Fetal ethanol exposure and postnatal brain hyperexcitability: “the tip of the neurobehavioural iceberg”.

Peter L. Carlen MD, FRCPC
Departments of Medicine (Neurology), Physiology, and IBBME
University of Toronto
Neurologist, Epilepsy Program
University Health Network, Toronto Western Hospital
Fundamental Neurobiology Division
Toronto Western Research Institute

Fetal Alcohol Syndrome

An understudied epidemic: clinical and neurobiological features

Ancient History

“Foolish, drunken and harebrained women most often bring forth children like unto themselves, morose and languid” – Aristotle

Ancient History

“… You will conceive and live birth to a son.
Now then, drink no wine or other fermented drink …”
Old Testament, Judges 13:7

Modern History

Lemoine P, Harousseau H, Borteyru J, et Menuet J. (1968). Les enfants des parents alcoholiques: anomolies observees a propos de 127 cas. *Quest Medical*. 25, 476-482.

Modern History

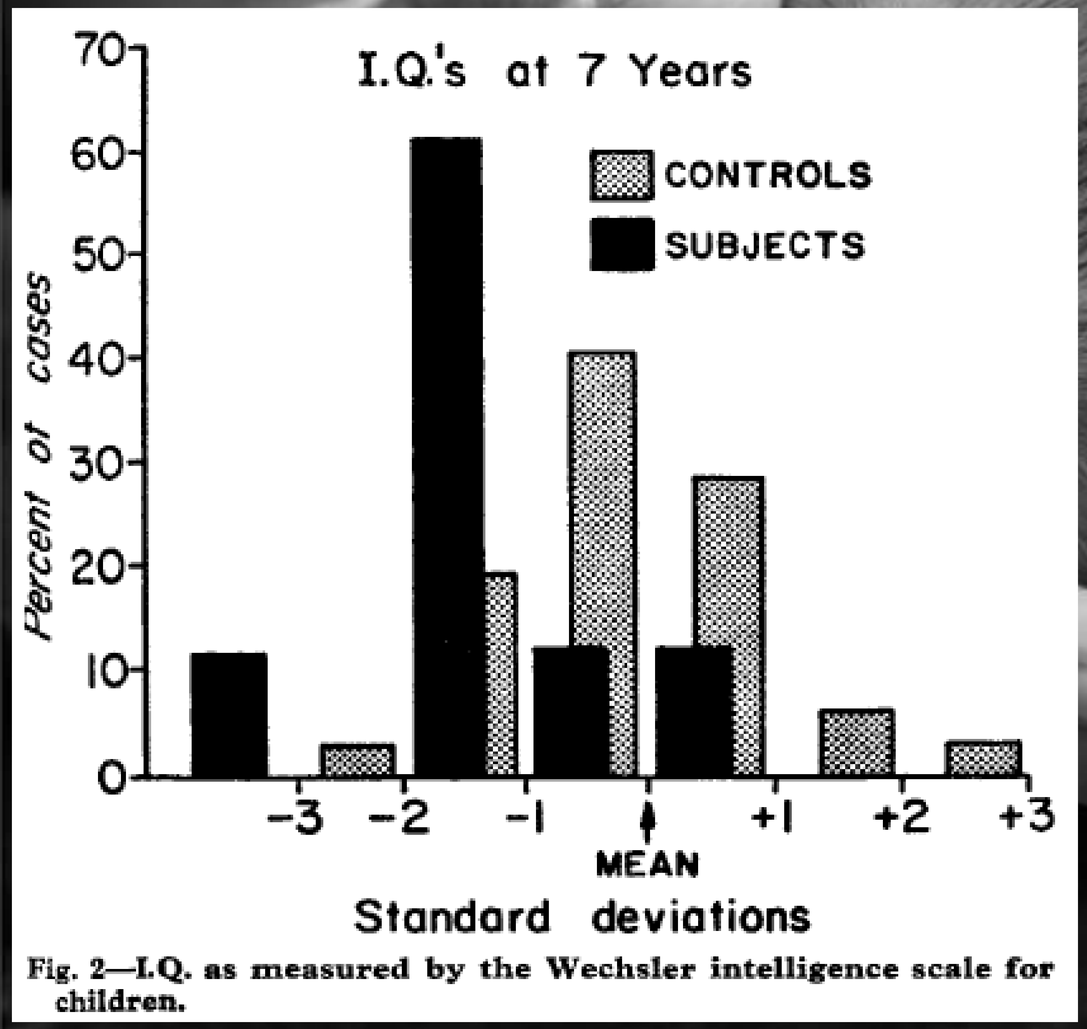

Fig. 2—I.Q. as measured by the Wechsler intelligence scale for children.

