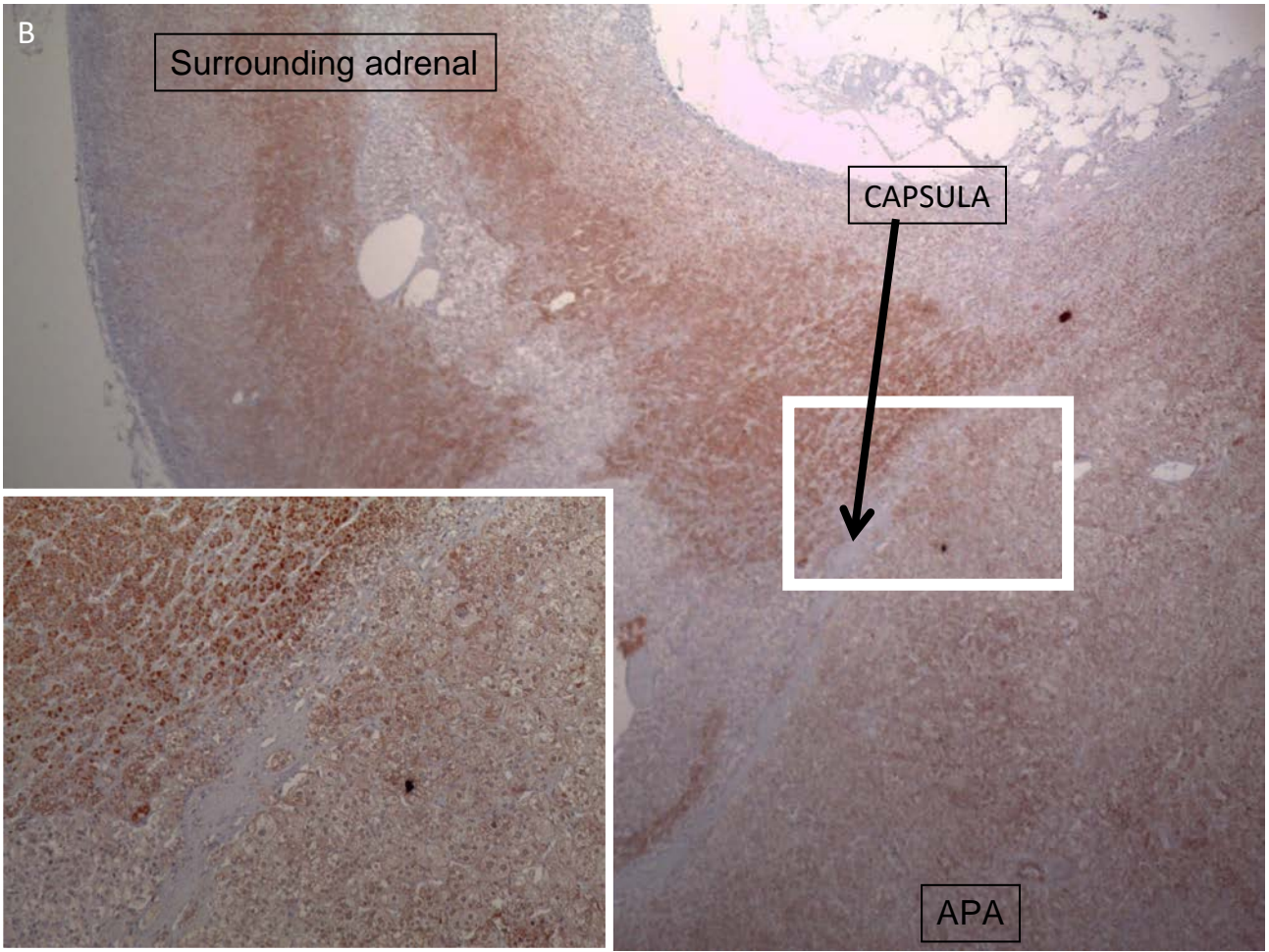
11 **Supplemental figure S1.**

12 Sections of adrenal tissue containing the aldosterone-producing adenoma (APA) and the
13 surrounding adrenal tissue were immuno-stained with antibodies against human aldosterone
14 synthase (CYP11B2) (**panel A**) and human 11β-hydroxylase (CYP11B1) (**panel B**).

