

Supplementary Table 1						
	P2-5 (n = 24)	P10-15 (n = 21)	P20-25 (n = 21)	P50-56 (n = 19)	P > 150 (n = 20)	oneway ANOVA + <i>post-hoc test</i>
C_m (nF)	0.04 ± 0.02	0.10 ± 0.04	0.16 ± 0.05	0.11 ± 0.02	0.14 ± 0.07	Kruskal-Wallis test: p < 0.0001 Dunn's multiple comparisons test: P2-5 vs. P10-15: p = 0.0005 P2-5 vs. P20-25: p < 0.0001 P2-5 vs. P50-56: p = 0.0001 P2-5 vs. P > 150: p < 0.0001 P10-15 vs. P 20-25: p = 0.0215 P10-15 vs. P 50-56: p > 0.9999 P10-15 vs. P > 150: p > 0.9999 P20-25 vs. P 50-56: p = 0.1059 P20-25 vs. P > 150: p > 0.9999 P50-56 vs. P 150: p > 0.9999
R_{in} (GΩ)	1.61 ± 0.88	0.74 ± 0.31	0.37 ± 0.28	0.25 ± 0.07	0.36 ± 0.24	Kruskal-Wallis test: p < 0.0001 Dunn's multiple comparisons test: P2-5 vs. P10-15: p > 0.9999 P2-5 vs. P20-25: p < 0.0001 P2-5 vs. P50-56: < 0.0001 P2-5 vs. P > 150: < 0.0001 P10-15 vs. P 20-25: p = 0.0098 P10-15 vs. P 50-56: p = 0.0001 P10-15 vs. P > 150: p = 0.0074 P20-25 vs. P 50-56: p > 0.9999 P20-25 vs. P > 150: p > 0.9999 P50-56 vs. P 150: p > 0.9999
E_{rest} (mV)	-58 ± 9	-68 ± 9	-70 ± 6	-72 ± 10	-72 ± 10	ANOVA: p < 0.0001 Bonferroni's multiple comparisons test P2-5 vs. P10-15: p = 0.0007 P2-5 vs. P20-25: p < 0.0001 P2-5 vs. P50-56: < 0.0001 P2-5 vs. P > 150: < 0.0001 P10-15 vs. P 20-25: p = 0.9792 P10-15 vs. P 50-56: p = 0.7538 P10-15 vs. P > 150: p = 0.7725 P20-25 vs. P 50-56: p = 0.9664 P20-25 vs. P > 150: p = 0.9736

						P50-56 vs. P 150: p > 0.9999
Development of passive membrane properties in M1LV neurons. Capacitance (C_m), input resistance (R_{in}), and resting membrane potential (E_{rest}) are reported as average \pm standard deviation. P values for multiple comparison and post-hoc test are reported for each paired comparison. The statistical tests used for normally distributed samples were one-way ANOVA and Bonferroni's multiple comparisons test (A. + Bonferroni). Non-normally distributed samples were compared with the Kruskal-Wallis test and Dunn's multiple comparisons test (K-W + Dunn's).						

Supplementary Table 2									
	P1	P3	P5	P7	P10	P15	P28	P55	P150
AIS length (μm)	(n = 4) 13.5 ± 0.8	(n = 6) 15.6 ± 0.4	(n = 6) 16.5 ± 0.4	(n = 6) 17.5 ± 0.8	(n = 6) 20.6 ± 1.4	(n = 6) 22.4 ± 2.1	(n = 6) 24.4 ± 1.7	(n = 6) 26.5 ± 1.2	(n = 6) 28.3 ± 0.5
AIS prox. diameter (μm)	(n = 3) 1.34 ± 0.09			(n = 3) 1.56 ± 0.15			(n = 3) 2.04 ± 0.15		
AIS dist. diameter (μm)	0.99 ± 0.05			1.07 ± 0.15			1.08 ± 0.04		
AIS distance from soma (μm)	1.79 ± 0.30			1.29 ± 0.21			1.07 ± 0.21		
Developmental AIS elongation in M1LV pyramidal neurons. AIS length, proximal and distal diameter, and distance from soma for M1LV neurons are reported as average \pm standard deviation. Number of animals (n) used for the analysis of each age group are reported below headers, indicating each respective age group (at least 100 AIS/animal).									

