

**ABSOLUTE QUANTITATION OF DRUG METABOLIZING CYTOCHROME P450 ENZYMES
AND ACCESSORY PROTEINS IN DOG LIVER MICROSOMES USING LABEL-FREE
STANDARD-FREE ANALYSIS REVEALS INTER-BREED VARIABILITY**

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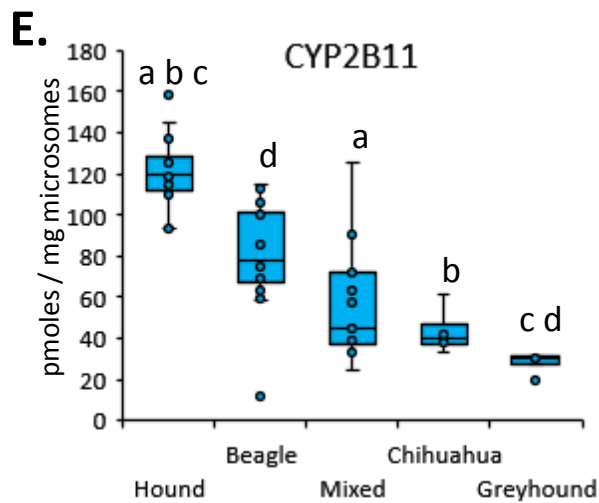
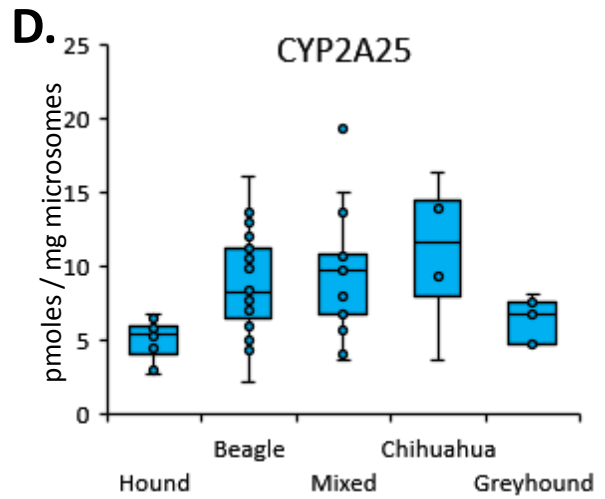