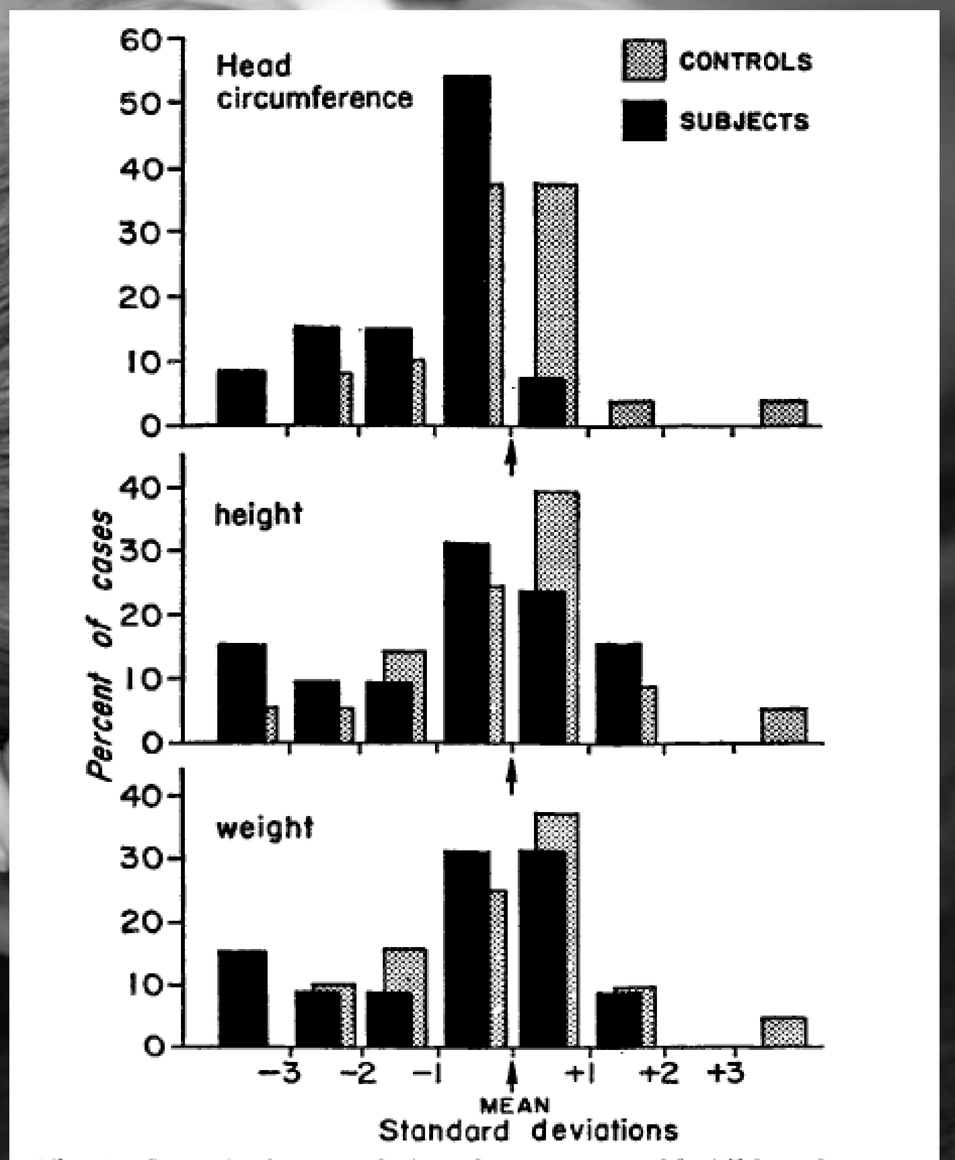

Fig. 4—Growth characteristics of seven-year-old children born to alcoholic mothers as contrasted to those of their matched controls.

Epidemiology

- Down Syndrome: 1 per 1000 births
- Tourette Syndrome: 1 per 1000 births
- Epilepsy: 3 per 1000 births
- Autism: 2 per 1000 births
- **FASD: 10 per 1000 births**
- ADHD: 30 per 1000 births

Epidemiology

- 95% will have mental health problems
- 82% will not be able to live independently
- 70% will have problems with employment
- 68% will have "disrupted school experience"
- 68% will experience trouble with the law
- 55% will be confined in prison, drug or alcohol treatment centre or mental institution
- 52% will exhibit inappropriate sexual behaviour
- 50% of males and 70% of females will have alcohol and drug problems

THE REMARKABLY HIGH PREVALENCE OF EPILEPSY IN SUBJECTS WITH FETAL ALCOHOL SPECTRUM DISORDERS

S. Bell[1], B.S. Stade[3,6,9], J.N. Reynolds[1,2], C. Rasmussen[4], G.P. Andrew[9], P. Hwang[6,7,8], P. Carlen.[5,8]

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Centre for Neuroscience Studies, and Department of [2]Pharmacology & Toxicology, Queen's University, Kingston, ON, [3]Department of Pediatrics, St. Michael's Health Centre, Toronto, ON M5C 2T2, [4]Department of Pediatrics, University of Alberta and Glenrose Rehabilitation Hospital, Edmonton, Alberta,[5]Toronto Western Research Institute, University of Toronto, [6]University Health Network, Toronto, ON, [7]North York General Hospital, [8]University of Toronto Epilepsy Research Centre, [9]The Applied Health Research Centre of the Keenan Research Centre Li Ka Shing Knowledge Institute St. Michael's Hospital

INTRODUCTION

- **Fetal alcohol spectrum disorders (FASD) describe the range of adverse developmental outcomes that occur in offspring as a consequence of maternal drinking during pregnancy.**
- **In Canada, FASD affects approximately 1 in every 100 individuals making it the leading preventable cause of mental deficiency in Canada and the western world. Also an estimated 50% of the prison population are FASD.**
- **Many individuals with FASD have brain injury and life-long intellectual, behavioural, and neurological dysfunction.**
- **Children with FASD may present a range of neurological deficits, including problems with learning, attention, memory, sensory-motor skills, executive function, and sleep disturbances.**
- **Epilepsy is a disorder characterized by spontaneous recurrence of unprovoked seizures, affecting approximately 0.6% of the general population in Canada, with an incidence of 0.5% per annum.**

HYPOTHESES

(1) Individuals with FASD have a higher prevalence of epilepsy and/or seizures than the general population.