Supplementary Table 3

ANOVA $p < 0.0001$

Bonferroni's multiple comparisons test:

P1 vs. P3: $p = 0.2014$	P3 vs. P5: $p = 0.9415$	P5 vs. P7: $p = 0.9067$	P07 vs. P10: $p = 0.0037$	P10 vs. P15: $p = 0.2318$	P15 vs. P28: $p = 0.1485$	P28 vs. P55: $p = 0.1381$	P55 vs. P150: $p = 0.4086$
P1 vs. P5 $p = 0.0143$	P3 vs. P7: $p = 0.2174$	P5 vs. P10: $p < 0.0001$	P07 vs. P15: $p < 0.0001$	P10 vs. P2: $p = 0.0001$	P15 vs. P55: $p < 0.0001$	P28 vs. P150: $p = 0.0008$	
P1 vs. P7: $p = 0.0004$	P3 vs. P10: $p < 0.0001$	P5 vs. P15: $p < 0.0001$	P15: $p < 0.0001$	P10 vs. P28: $p < 0.0001$	P55: $p < 0.0001$		
P1 vs. P10: $p < 0.0001$	P3 vs. P15: $p < 0.0001$	P5 vs. P28: $p < 0.0001$	P07 vs. P28: $p < 0.0001$	P55: $p < 0.0001$	P15 vs. P150: $p < 0.0001$		
P1 vs. P15: $p < 0.0001$	P3 vs. P28: $p < 0.0001$	P5 vs. P55: $p < 0.0001$	P07 vs. P55: $p < 0.0001$	P10 vs. P150: $p < 0.0001$			
P1 vs. P28: $p < 0.0001$	P3 vs. P55: $p < 0.0001$	P5 vs. P150: $p < 0.0001$	P55: $p < 0.0001$				
P1 vs. P55: $p < 0.0001$	P3 vs. P150: $p < 0.0001$		P07 vs. P150: $p < 0.0001$				
P1 vs. P150: $p < 0.0001$							

Developmental AIS elongation in M1LV pyramidal neurons: AIS length; oneway ANOVA + post-hoc test.

Statistical comparison of AIS length between age groups, referring to data in Supplementary Table 2. P values for post-hoc test are reported for each paired comparison. The statistical tests used for comparing these (normally distributed) samples was one-way ANOVA and Bonferroni's multiple comparisons test (A. + Bonferroni).

Supplementary Table 4

	P2-5 (n = 24)	P10-15 (n = 21)	P20-25 (n = 21)	P50-56 (n = 19)	P > 150 (n = 20)	oneway ANOVA + <i>post-hoc test</i>
rheobase (pA)	11.9 ± 9.5	24.5 ± 20.0	57.4 ± 46.5	75.8 ± 37.6	51.7 ± 29.0	Kruskal-Wallis test: p < 0.0001 (K-W + Dunn's multiple comparisons test: P2-5 vs. P10-15: p = 0.8636 P2-5 vs. P20-25: p < 0.0001 P2-5 vs. P50-56: < 0.0001 P2-5 vs. P > 150: < 0.0001 P10-15 vs. P20-25: p = 0.0560 P10-15 vs. P50-56: p = 0.0003 P10-15 vs. P > 150: p = 0.0698 P20-25 vs. P50-56: p > 0.9999 P20-25 vs. P > 150: p > 0.9999 P50-56 vs. P > 150: p > 0.9999
Max gain dAP frequency/ dI_{input} (Hz/pA)	14x10 ⁻³ ± 7x10 ⁻³	7x10 ⁻³ ± 3x10 ⁻³	5x10 ⁻³ ± 1x10 ⁻³	7x10 ⁻³ ± 2x10 ⁻³	6x10 ⁻³ ± 2x10 ⁻³	ANOVA: p < 0.0001 Bonferroni's multiple comparisons test: P2-5 vs. P10-15: p < 0.0627 P2-5 vs. P20-25: p = 0.0001 P2-5 vs. P50-56: p = 0.0818 P2-5 vs. P > 150: p = 0.001 P10-15 vs. P20-25: p > 0.5937 P10-15 vs. P50-56: p > 0.9999 P10-15 vs. P > 150: p > 0.9999 P20-25 vs. P50-56: p = 0.7328 P20-25 vs. P > 150: p > 0.9999 P50-56 vs. P > 150: p > 0.9999
max AP freq. (Hz)	19 ± 14	19 ± 8	29 ± 12	30 ± 7	31 ± 8	Kruskal-Wallis test: p < 0.0001 (K-W + Dunn's multiple comparisons test: P2-5 vs. P10-15: p > 0.9999 P2-5 vs. P20-25: p = 0.1564 P2-5 vs. P50-56: p = 0.0065 P2-5 vs. P > 150: p = 0.0023 P10-15 vs. P20-25: p = 0.0555 P10-15 vs. P50-56: p = 0.0019 P10-15 vs. P > 150: p = 0.0006