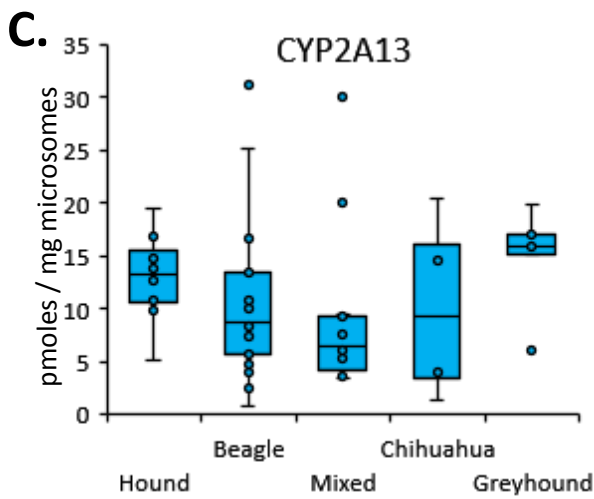
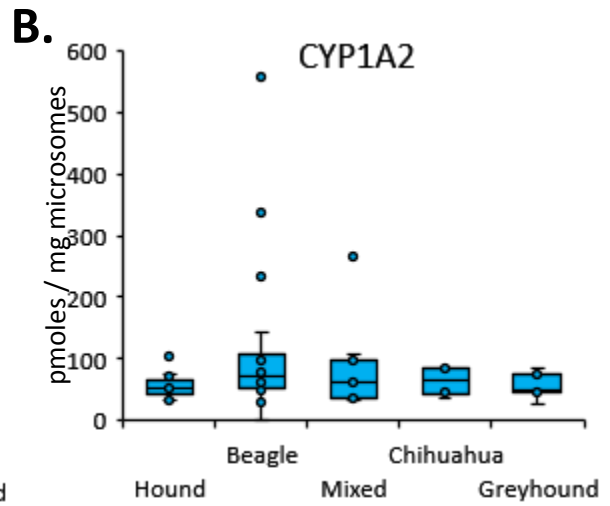
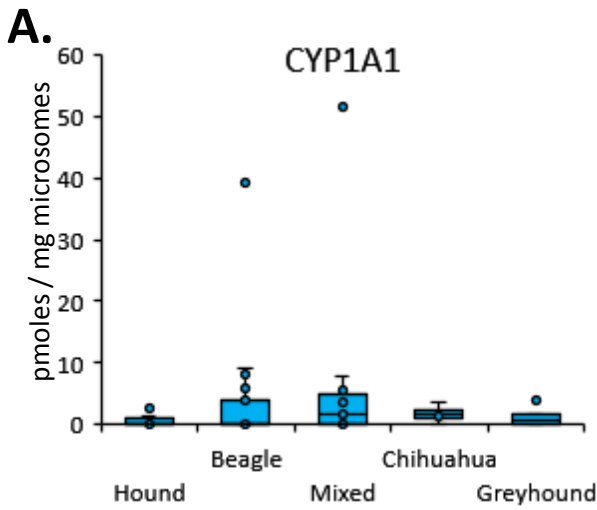
SUPPLEMENTAL DATA

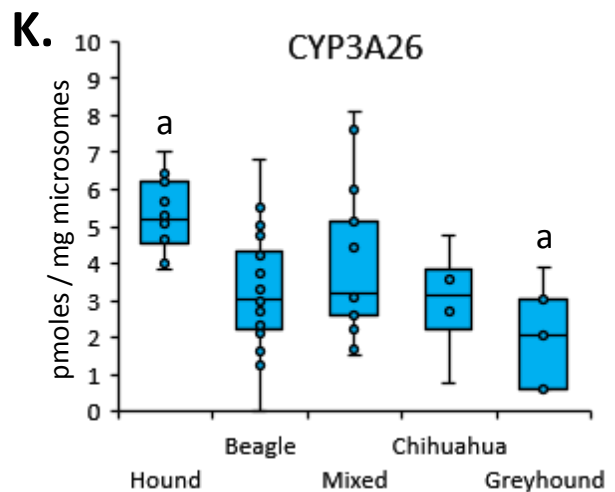
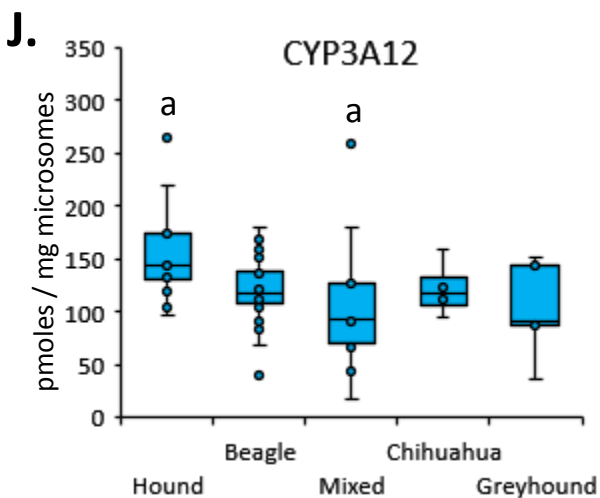
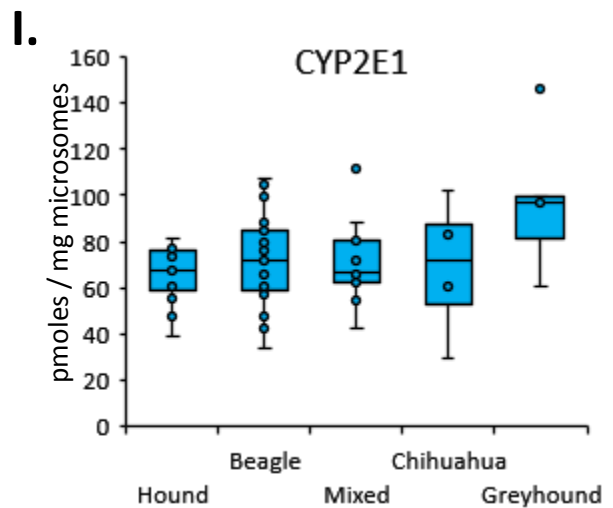
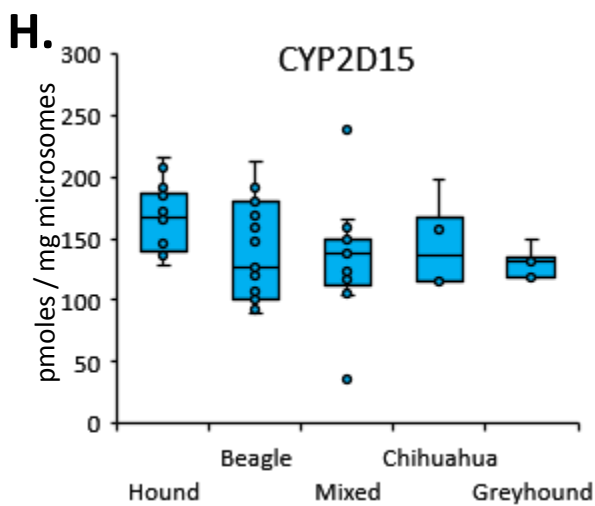