(2) Epilepsy and/or seizures are not associated with a specific diagnostic subgroup (ARND, pFAS, or FAS).

(3) A diagnosis of FASD is independent of other risk factors associated with epilepsy and seizures in individuals with FASD.

METHODS

- A retrospective chart review was conducted on all active charts (n=1063) at St. Michael's Hospital (Toronto, Ontario) and Glenrose Rehabilitation Hospital (Edmonton, Alberta) FASD clinics.
- Information was gathered on 425 subjects with a diagnosis of Fetal Alcohol Syndrome (FAS), partial FAS (pFAS) or Alcohol Related Neurodevelopmental Disorder (ARND).
- Information regarding maternal drinking history was extracted including: pattern, magnitude, consumption and type of beverage.
- In subjects with epilepsy or spontaneous seizures, information on type of seizure, age of onset, frequency and treatment was collected.
- Two neurologists reviewed all records of subjects with epilepsy, seizures, or questionable epilepsy/seizures.
- The relationship between FASD diagnosis and other risk factors for co-occurrence of epilepsy (e.g. type of birth, prenatal care, family history, trauma) to the occurrence of seizures in children with prenatal alcohol exposure was examined.
- Chi-square tests and multivariate multinomial logistic regression were performed.

Demographic characteristics.

Characteristics	Subjects (n=425)
Age Mean (SD) Range Toronto Edmonton Age group 2-14 15 +	 15.2(7.6) 2-49 6-22 219(51.5%) 206(48.5%)
Gender, n(%) Male Female	 254(59.8%) 171(40.2%)
Comorbidities ADHD ODD Autism Depression PTSD ROD	276(64.9%) 200(47.1%) 42(9.9%) 8(1.9%) 25(5.9%) 17(4%) 17(4%)

Abbreviations: ADHD (Attention Deficit Hyperactivity Disorder), ODD (Oppositional Defiant Disorder), PTSD (Post-Traumatic Stress Disorder), ROD (Reactive Attachment Disorder).

The prevalence of epilepsy and seizures in the FASD population.

FASD Diagnosis	No Seizures	≥ 1 Seizure Episode(s)	Epilepsy	All Seizures
FAS, N (%)	12(80.0)	3(20.0)	0(0)	3(20.0)
PFAS, N (%)	61(85.9)	7(9.9)	3(4.2)	10(14.1)
ARND, N (%)	277(81.7)	40(11.8)	22(6.5)	62(18.23)
Overall, N (%)	350(82.3)	**50(11.8)**	**25(5.9)**	**75(17.7)**

Maternal Drinking Patterns in Epilepsy and Seizure Subjects.

Trimester of Alcohol Exposure	Epilepsy	≥ 1 Seizure Episode(s)
1st	5	8
2nd	0	1
3rd	0	0
1st and 2nd	0	2
1st and 3rd	1	0
All 3 trimesters	5	14

Information available for 36/75 subjects.

Classification of Seizures (number of subjects per category)

Classification	Epilepsy	≥1 Seizure Episode(s)
Generalized	8	2
Partial	3	2
Complex Partial or Absence	9	22
Febrile	0	4
Unclassified	5	20
Total	**25**	**50**

SUMMARY and DISCUSSION

- **These results indicate that there is a remarkably high prevalence of epilepsy in the FASD population of two specialized FASD clinics compared to that of the general population.**
- **There was a high frequency of heavy drinkers among multi-drug users. Most of these women drank heavily (5 or more drinks) regularly, throughout pregnancy.**
- **In the group of individuals with an unnatural type of birth, five of the cases were emergency cesarean sections, and there were four cases of respiratory problems, which may have contributed to neonatal hypoxic brain damage and subsequent seizures.**
- **Future studies, which must be prospective, should be aimed at identifying the types of epilepsy, the roles of other risk factors and the underlying brain mechanisms responsible for the high prevalence of epilepsy and seizures in FASD.**

IMPLICATIONS

- **Indicates a need to screen individuals with FASD for epilepsy, and to screen individuals with epilepsy for FASD.**
- **Prevention and early treatment of seizures.**
- **Future studies are needed to understand the brain mechanisms that link the effects of prenatal alcohol exposure and a reduced seizure threshold.**
- **Could the seizures be a manifestation of underlying hyperexcitable brain circuitry in FASD subjects, even those without a history of seizures, which could predispose to ADD and other psychopathologies?**

Alcohol drinking pattern during pregnancy and risk of infant mortality.

Strandberg-Larsen K, Grønboek M, Andersen AM, Andersen PK, Olsen J. Epidemiology. 2009 Nov;20(6):884-91.

- The association of maternal average alcohol intake and binge drinking (>or=5 drinks per sitting) with infant mortality, both in the neonatal and postneonatal period was examined.
- METHODS: Participants were 79,216 mothers who were enrolled in the Danish National Birth Cohort in 1996-2002, gave birth to a live-born singleton, and provided information while they were pregnant on alcohol consumption during pregnancy. Information on infant mortality and causes of death was obtained from national registries and medical records.
- RESULTS: During the first year of life, 279 children (0.35%) died, 204 during the neonatal period. Infant mortality was not associated with alcohol drinking, even at a consumption level of either 4+ drinks per week or 3+ occasions of binge drinking. **When restricting analyses to term births, both infant mortality and postneonatal mortality were associated with a weekly average intake of 4+ drinks or 3+ binge episodes during the 11th to 16th weeks of gestation (1st trimester).**

Binge drinking during pregnancy and risk of seizures in childhood: a study based on the Danish National Birth Cohort.