						<i>P</i> 20-25 vs. <i>P</i> 50-56: <i>p</i> > 0.9999 <i>P</i> 20-25 vs. <i>P</i> > 150: <i>p</i> > 0.9999 <i>P</i> 50-56 vs. <i>P</i> 150: <i>p</i> > 0.9999
AP threshold (mV)	-30 ± 7	-42 ± 7	-43 ± 7	-42 ± 8	-44 ± 7	Kruskal-Wallis test: <i>p</i> < 0.0001 (K-W + Dunn's multiple comparisons test: <i>P</i> 2-5 vs. <i>P</i> 10-15: <i>p</i> = 0.0005 <i>P</i> 2-5 vs. <i>P</i> 20-25: <i>p</i> < 0.0001 <i>P</i> 2-5 vs. <i>P</i> 50-56: <i>p</i> = 0.0027 <i>P</i> 2-5 vs. <i>P</i> > 150: <i>p</i> < 0.0001 <i>P</i> 10-15 vs. <i>P</i> 20-25: <i>p</i> > 0.9999 <i>P</i> 10-15 vs. <i>P</i> 50-56: > 0.9999 <i>P</i> 10-15 vs. <i>P</i> > 150: > 0.9999 <i>P</i> 20-25 vs. <i>P</i> 50-56: > 0.9999 <i>P</i> 20-25 vs. <i>P</i> > 150: > 0.9999 <i>P</i> 50-56 vs. <i>P</i> 150: > 0.9999
max dV/dT (V/s)	95 ± 41	158 ± 27	254 ± 61	219 ± 44	218 ± 65	Kruskal-Wallis test: <i>p</i> < 0.0001 (K-W + Dunn's multiple comparisons test: <i>P</i> 2-5 vs. <i>P</i> 10-15: <i>p</i> = 0.0937 <i>P</i> 2-5 vs. <i>P</i> 20-25: <i>p</i> < 0.0001 <i>P</i> 2-5 vs. <i>P</i> 50-56: <i>p</i> < 0.0001 <i>P</i> 2-5 vs. <i>P</i> > 150: <i>p</i> < 0.0001 <i>P</i> 10-15 vs. <i>P</i> 20-25: <i>p</i> = 0.0002 <i>P</i> 10-15 vs. <i>P</i> 50-56: <i>p</i> = 0.0184 <i>P</i> 10-15 vs. <i>P</i> > 150: <i>p</i> = 0.0441 <i>P</i> 20-25 vs. <i>P</i> 50-56: <i>p</i> > 0.9999 <i>P</i> 20-25 vs. <i>P</i> > 150: <i>p</i> > 0.9999 <i>P</i> 50-56 vs. <i>P</i> 150: <i>p</i> > 0.9999
min dV/dT (V/s)	-21 ± 11	-31 ± 7	-53 ± 20	-52 ± 7	-61 ± 14	Kruskal-Wallis test: <i>p</i> < 0.0001 (K-W + Dunn's multiple comparisons test: <i>P</i> 2-5 vs. <i>P</i> 10-15: <i>p</i> = 0.0529 <i>P</i> 2-5 vs. <i>P</i> 20-25: <i>p</i> < 0.0001 <i>P</i> 2-5 vs. <i>P</i> 50-56: <i>p</i> < 0.0001 <i>P</i> 2-5 vs. <i>P</i> > 150: <i>p</i> < 0.0001 <i>P</i> 10-15 vs. <i>P</i> 20-25: <i>p</i> < 0.0001 <i>P</i> 10-15 vs. <i>P</i> 50-56: <i>p</i> < 0.0001 <i>P</i> 10-15 vs. <i>P</i> > 150: <i>p</i> < 0.0001 <i>P</i> 20-25 vs. <i>P</i> 50-56: <i>p</i> > 0.9999 <i>P</i> 20-25 vs. <i>P</i> > 150: <i>p</i> = 0.1867 <i>P</i> 50-56 vs. <i>P</i> 150: <i>p</i> = 0.1666