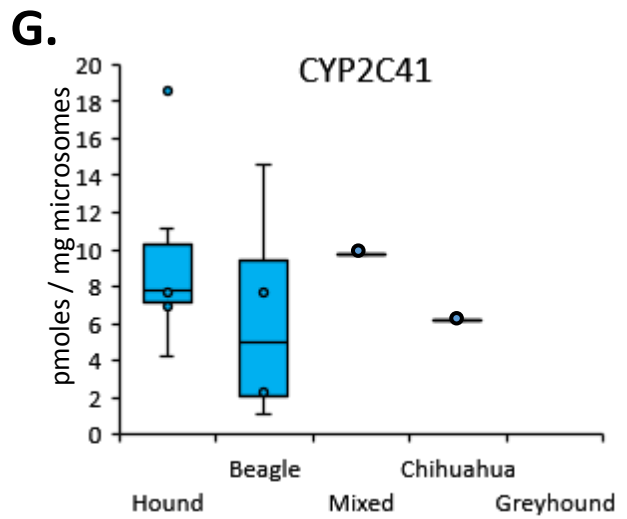
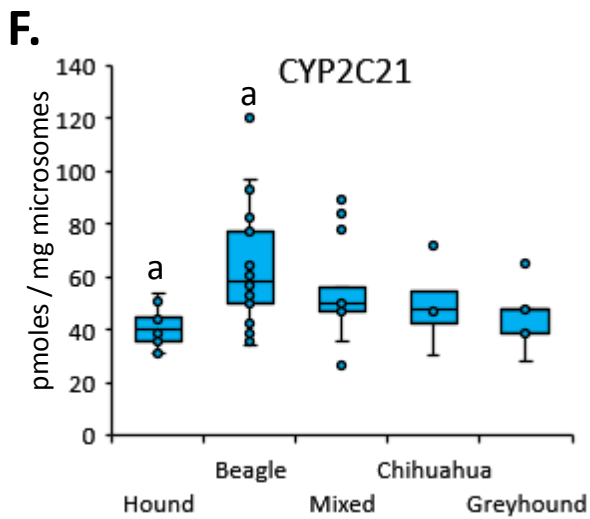
Table S1. Abundance of CYP enzymes and accessory proteins (P450 oxidoreductase, POR; cytochrome b5A, CYB5A) in a bank of liver microsomes (N =59) representing 4 different breeds and mixed-breed dogs measured by LC-MS/MS.