Sun Y, Strandberg Larsen K, Vestergaard M, Christensen J, Nybo Andersen AM, Grønbaek M, Olsen J. Am J Epidemiol. 2009 Feb 1;169(3):313-22.

- A population-based cohort study of 80,526 liveborn singletons was done using the Danish National Birth Cohort (1996-2002).
- maternal binge drinking was defined as intake of > or = 5 drinks on a single occasion
- Children were followed for up to 8 years.
- Results showed that exposure to binge drinking episodes during pregnancy was **not** associated with an increased risk of seizure disorders in children, except for those exposed at 11-16 gestational weeks.
- These children had a **3.15-fold increased risk of neonatal seizures** (95% confidence interval: 1.37, 7.25) and a **1.81-fold increased risk of epilepsy** (95% confidence interval: 1.13, 2.90).

HIPPOCAMPAL EXCITABILITY IN YOUNG POSTNATAL GUINEA PIG FOLLOWING PRENATAL ALCOHOL EXPOSURE

B. Sutarjono; C. Florez; J.F. Brien; J.N. Reynolds; B. Kapur;
P.L. Carlen.

- Chronic prenatal ethanol exposure (CPEE) is often associated with persistent deficits in learning and memory in postnatal offspring.
- Timed pregnant guinea pigs consumed *ad libitum* either water or aqueous ethanol solution
- Hippocampal brain slices (350 µm) were prepared from postnatal offspring 21-23. Extracellular recordings of the field excitatory postsynaptic potentials (fEPSPs) were obtained in the CA1 stratum radium of the hippocampus. Intracellular recordings from CA1 pyramidal neurons were also performed

Extracellular Recordings

Epileptiform Acitivity Summary

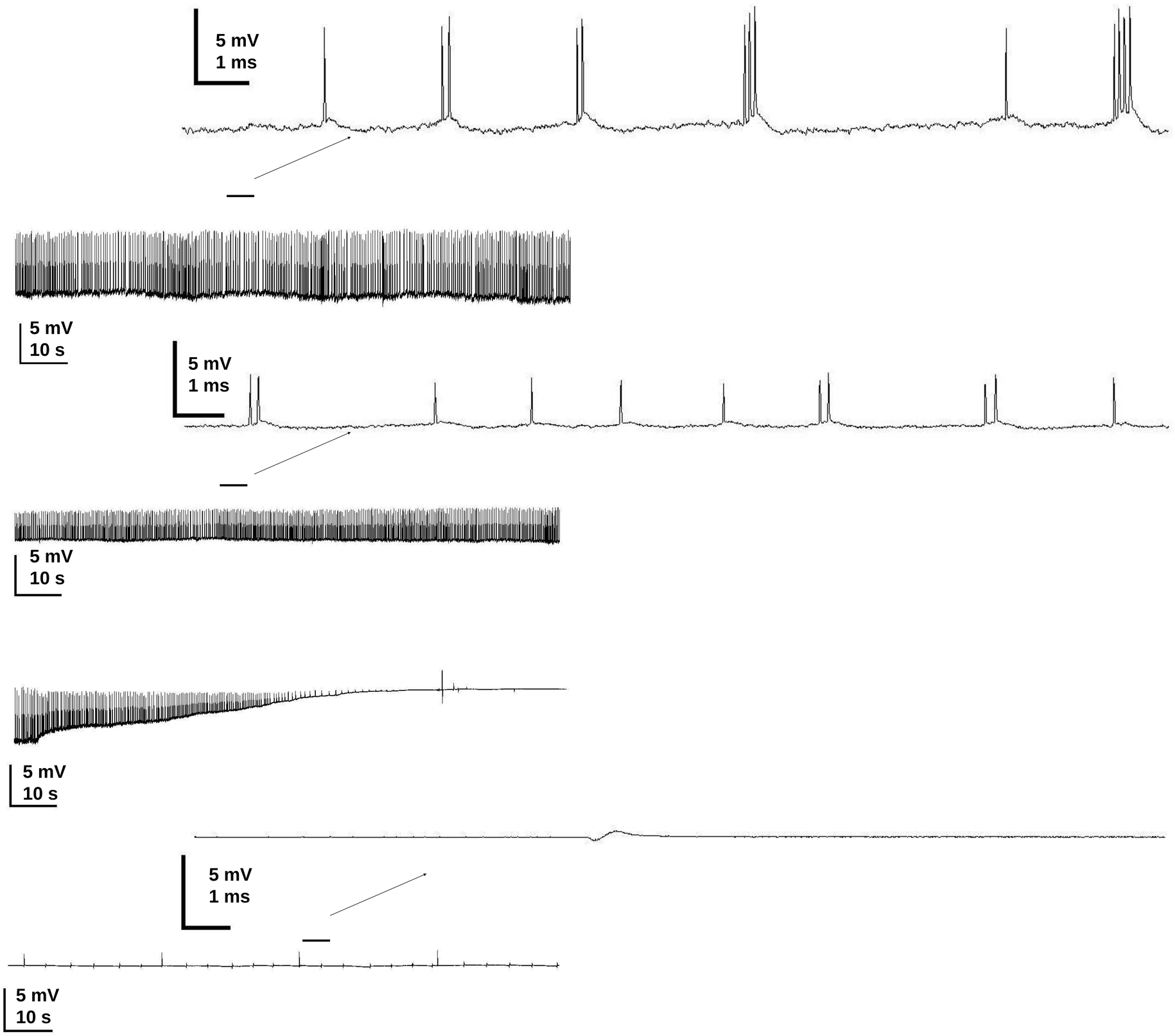
	Control	2nd and 3rd trimesters	1st and 2nd trimesters	1st, 2nd and 3rd trimesters
Number of guinea pigs	19	10	8	24
Epileptiform Activity/Sharp wave ripples	0	2	7	24
Average	0	20.0%	87.5%	100.0%
Number of slices	83	33	29	75
Exhibiting spontaneous spiking activity	16	11	22	66
Average	19.3%	33.3%	75.9%	88.5%