AP half-width (ms)	3.5 ± 1.6	2.3 ± 0.4	1.7 ± 0.5	1.7 ± 0.2	1.4 ± 0.3	Kruskal-Wallis test: p < 0.0001 Dunn's multiple comparisons test:P2-5 vs. P10-15: p > 0.9999 P2-5 vs. P20-25: p < 0.0001 P2-5 vs. P50-56: p = 0.0001 P2-5 vs. P > 150: p < 0.0001 P10-15 vs. P 20-25: p = 0.0036 P10-15 vs. P 50-56: p = 0.0140 P10-15 vs. P > 150: p < 0.0001 P20-25 vs. P 50-56: p > 0.9999 P20-25 vs. P > 150: p = 0.8198 P50-56 vs. P 150: p = 0.3962
Developmental changes in intrinsic membrane properties and input – output gain. Intrinsic membrane properties, maximal input-output gain, rheobase, maximal AP frequency, AP threshold, max dV/dt, min dV/dt, and AP half-width of M1LV neuron are reported as average ± standard deviation. P values for multiple comparison and post-hoc test are reported for each paired comparison. The statistical tests used for normally distributed samples were one-way ANOVA and Bonferroni's multiple comparisons test (A. + Bonferroni). Non-normally distributed samples were compared with the Kruskal-Wallis test and Dunn's multiple comparisons test (K-W + Dunn's).						

Supplementary Table 5.

	P2-5 (n = 24)	P10-15 (n = 21)	P20-25 (n = 21)	P50-56 (n = 19)	P > 150 (n = 20)	oneway ANOVA + <i>post-hoc test</i>
IS (V/s)	100 ± 40	150 ± 31	179 ± 60	180 ± 31	190 ± 47	Kruskal-Wallis test: p < 0.0001 Dunn's multiple comparisons test: P2-5 vs. P10-15: p = 0.1341 P2-5 vs. P20-25: p = 0.0002 P2-5 vs. P50-56: p < 0.0001 P2-5 vs. P > 150: p < 0.0001 P10-15 vs. P20-25: p = 0.5839 P10-15 vs. P50-56: p = 0.2295 P10-15 vs. P > 150: p = 0.0797 P20-25 vs. P50-56: p > 0.9999 P20-25 vs. P > 150: p > 0.9999 P50-56 vs. P > 150: p > 0.9999
SD (V/s)	n.a.	130 ± 31	248 ± 66	250 ± 50	274 ± 52	Kruskal-Wallis test: p < 0.0001 Dunn's multiple comparisons test: P10-15 vs. P20-25: p < 0.0001 P10-15 vs. P50-56: p < 0.0001 P10-15 vs. P > 150: p < 0.0001 P20-25 vs. P50-56: p = 0.9998 P20-25 vs. P > 150: p > 0.4143 P50-56 vs. P > 150: p > 0.4942
Est $I_{ln\text{-}AIS}$ (pA)	126 ± 50	273 ± 58	451 ± 148	490.5 ± 84.98	550.6 ± 136.7	Kruskal-Wallis test: p < 0.0001 Dunn's multiple comparisons test: P2-5 vs. P10-15: p < 0.0001 P2-5 vs. P20-25: p < 0.0001 P2-5 vs. P50-56: p < 0.0001 P2-5 vs. P > 150: p < 0.0001 P10-15 vs. P20-25: p = 0.0009 P10-15 vs. P50-56: p < 0.0001 P10-15 vs. P > 150: p < 0.0001 P20-25 vs. P50-56: p = 0.9778 P20-25 vs. P > 150: p > 0.3143

