

Supplementary materials

Diagnostic accuracy of aldosterone and renin measurement by chemiluminescence for screening of patients with primary aldosteronism

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Table S1. Correlation analysis for DRC by CLIA vs PRA by RIA and aldosterone by CLIA vs RIA measurements. PRA and DRC are reported as international unit (SI) and after normalization by Z-score. The table reports sample size (N), Pearson's coefficient (R), and the equation of regression line. *Quadratic fit-line R value and equation line. Sub-analyses were performed for PRA lower and equal or higher than $1 \text{ ng x ml}^{-1} \text{ x h}^{-1}$, and for aldosterone (RIA) lower and equal or higher than 100 ng/L . *P*-value < 0.05 were considered significant. AC, aldosterone concentration; CLIA, chemiluminescence; DRC, direct renin concentration; RIA, radio-immuno assay.

Variable		N	R	P-value	Regression line
All data					
DRC (CLIA) vs. PRA (RIA)	Z-score	918	+ 0.422	<0.001	$Y = 0.42 * X - 0.001$
			+ 0.423	<0.001	$Y = 6.97 * X + 25.88$
	SI		+ 0.553*	<0.001	$Y = 6.9 + 16.6 * X - 0.07 * X^2$
AC (CLIA) vs. AC (RIA) – ng/L		929	+ 0.945	<0.001	$Y = 0.89 * X + 19.03$
PRA < $1 \text{ ng x ml}^{-1} \text{ x h}^{-1}$					
DRC (CLIA) vs. PRA (RIA)	Z-score	524	– 0.018	0.677	$Y = - 0.58 * X - 0.37$
	SI		– 0.026	0.558	$Y = - 13.89 * X + 20.33$
PRA $\geq 1 \text{ ng x ml}^{-1} \text{ x h}^{-1}$					
DRC (CLIA) vs. PRA (RIA)	Z-score	394	+ 0.668	<0.001	$Y = 0.39 * X + 0.15$
	SI		+ 0.668	<0.001	$Y = 6.35 * X + 47.29$
Aldosterone (RIA) < 100 ng/L					
AC (CLIA) vs. AC (RIA) – ng/L		410	+ 0.656	<0.001	$Y = 0.96 * X + 22.01$
Aldosterone (RIA) $\geq 100 \text{ ng/L}$					
AC (CLIA) vs. AC (RIA) – ng/L		519	+ 0.942	<0.001	$Y = 0.91 * X + 9.68$

Table S2. Bland-Altman analysis for DRC by CLIA vs PRA by RIA and aldosterone by CLIA vs RIA measurements. PRA and DRC are reported after normalization by Z-score. The table reports sample size (N), mean value for the x-axis (Z-score for PRA/DRC, and mean of aldosterone measured by CLIA and RIA), mean difference (reported as absolute value, n, or as percentage, %). Sub-analyses were performed for PRA lower and equal or higher than $1 \text{ ng} \times \text{mL}^{-1} \times \text{h}^{-1}$, for aldosterone (RIA) lower and equal or higher than 100 ng/L , and for quartiles of PRA/DRC and aldosterone. AC, aldosterone concentration; CLIA, chemiluminescence; DRC, direct renin concentration; N.A., not available; RIA, radio-immuno assay.

Variable	N	Mean (x-axis)	Mean Difference (n) (y-axis)	Mean Difference (%) (y-axis)
All data				
DRC (CLIA) vs. PRA (RIA) – Z score	918	– 0.22	N.A.	+ 5.8± 4.10
AC (CLIA) vs. AC (RIA) – ng/L	929	+ 157	+ 2.2± 2.15	+ 2.3± 1.32
PRA < 1 ng/mL/min (mean Z-score < -0.19)				
DRC (CLIA) vs. PRA (RIA) – Z score	524	– 0.26	N.A.	+ 2.4± 0.72
PRA ≥ 1 ng x mL⁻¹ x h⁻¹ (mean Z-score ≥ -0.19)				
DRC (CLIA) vs. PRA (RIA) – Z score	394	+ 0.35	N.A.	+ 10.4± 9.54
PRA/DRC Quartile (mean Z-score)				
I Quartile (<–0.28)	202	– 0.30	N.A.	+ 4.7± 0.31
II Quartile (–0.28 - –0.21)	232	– 0.26	N.A.	+ 3.0 ± 0.99
III Quartile (–0.22 - –0.06)	250	– 0.17	N.A.	+ 0.2 ± 3.85
IV Quartile (≥–0.07)	234	+ 0.69	N.A.	+ 15.5 ± 15.54
Aldosterone < 100 ng/L (mean CLIA/RIA)				
AC (CLIA) vs. AC (RIA)	371	+ 65.2	+ 13.0± 1.26	+ 12.7± 2.00
Aldosterone ≥ 100 ng/L (mean CLIA/RIA)				
AC (CLIA) vs. AC (RIA)	558	+ 218.8	– 4.9± 3.45	– 4.5± 1.70
Aldosterone Quartile (ng/L; mean CLIA/RIA)				
I Quartile (< 76.3)	232	+ 51.7	+ 11.1 ± 1.47	+ 12.4 ± 2.78
II Quartile (76.3 – 119.0)	232	+ 97.1	+ 14.8 ± 1.96	+ 10.5 ± 2.04
III Quartile (120.0 – 185.0)	231	+ 149.0	+ 10.4 ± 3.05	+ 1.7 ± 2.41
IV Quartile (≥186.0)	234	+ 330.4	– 26.9 ± 7.30	– 15.1 ± 2.89