Liver ID	Breed	Sex	CYP1A1	CYP1A2	CYP2A13	CYP2A25	CYP2B11	CYP2C21	CYP2C41	CYP2D15	CYP2E1	CYP3A12	CYP3A26	POR	CYB5A
DLM01	Research Beagle	M	--	78.7	8.9	6.0	113.5	42.2	--	163.1	57.1	158.0	3.0	45.1	463.8
DLM02	Research Beagle	M	--	64.9	13.7	6.5	245.9	50.4	--	92.3	58.4	136.3	2.3	33.3	506.1
DLM03	Greyhound	M	--	73.8	15.9	6.8	30.3	47.5	--	148.9	99.2	87.1	--	31.9	382.4
DLM04	Greyhound	M	--	25.0	15.1	7.6	27.8	38.7	--	131.2	146.1	151.7	2.1	41.7	328.3
DLM05	Mixed	F	51.7	266.5	4.2	9.8	72.0	28.7	--	36.1	42.5	179.2	3.2	84.8	402.1
DLM06	Mixed	F	4.9	108.0	7.6	13.7	39.6	26.4	--	105.5	81.1	93.7	1.7	51.6	425.3
DLM07	Mixed	F	7.9	102.8	4.3	15.0	73.9	46.9	--	140.2	71.7	133.0	2.2	35.6	431.0
DLM08	Greyhound	M	3.9	83.7	6.1	4.7	19.9	28.3	--	118.2	60.3	36.8	--	23.9	313.4
DLM09	Greyhound	F	1.5	48.4	17.1	4.8	30.9	47.8	--	118.1	81.1	91.2	3.0	30.0	459.5
DLM10	Greyhound	F	--	44.5	19.9	8.2	31.3	65.0	--	135.1	97.0	143.8	3.9	35.0	491.2
DLM11	Mixed	F	3.8	61.5	9.4	6.8	57.2	48.3	--	158.7	80.3	43.7	1.5	28.1	393.5
DLM12	Mixed	M	--	37.8	9.3	5.6	125.1	46.6	--	111.8	54.6	65.6	3.1	26.0	381.1
DLM13	Mixed	M	--	37.0	6.5	3.6	32.9	35.9	--	123.3	62.5	70.0	3.1	19.1	296.4
DLM14	Mixed	F	1.2	35.5	6.1	10.1	36.6	50.7	--	148.5	61.9	93.4	2.6	34.6	428.8
DLM15	Research Beagle	M	--	39.0	6.0	7.3	64.8	34.2	--	125.8	76.2	40.6	2.1	24.4	254.2
DLM16	Research Beagle	M	--	49.9	4.0	7.1	76.8	39.0	--	127.3	75.9	90.6	2.7	28.0	329.1
DLM17	Mixed	M	--	36.2	20.0	4.1	24.4	50.0	--	104.6	65.9	127.3	4.5	23.2	398.0
DLM18	Mixed	M	1.6	71.1	3.7	7.9	35.6	50.3	--	165.2	65.0	94.7	4.6	41.0	424.4
DLM19	Mixed	M	5.7	97.9	8.3	19.3	91.0	84.0	--	140.5	67.0	258.7	6.0	50.8	546.5
DLM20	Research Beagle	M	3.9	107.1	--	8.8	114.6	51.4	--	120.2	33.5	118.4	2.2	41.1	351.7
DLM21	Mixed	F	--	40.1	5.3	10.8	39.4	89.6	9.7	137.8	72.9	91.8	7.6	45.8	364.8
DLM22	Mixed	M	4.5	73.3	3.4	10.6	63.2	78.2	--	116.4	88.4	91.0	5.2	45.6	354.9
DLM23	Chihuahua	M	3.7	84.3	1.3	16.4	38.1	48.5	6.2	114.8	60.8	111.2	4.8	36.5	420.6
DLM24	Chihuahua	M	--	44.9	4.1	13.9	61.3	72.1	--	198.4	102.0	123.6	--	42.2	469.7
DLM25	Chihuahua	M	1.9	86.1	20.5	9.4	42.1	46.9	--	157.0	82.9	94.4	2.7	45.9	432.9