						P_{50-56} vs. P_{150} : $p > 0.6648$
Developmental changes in IS and SD component. AP IS and SD component, and estimated inward current at the AIS (Est $I_{in\text{-AIS}}$) of M1LV neuron are reported as average \pm standard deviation. P values for multiple comparison and post-hoc test are reported for each paired comparison. These (non-normally distributed) samples were compared with the Kruskal-Wallis test and Dunn's multiple comparisons test (K-W + Dunn's).						

Supplementary Table 6.

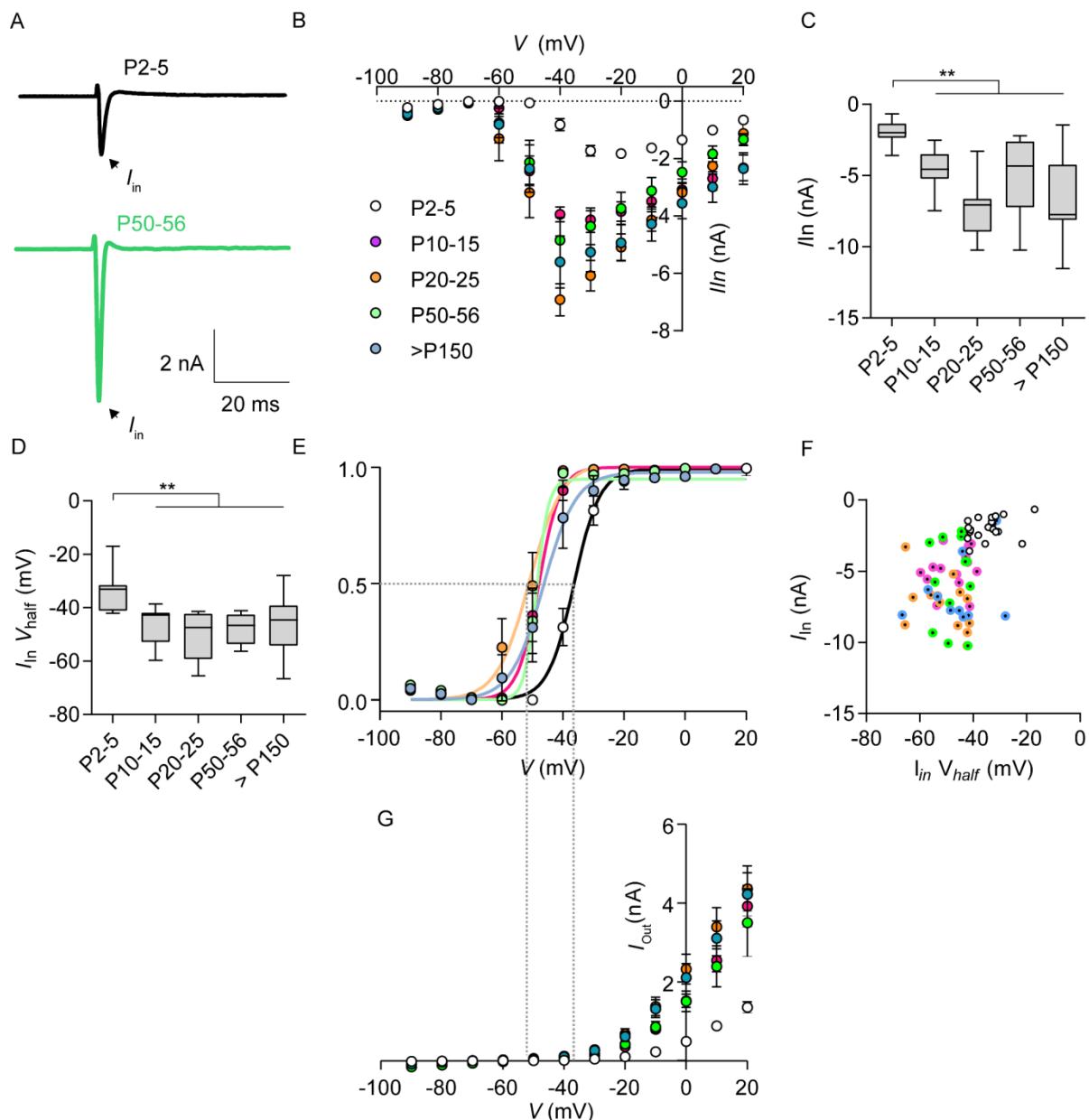
	P2-5 (n _{mono} /n _{tri} = 21/0)	P10-15 (n _{mono} /n _t ri = 19/0)	P20-25 (n _{mono} /n _t ri = 20/7)	P50-56 (n _{mono} /n _t ri = 18/6)	P > 150 (n _{mono} /n _t ri = 16/14)	oneway ANOVA + <i>post-hoc test</i>
AHP_{mono/sl} ow (mV)	19 ± 3	17 ± 3	13 ± 3	11 ± 3	14 ± 4	ANOVA: p < 0.0001 Bonferroni's multiple comparisons test: P2-5 vs. P10- 15: p = 0.2449 P2-5 vs. P20-25: p < 0.0001 P2-5 vs. P50-56: p < 0.0001 P2-5 vs. P > 150: p = 0.0002 P10-15 vs. P20-25: p = 0.0010 P10-15 vs. P50-56: p < 0.0001 P10-15 vs. P > 150: p = 0.2480 P20-25 vs. P50-56: p > 0.9999 P20-25 vs. P > 150: p > 0.9999 P50-56 vs. P > 150: p = 0.0817