A **Pretreatment**

B Chemical synaptic transmission blockers, **10 uM CNQX + 60 uM APV + 10 uM Gabazine,** have little effect

C Gap junctional blocker: **300 uM Octanol (after 18 min),** blocks the epileptiform activity

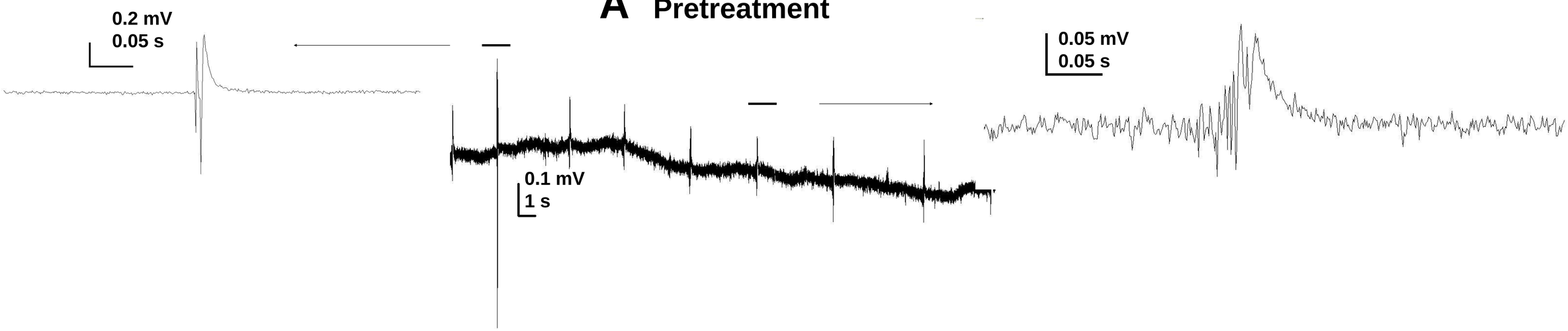
D **Washout (30 min)**