DLM26	Chihuahua	F	1.3	35.6	14.6	3.6	32.7	30.0	--	115.4	29.8	158.7	3.5	33.3	259.9
DLM27	Mixed	M	--	32.7	30.1	7.1	45.0	55.9	--	238.9	111.7	16.5	8.1	57.0	505.0
DLM28	Research Beagle	F	6.7	234.1	7.4	2.2	62.1	77.2	--	95.1	59.2	107.9	2.2	60.8	400.8
DLM29	Research Beagle	F	39.2	349.4	10.9	5.0	89.3	82.4	--	93.0	104.7	135.4	3.3	48.3	572.8
DLM30	Research Beagle	M	9.2	337.8	5.6	12.5	101.3	93.3	--	100.1	60.6	138.6	3.0	58.0	539.5
DLM31	Research Beagle	M	6.0	557.3	2.5	11.2	113.1	50.1	14.6	92.0	74.3	174.9	6.8	64.9	478.4
DLM32	Research Beagle	M	--	143.6	4.8	9.8	101.1	65.4	--	121.2	107.5	180.7	3.3	46.4	567.7
DLM33	Research Beagle	F	8.2	241.8	5.5	5.4	66.9	79.7	--	99.9	65.5	109.7	3.0	54.9	470.8
DLM34	Research Hound	F	1.2	43.7	19.5	5.6	121.0	39.0	--	164.7	47.4	264.7	6.3	44.3	465.8
DLM35	Research Hound	F	2.6	76.0	13.9	6.8	125.8	41.8	--	214.9	67.9	143.6	6.5	43.7	360.2
DLM36	Research Hound	F	1.5	63.6	17.2	6.5	158.6	41.9	7.9	184.6	55.1	145.8	6.2	37.1	447.4
DLM37	Research Hound	F	--	40.0	9.8	5.3	118.4	31.5	6.9	136.1	77.3	173.7	3.8	40.1	357.7
DLM38	Research Hound	F	--	31.6	10.7	5.8	121.6	53.6	--	172.6	60.6	175.3	5.3	36.8	461.4
DLM39	Research Hound	F	--	52.7	11.0	2.7	144.8	43.8	--	169.3	39.0	133.2	5.7	44.9	424.1
DLM40	Research Hound	F	--	33.4	12.8	3.0	93.3	35.8	--	128.3	73.7	119.2	4.1	41.9	400.7
DLM41	Research Hound	F	--	104.0	5.1	5.4	109.5	30.9	7.7	140.5	67.5	218.6	4.7	33.9	410.9
DLM42	Research Hound	F	--	42.9	14.7	3.2	113.2	46.2	--	146.8	62.7	96.8	4.0	42.0	370.2
DLM43	Research Hound	F	--	53.9	10.5	4.4	93.7	51.0	4.2	208.3	81.2	103.6	5.1	33.9	449.1
DLM44	Research Hound	F	--	58.3	16.9	6.7	137.0	38.0	18.6	192.1	77.5	137.5	7.1	39.3	457.5
DLM45	Research Hound	F	--	72.9	15.1	5.2	114.4	36.1	11.1	137.1	76.1	173.9	4.9	35.6	482.9
DLM46	Research Beagle	M	--	63.2	10.0	12.0	105.7	56.5	--	190.8	68.6	108.4	4.2	29.9	512.5
DLM47	Research Beagle	M	--	72.5	7.8	6.5	58.7	44.3	--	107.7	84.3	93.9	2.7	29.7	439.1
DLM48	Research Beagle	M	--	63.9	8.3	10.5	70.2	50.0	--	161.7	56.8	120.9	4.4	28.7	388.4
DLM49	Research Beagle	M	--	48.1	11.3	7.2	86.1	58.5	--	212.6	47.5	138.7	5.0	30.8	488.8