AHP_{fast} (mV)	n.a.	n.a.	6.1 ± 2.6	13.2 ± 2.9	9.1 ± 3.7	ANOVA: p < 0.0001 Bonferroni's multiple comparisons test: P20-25 vs. P50-56: p = 0.0025 P20-25 vs. P > 150: p = 0.1773 P50-56 vs. P > 150: p = 0.0619
ADP (mV)	n.a.	n.a.	2.2 ± 1.8	1.8 ± 1.0	3.9 ± 2.8	ANOVA: p < 0.0968 Bonferroni's multiple comparisons test: P20-25 vs. P50-56: p > 0.9999 P20-25 vs. P > 150: p = 0.3324 P50-56 vs. P > 150: p = 0.1771

Developmental changes of AHP and ADP. Average amplitude of AHP_{mono/slow}, AHP_{fast} and ADP of M1LV neurons are reported as average ± standard deviation. P values for multiple comparison and post-hoc test are reported for each paired comparison. Below the table headers indicating age groups, the number of neurons showing mono-phasic events (n_{mono}) and tri-phasic events (n_{tri}) are reported side by side (n_{mono}/n_{tri}). The statistical tests used for comparing these (normally distributed) samples was one-way ANOVA and Bonferroni's multiple comparisons test (A. + Bonferroni).

Supplementary Table 7.