Table S3. Diagnostic performance of ADRR vs ARR. The table reports ROC curve analysis comparing ADRR calculated using aldosterone and DRC by CLIA and ARR calculated using aldosterone and PRA by RIA. Sample size (N; PA, UPA, and BiPA patients are reported for each analysis between square brackets), area under the curve (AUC), 95% confidence interval (95% CI) and asymptotical significance are shown together with cut-off corresponding to highest accuracy (according to Youden index*), highest sensitivity (#) and specificity (§). *P*-value < 0.05 were considered significant. ADRR, aldosterone-to-direct renin ratio; AC, aldosterone concentration; ARR, aldosterone-to-renin ratio; BiPA, bilateral primary aldosteronism; DRC, direct renin concentration; PA, primary aldosteronism; PRA, plasma renin activity; RIA, radio-immuno assay; UPA, unilateral primary aldosteronism.

	Variable	N	AUC	95% CI	<i>P</i> -value	Cut-off	Sens (%)	Spec (%)
PA Patients	ADRR (DRC – CLIA) (AC ≥ 100 ng/L)	423 [87]	0.928	0.904-0.954	<0.001	9 [#]	100.0	53.6
						20	93.3	81.1
						25*	91.1	85.3
						27	86.5	87.1
						436 [§]	3.4	100.0
ARR (PRA – RIA) (AC ≥ 100 ng/L)	359 [89]	0.943	0.920-0.966	<0.001	200 [#]	100.0	66.5	
					300	93.1	78.7	
					436*	89.7	87.1	
					2600 [§]	13.8	100.0	
					UPA Patients	ADRR (DRC – CLIA) (AC ≥ 100 ng/L)	423 [23]	0.884
20	95.7	69.3						
27	87.0	75.0						
37*	86.9	78.8						
483 [§]	4.3	100.0						
ARR (PRA – RIA) (AC ≥ 100 ng/L)	359 [23]	0.885	0.835-0.934	<0.001	200 [#]	100.0	53.9	
					300	95.7	65.2	
					460*	95.7	74.1	
					8000 [§]	8.7	100.0	
					BiPA Patients	ADRR (DRC – CLIA) (AC ≥ 100 ng/L)	400 [33]	0.866
17.5*	93.4	71.0						
20	87.9	74.3						
27	78.8	80.1						
436 [§]	6.1	99.7						
ARR (PRA – RIA) (AC ≥ 100 ng/L)	336 [33]	0.906	0.869-0.944	<0.001	230 [#]	100.0	65.4	
					300	93.9	71.6	
					472*	87.9	81.2	
					2645 [§]	15.2	99.7	

Figure S1. Correlation sub-analyses for aldosterone and PRA/DRC assessed by Pearson's R test. Green dot: patients with EH; red dot: patients with PA; grey dot: patients without a defined diagnosis; dashed lines: 95% confidence interval; continuous line: regression curve. P -value < 0.05 was considered significant. **(A)** Aldosterone concentration by CLIA vs RIA in patients with an AC < 100 ng/L. **(B)** Aldosterone concentration by CLIA vs RIA in patients with an AC \geq 100 ng/L. **(C)** DRC vs PRA in patients with a PRA < 1 ng \times ml⁻¹ \times h⁻¹. **(D)** DRC vs PRA in patients with a PRA \geq 1 ng \times ml⁻¹ \times h⁻¹. AC, aldosterone concentration; CLIA, chemiluminescence; DRC, direct renin concentration; EH, essential hypertension; PA, primary aldosteronism; PRA, plasma renin activity; RIA, radio-immuno assay; Und, undefined diagnosis.