DLM50	Research Beagle	M	--	57.7	4.8	8.4	69.4	57.1	--	109.5	42.9	112.5	5.5	32.6	383.6
DLM51	Research Beagle	M	--	53.5	8.6	16.0	75.3	65.6	--	169.4	72.2	151.1	2.2	33.8	506.5
DLM52	Research Beagle	M	4.3	96.6	13.8	9.8	77.8	53.2	2.3	180.8	88.1	158.9	3.2	48.8	460.9
DLM53	Research Beagle	M	--	30.1	11.2	4.3	102.5	120.0	--	147.6	91.4	103.5	--	42.4	497.6
DLM54	Research Beagle	M	--	35.6	13.9	8.2	63.4	77.9	--	192.1	79.6	82.6	1.2	42.9	430.0
DLM55	Research Beagle	M	--	--	13.5	4.4	77.1	96.8	--	181.7	99.8	118.0	3.7	44.5	547.9
DLM56	Research Beagle	M	--	82.0	8.4	13.6	100.3	64.0	1.1	183.4	71.5	167.9	4.8	35.7	470.5
DLM57	Research Beagle	F	--	77.4	16.7	7.7	11.6	35.5	--	88.6	99.6	68.6	1.6	38.6	424.7
DLM58	Research Beagle	F	--	77.7	31.2	13.0	59.3	62.7	7.7	159.1	73.9	123.3	4.3	48.6	462.9
DLM59	Research Beagle	F	--	70.9	25.3	11.4	78.9	60.4	--	190.7	71.8	111.4	5.2	55.3	460.7





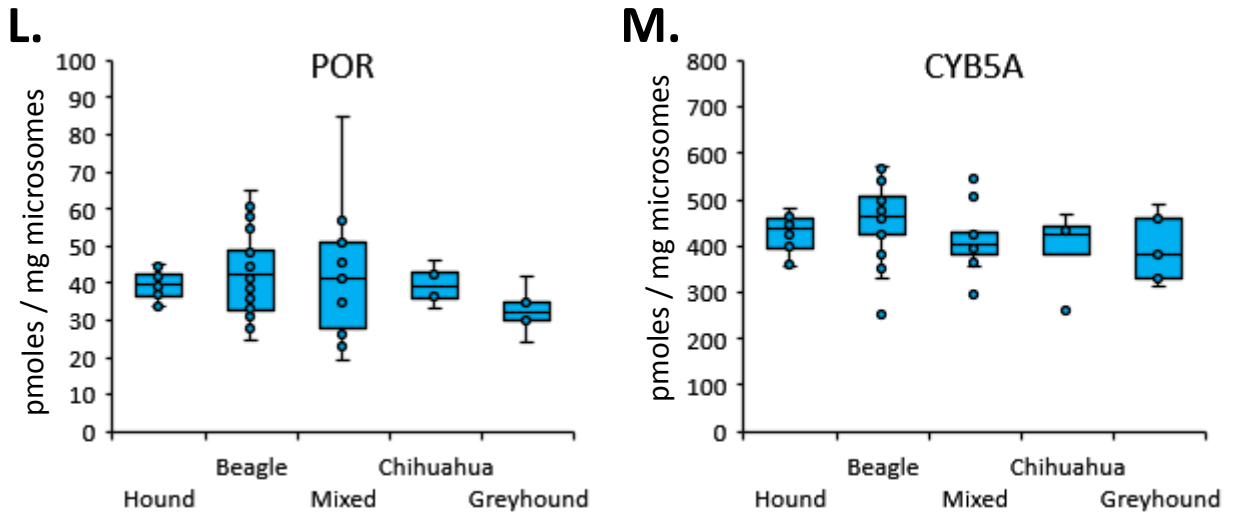


Figure S1 (panels A – M). Dog breed-associated variability in liver microsomal protein abundance (in pmoles / mg) of CYP1A1 (A), CYP1A2 (B), CYP2A13 (C), CYP2A25 (D), CYP2B11 (E), CYP2C21 (F), CYP2C41 (G), CYP2D15 (H), CYP2E1 (I), CYP3A12 (J), CYP3A26 (K), POR (L), and CYB5A (M). Shown in each plot are box and whisker plots representing the median, 25th percentile, 75th percentile, range, and outlier points for each breed. In most plots microsomal abundance data are from 12 purpose-bred research Hounds, 25 purpose-bred research Beagles, 13 mixed-breed dogs, 4 Chihuahuas, and 5 Greyhounds. However, the data for CYP2C41 (panel G) are limited to the 12 dogs (out of 59 dogs) that expressed at least one copy of the CYP2C41 gene, which included 6 Research Hounds, 4 Beagles, 1 Mixed-breed, and 1 Chihuahua. Differences between breeds were analyzed by ANOVA on rank transformed data followed by Dunn’s multiple comparisons testing. Lowercase letters (a, b, c, or d) shown on some plots (panels E, F, J, K) indicate breed groups that were significantly different from each other by Dunn’s test ($P < 0.05$ with same letter).