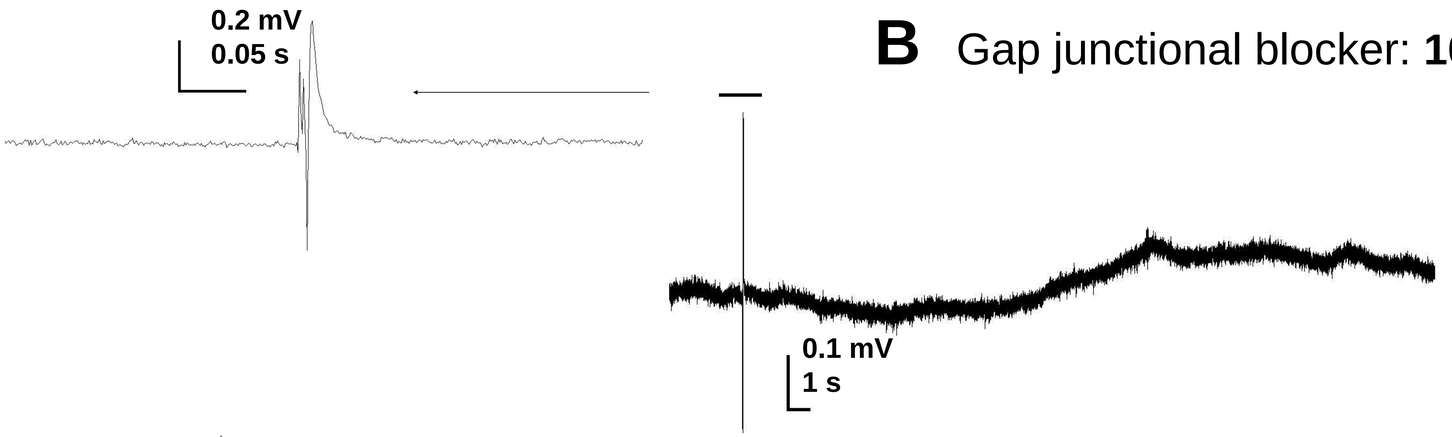
Evoked Response

Spontaneous Activity

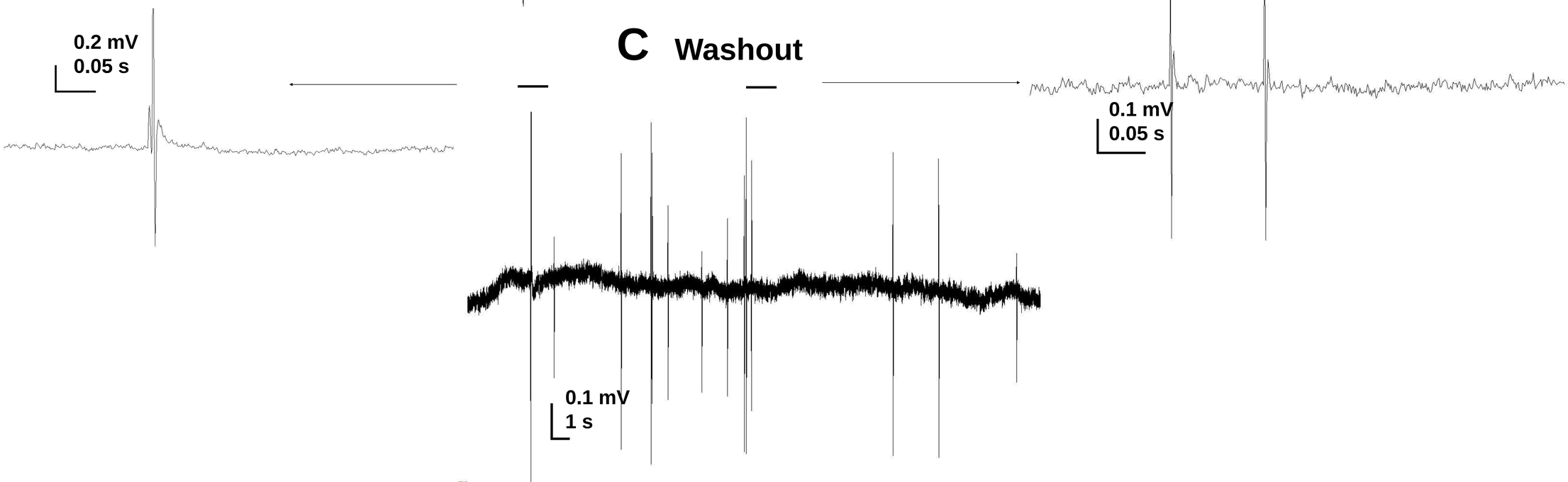
A **Pretreatment**



B Gap junctional blocker: **100 uM Carbenoxolone**



C **Washout**

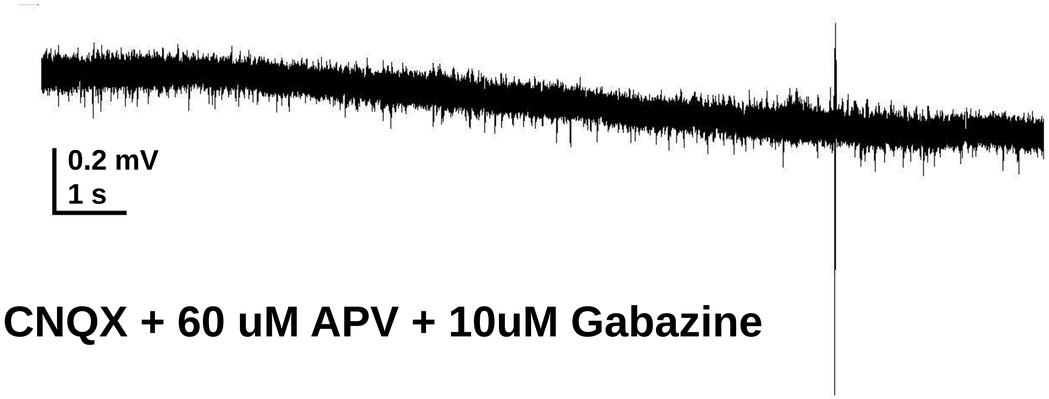


Extra- and Intracellular Recordings (Submerged Slices)