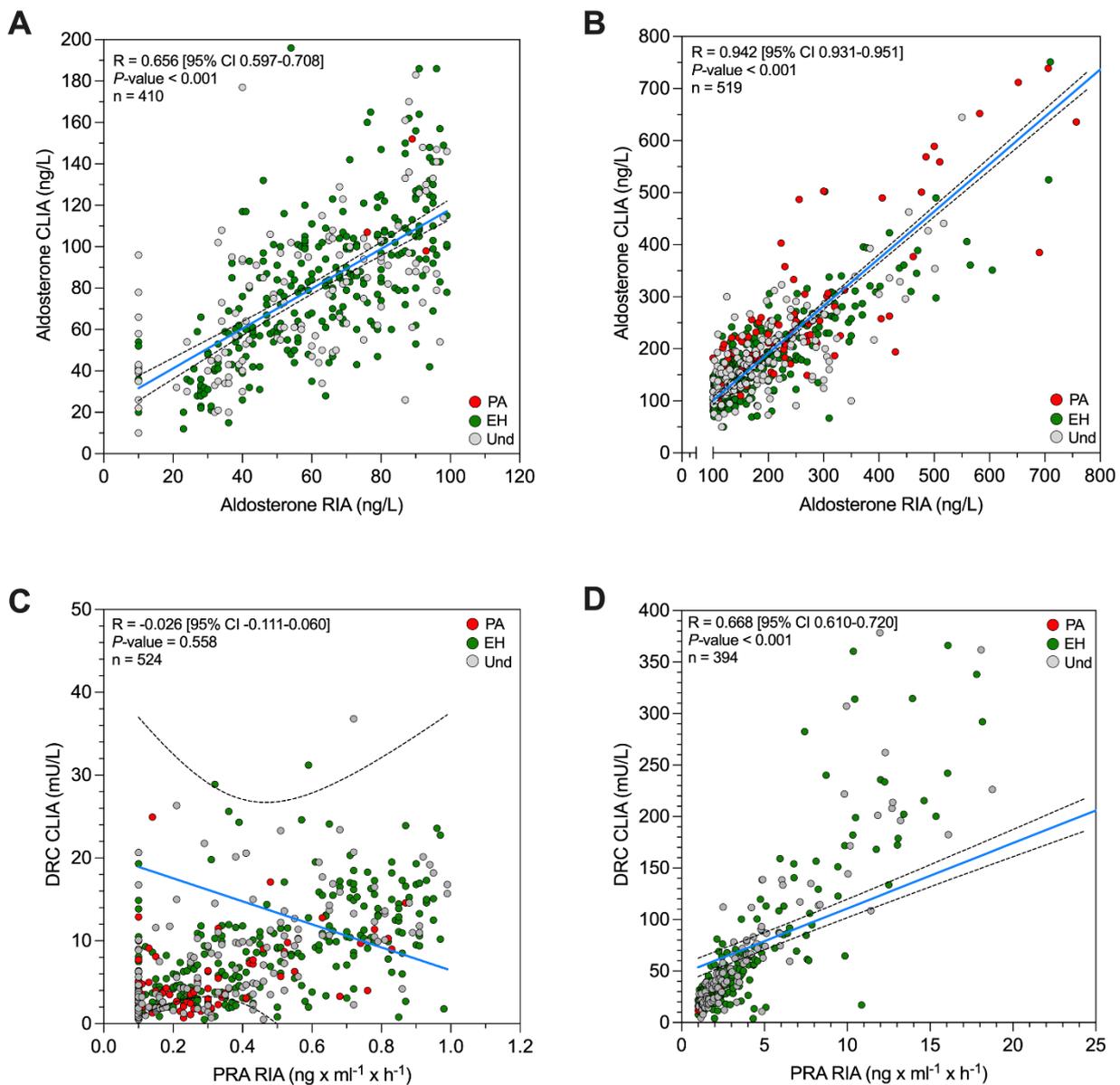


Figure S2. Bland-Altman sub-analyses for aldosterone and PRA/DRC. Green dot: patients with EH; red dot: patients with PA; grey dot: patients without a defined diagnosis. Continuous red line represents mean difference between measurement of DRC by CLIA and PRA by RIA; blue dashed lines represent 95% confidence interval. **(A)** On x-axis mean AC measurement by CLIA and RIA assays; on y-axis difference between AC measurement by CLIA and RIA assays. Aldosterone is reported in ng/L. **(B)** On x-axis mean measurement of DRC by CLIA and PRA by RIA assays; on y-axis difference between measurement of DRC by CLIA and PRA by RIA assays. DRC and PRA are reported after normalization by Z-score. AC, aldosterone concentration; CLIA, chemiluminescence; DRC, direct renin concentration; EH, essential hypertension; PA, primary aldosteronism; PRA, plasma renin activity; RIA, radio-immuno assay; Und, undefined diagnosis.