	P2-5 (n = 21)	P10-15 (n = 19)	P20-25 (n = 14)	P50-56 (n = 15)	P > 150 (n = 13)	oneway ANOVA + <i>post-hoc test</i>
I_{in} (nA)	-2.0 ± 0.7	-4.6 ± 1.4	-7.5 ± 1.9	-5.4 ± 2.8	-6.6 ± 2.6	ANOVA: p < 0.0001 Bonferroni's multiple comparisons test: P2-5 vs. P10-15: p = 0.0005 P2-5 vs. P20-25: p < 0.0001 P2-5 vs. P50-56: p < 0.0001 P2-5 vs. P > 150: p < 0.0001 P10-15 vs. P20-25: p = 0.0005 P10-15 vs. P50-56: p = 0.7067 P10-15 vs. P > 150: p = 0.0303 P20-25 vs. P50-56: p = 0.0412 P20-25 vs. P > 150: p = 0.7935 P50-56 vs. P > 150: p = 0.4575
$I_{in} V_{Half}$ (mV)	-34 ± 7	-47 ± 7	-51 ± 9	-48 ± 6	-46 ± 11	Kruskal-Wallis test: p < 0.0001 Dunn's multiple comparisons test: P2-5 vs. P10-15: p = 0.0002 P2-5 vs. P20-25: p < 0.0001 P2-5 vs. P50-56: p = 0.0001 P2-5 vs. P > 150: p = 0.0062 P10-15 vs. P20-25: p > 0.9999 P10-15 vs. P50-56: p > 0.9999 P10-15 vs. P > 150: p > 0.9999 P20-25 vs. P50-56: p > 0.9999 P20-25 vs. P > 150: p > 0.9999 P50-56 vs. P > 150: p > 0.9999
I_{out} (nA)	1.3 ± 0.6	3.9 ± 2.3	4.4 ± 2.0	3.6 ± 3.1	4.0 ± 1.9	Kruskal-Wallis test: p < 0.0001 Dunn's multiple comparisons test: P2-5 vs. P10-15: p < 0.0001 P2-5 vs. P20-25: p < 0.0001 P2-5 vs. P50-56: p = 0.0378 P2-5 vs. P > 150: p < 0.0001 P10-15 vs. P20-25: p > 0.9999

						$P10-15$ vs. $P 50-56$: $p > 0.9999$ $P10-15$ vs. $P > 150$: $p > 0.9999$ $P20-25$ vs. $P 50-56$: $p = 0.4134$ $P20-25$ vs. $P > 150$: $p > 0.9999$ $P50-56$ vs. $P 150$: $p > 0.9999$
Developmental changes in inward and outward voltage-activated currents. Voltage sensitive inward current (I_{in}), voltage of I_{in} half-maximal activation ($I_{in} V_{Half}$), steady state outward current (I_{out}), fast outward current ($I_{out\ fast}$), and $I_{out\ fast}$ decay constant (τ) of M1LV neuron are reported as average \pm standard deviation. P values for multiple comparison and post-hoc test are reported for each paired comparison. The statistical tests used for normally distributed samples were one-way ANOVA and Bonferroni's multiple comparisons test (A. + Bonferroni). Non-normally distributed samples were compared with the Kruskal-Wallis test and Dunn's multiple comparisons test (K-W + Dunn's).						



Supplementary Fig. 1 Development of voltage-activated currents. **A.** Typical inward current (I_{in}) of P2-5 neurons (black) and P50-56 neurons (green) upon depolarization (-70 mV to -40 mV). Peak I_{in} is highlighted by arrowheads. **B.** Current – voltage relation of peak I_{in} elicited by depolarizing steps (500 ms) of increasing voltage (from -90 mV to +20 mV, holding potential: -70 mV). **C.** Maximal amplitude of I_{in} . **D.** Inward current half-maximal activation ($I_{in} V_{half}$). **E.** Fractional activation of I_{in} for different age groups. Note the smaller voltage sensitivity at P2-5. **F.** Relation between amplitude of peak I_{in} and $I_{in} V_{half}$, samples are color coded as in panel B. **G.**

Current – voltage relation of outward currents elicited by depolarizing voltage steps (500 ms) of increasing amplitude (from -90 mV to +20 mV, holding potential: -70 mV). Note reduced amplitude and voltage dependence at P2-5. ** $p < 0.01$; P2-5: n = 21; P10-15: n = 19; P20-25: n = 14; P50-56: n = 15; P > 150: n = 13.