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Unstimulated AVS	Routine Cortisol (µg/dl)	Aldosterone (ng/dl)	SI	ACR
Infra-renal inferior vena cava	10.3	39.6	-	3.8
Left adrenal vein	65.1	78.2	6.3	1.2
Right adrenal vein – Sample 1 (APCC)	244	251	23.7	1
Right adrenal vein – Sample 2 (APA)	269	5310	26.1	19.7
Right adrenal vein – Sample 3 (central vein)	210	3767	20.4	17.9
Cosyntropin-stimulated AVS	Routine Cortisol (µg/dl)	Aldosterone (ng/dl)	SI	ACR
Infra-renal inferior vena cava	29	57.2	-	2
Left adrenal vein	183	1268	8.2	0.7
Right adrenal vein – Sample 1 (APCC)	778	622	26.8	0.8
Right adrenal vein – Sample 2 (APA)	575	6831	19.8	11.9
Right adrenal vein – Sample 3 (central vein)	625	5123	21.6	8.2

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2 **Table S1. Hormonal measurements from adrenal venous sampling of a second case.** AVS=

3 Adrenal Venous Sampling; SI= Selectivity Index; ACR= Aldosterone/Cortisol Ratio; APCC=

4 aldosterone-producing cell cluster.

5 This table refers to a different patient who underwent super-selective AVS: the LI (ACR right/ACR

6 left) from the central adrenal vein is 14.9 during unstimulated AVS and 11.7 during cosyntropin

7 infusion. ACR from the left adrenal vein and from the adrenal branch draining the area where

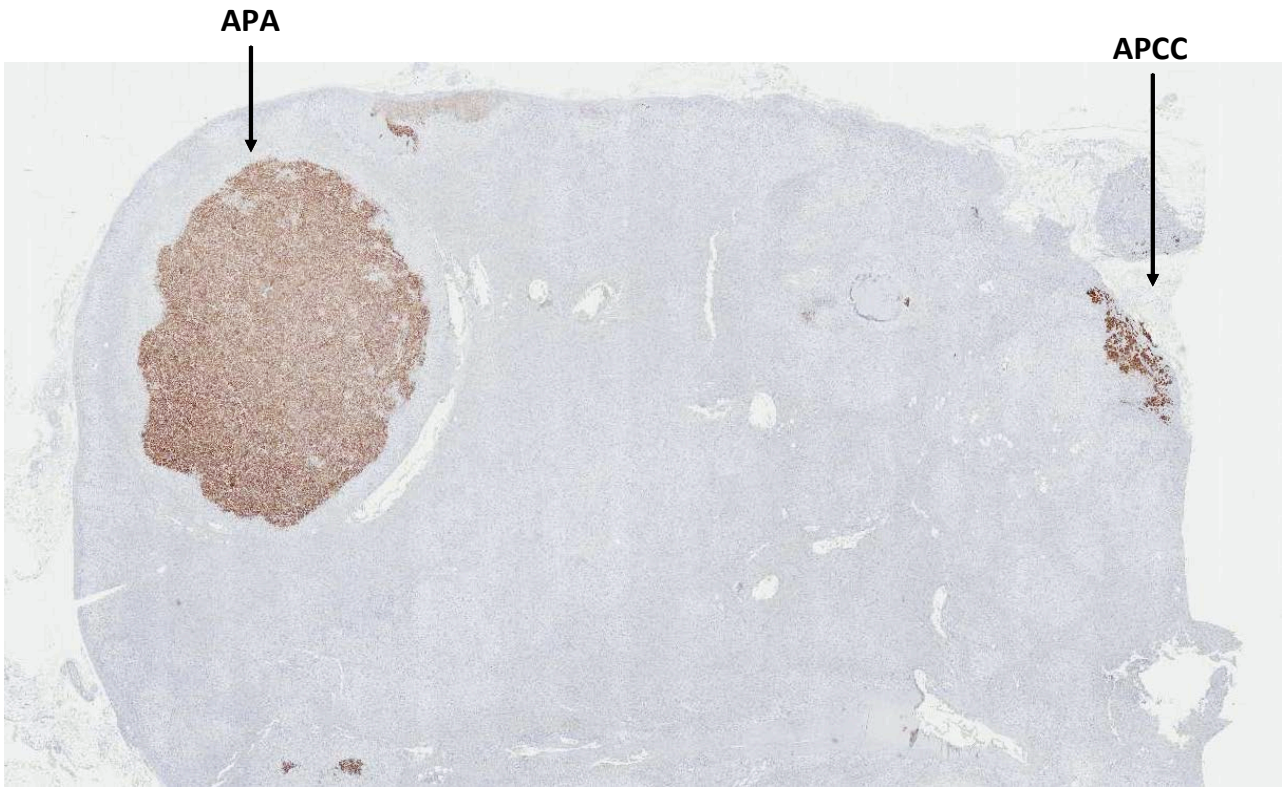
8 APCC is present are less than ACR from the peripheral vein, both under basal condition and after

9 cosyntropin infusion. The presence of an APCC outside the main nodule does not affect

10 significantly aldosterone production and ACRs in this patient.

1 **Supplemental figure S2.**

2 Sections of adrenal tissue from patient described in table S1, containing the aldosterone-producing
3 adenoma (APA) and the surrounding adrenal tissue containing an aldosterone-producing cell cluster
4 (APCC), immuno-stained with antibodies against human aldosterone synthase (CYP11B2).



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