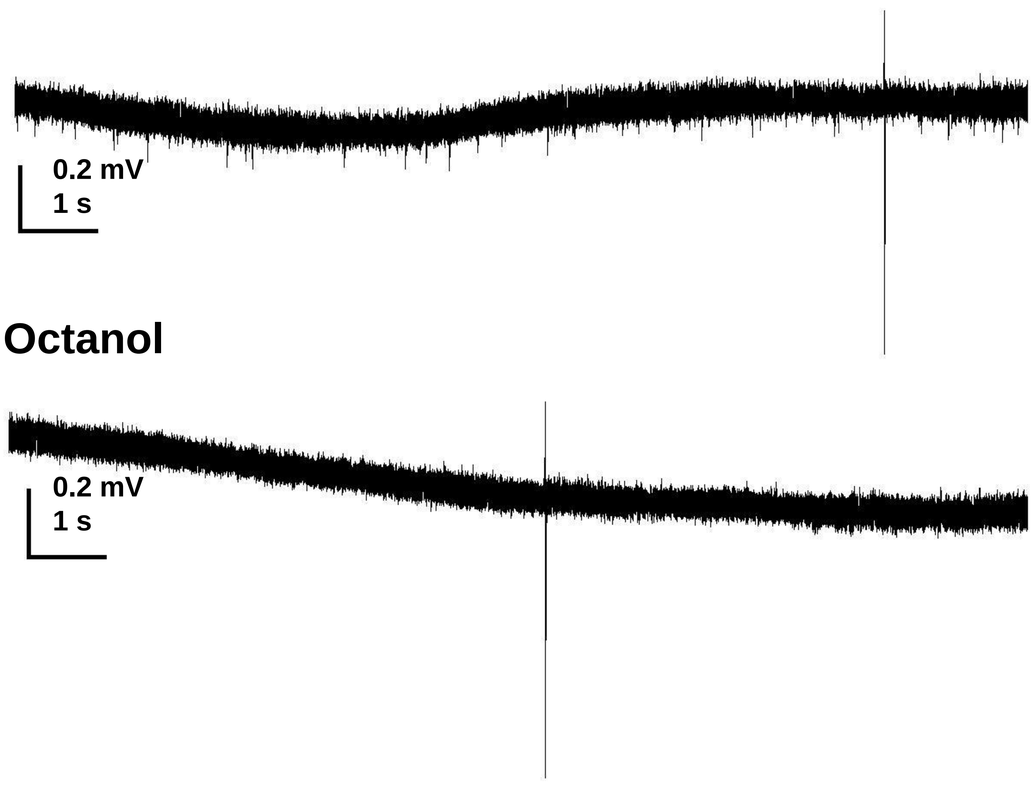
A Pretreatment

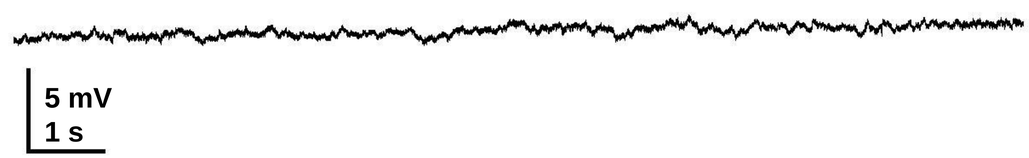
Extracellular

Intracellular

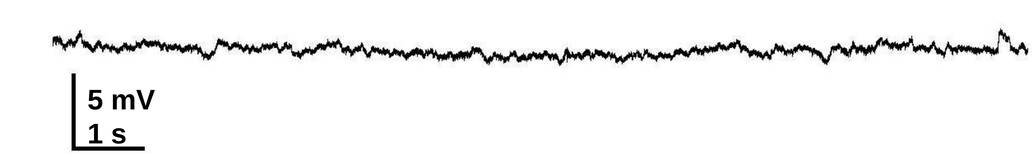


B 10 uM CNQX + 60 uM APV + 10uM Gabazine

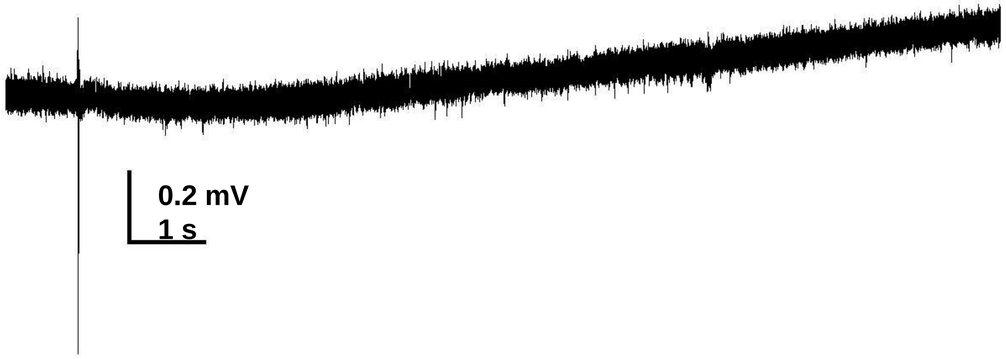


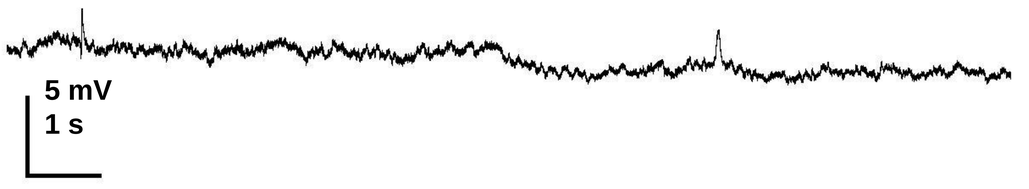


C 300 uM Octanol



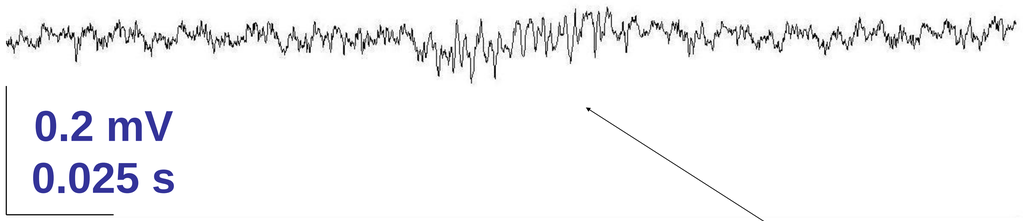
D Washout





C

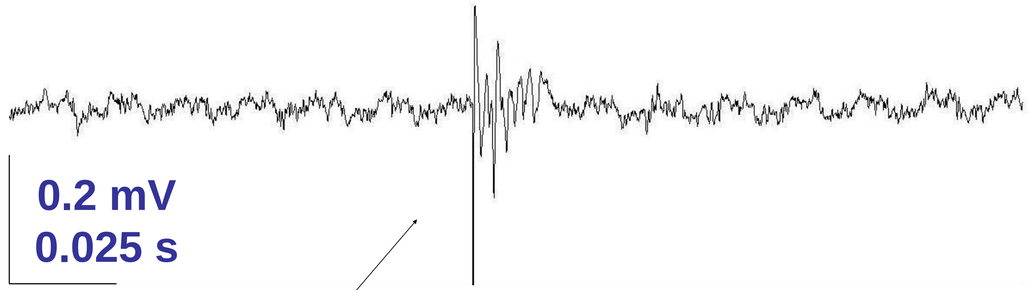
spontaneous



0.2 mV
0.025 s

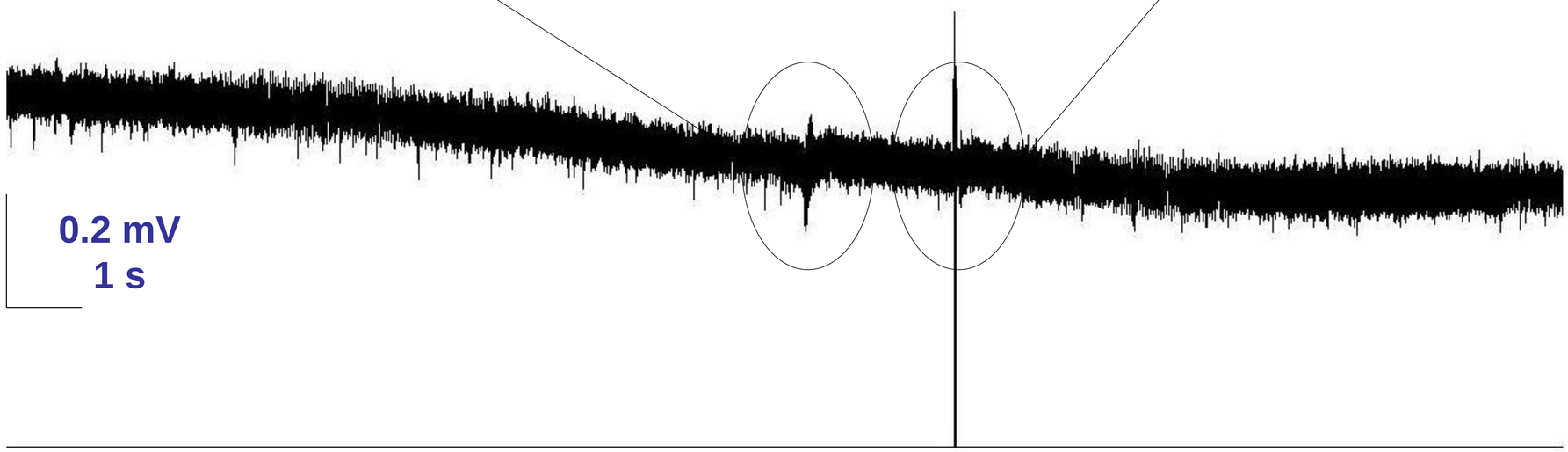
D

evoked



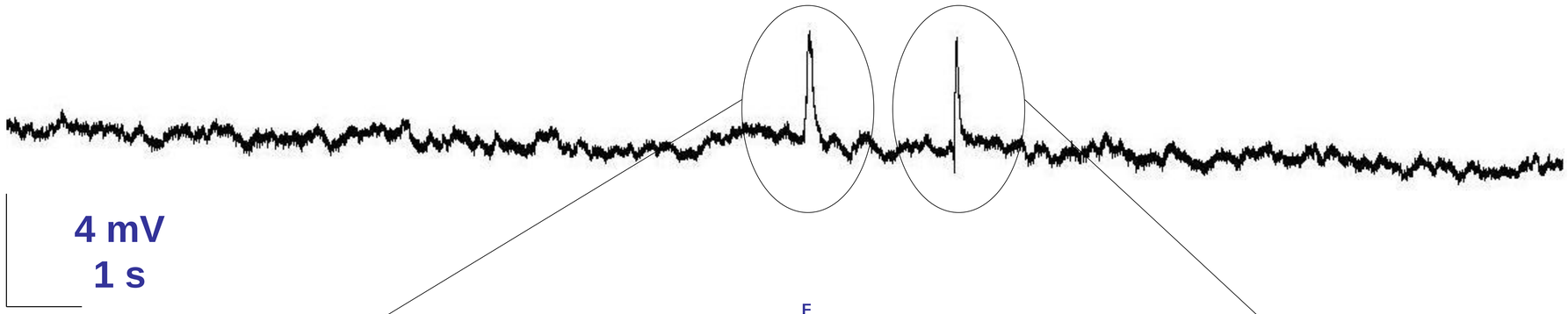
0.2 mV
0.025 s

A



0.2 mV
1 s

B



4 mV
1 s

E

4 mV
0.025 s

F

4 mV
0.025 s

Summary

- Guinea pigs prenatally exposed to 5% EtOH exhibit epileptiform activity in the CA1 of the hippocampus if exposed at least in the first trimester. Second and 3rd trimester exposure without 1st trimester exposure to ethanol did not create the brain hyperexcitability.
- Chemical synaptic transmission blockade (10 uM CNQX, 60 uM APV, and 10 uM gabazine) did **not** stop the epileptiform activity (n = 25)
- The gap junctional blockers (300 uM octanol, n = 9; 100 uM carbenoxolone, n = 5) stopped the epileptiform activity.

- <not shown>
  - Epileptiform activity present in fetuses
  - Epileptiform activity present in adult guinea pigs

CONCLUSIONS

•Brain tissue of young guinea pigs exposed to ethanol, at least in the first trimester, is abnormally hyperexcitable for at least a few months after birth.

•This brain hyperexcitability is related to enhanced gap junctional mechanisms and is not dependent on chemical synaptic transmission.

•Hence altered gap junctional communication and expression have to be investigated in this animal model. Could interfering with gap junctional communication be a therapeutic strategy for FASD?

•First trimester exposure to ethanol appears to be the most dangerous for causing the epileptiform manifestations of the FASD.

•Epigenetic mechanisms are proposed to underly the above findings related to early prenatal ethanol exposure.

•Could this brain hyperexcitability underly the many neurobehavioural abnormalities of the FASD?

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