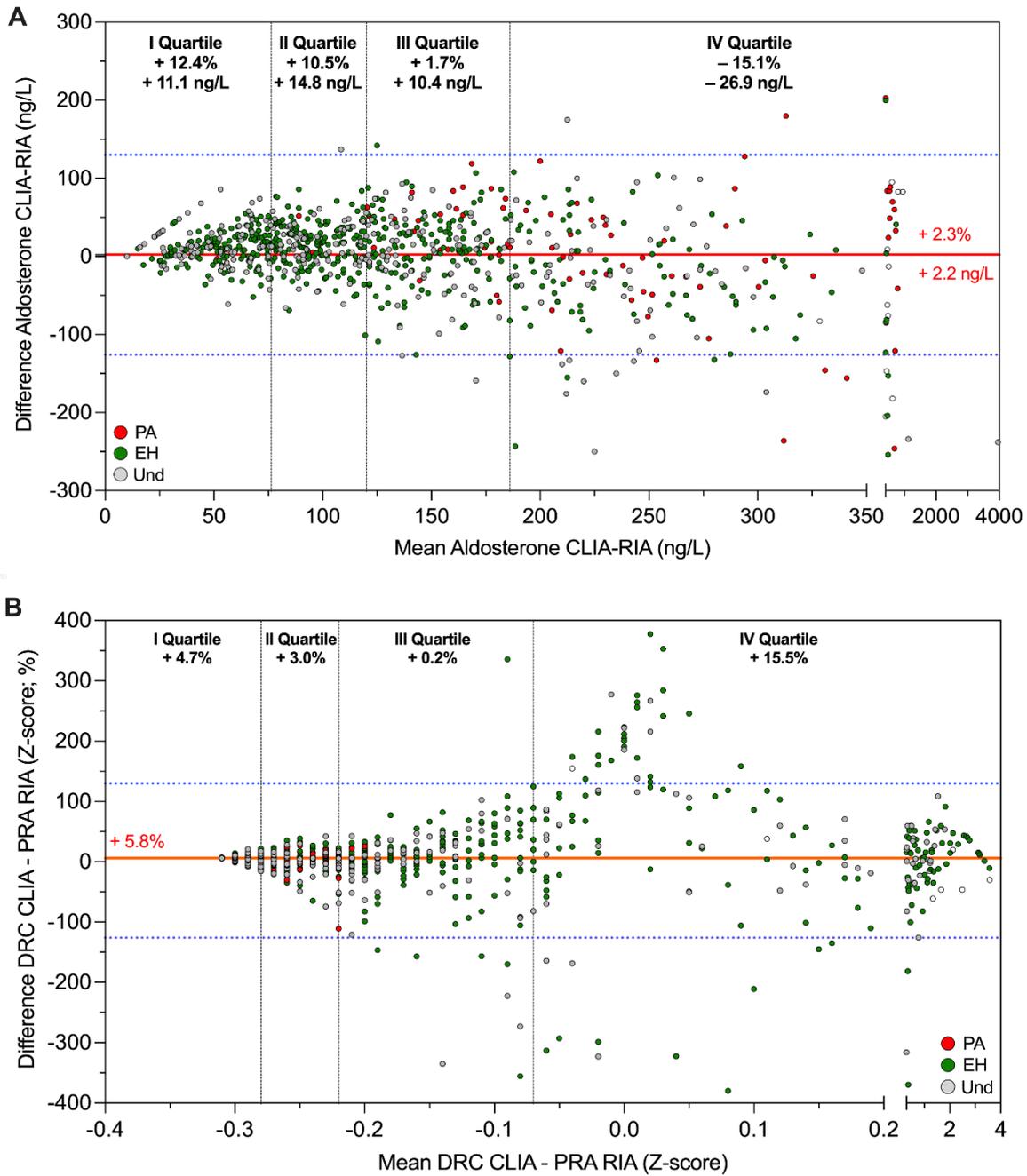


Figure S3. Correlation sub-analyses for PRA/DRC after normalization by Z-score assessed by Pearson's R test. Green dot: patients with EH; red dot: patients with PA; grey dot: patients without a defined diagnosis; dashed lines: 95% confidence interval; continuous line: regression curve. *P*-value < 0.05 was considered significant. **(A)** DRC vs PRA after normalization by Z-score. **(B)** DRC vs PRA in patients with a PRA < 1 ng x ml⁻¹ x h⁻¹ after normalization by Z-score. **(C)** DRC vs PRA in patients with a PRA ≥ 1 ng x ml⁻¹ x h⁻¹ after normalization by Z-score. CLIA, chemiluminescence; DRC, direct renin concentration; EH, essential hypertension; PA, primary aldosteronism; PRA, plasma renin activity; RIA, radio-immuno assay; Und, undefined diagnosis.

