Histo-neuro-behavioral effects of a single, very mild trauma on senescent rodent brains: a systematic review

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Abstract

Post-concussion syndrome, recently recognized as a complication of mild traumatic brain injury, is considered a consequence of the summative effect of multiple concussions received over lifetime. In elderlies, the main mild brain trauma mechanism is fall (low impact force). Many falls are often not reported or noticed but may generate serious medical and medico-legal consequences.

Our research question was to find if a single, very mild brain trauma can induce neuro-behavioral consequences in elderlies. One database was queried (PubMed – MeSH terminology) looking for histopathological, neuro-cognitive and behavioral changes that can be generated by sub-concussional trauma in senescent rodents, in comparison with young animals.

41 published research articles were selected. 17 of them used very mild brain trauma in young and senescent animals, in the same experiment (6 rats and 11 mice). 24 articles evaluated the effect of sub-threshold brain trauma in adult animals (no control group). Five trauma models were used (blast models were excluded). Neuro-inflammatory changes were detected immediate after very mild primary impact. In young animals, observed pathology disappeared fast (after 3 to 7 days). Increased apoptosis, mild axonal injury in white matter tracts plus maladaptive astrogliosis and microglial activation was stronger in aged animals, persisted over time (8 months) and significantly altered animals’ cognition and behavior. Associated preexisting pathology (hypertension, tau protein deposits, microbleeds, reactive inflammation) was often responsible for amplification of the primary impact results.

As translation of observation is the weak spot of pathology and behavior animal research, further investigation is needed before to conclude that even a single, very mild brain trauma may have medical consequences on human senescent brain.

Keywords:

Very mild Traumatic Brain Injury, Animal models (rodents), Post-concussion syndrome, neuro-behavioral changes, “inflammaging”, brain apoptosis
Introduction

Traumatic brain injury (TBI) is a common medical condition that may lead to long-term disability or death. Often seen in clinical practice, the complex, heterogeneous consequences of TBI are still a subject of research and debate [1]. TBI is labeled in many ways. The most popular classification system is the Glasgow Coma Scale (GCS). Under it, the broad spectrum of brain trauma pathology is classified as mild, moderate and severe. Mild TBI (mTBI), the least severe form of head trauma, represents more than 80% of the cases. Most of the patients with mTBI have a favourable evolution with full, rapid recovery. A minority of patients (10-20%) will develop prolonged neurocognitive and behavioral changes [2]. For these patients, neurological, cognitive, emotional and behavioral changes may organize in time, under the label of various chronic neuropsychiatric conditions [3]. Many factors may dictate the heterogeneous clinical response after mTBI: age at the moment of impact [4], type and intensity of trauma, existence of associated pathology [5] or the number of hits over lifetime [6].

mTBI has a recognized bimodal distribution. Young (bellow 21 years) and old people (65 years and above) are more frequently affected [7]. When effects of mTBI are compared in the two age groups, elderlies (65 and over) have a worse immediate clinical outcome, a higher degree of medical resource utilization (and medical related costs), an increased higher risk for progressive cognitive decline in time and a reduced response to medical rehabilitation [8,9]. In geriatric patients, fall is the recognized cause of the mTBI. 35% of old people (65 and over) living alone, in long term care facilities or in assisted living housing fall at least once per year. This percentage increase to 50%- 60% for the group 80 years and over [10]. These numbers can be
even higher as not all fall victims are medically evaluated if the trauma is very mild (sub-concussion), if the fall is unwitnessed and not mentioned to the caregivers or family [11].

Beside immediate and medium-term complications after mTBI, in elderly, a progression to chronic traumatic encephalopathy (CTE) [12], Alzheimer Disease (AD) [13] or Parkinson disease (PD) [14] was described, frequently in association with pre-existing pathology [15].

CTE (sometimes named post-concussion syndrome) was a subject of medical controversy for more than 130 years [16]. Only recently CTE was recognized as one of major chronic neuro-psychologic complication of mTBI [17]. CTE is considered now a spectrum disease [18] where progressive brain atrophy is associated with progressive cognitive decline. Initially was linked with professional exposure (military personnel or professional athletes) to multiple mild head injuries over a lifetime [19]. It is widely accepted now that CTE may appear in geriatric population without history of repeated head trauma [20,21].

We hypothesized that a single sub-concussive trauma (without any loss of conscience, GCS =15), in specific conditions (old, frail geriatric patients with associated pathology) may contribute to CTE development that often remains undetected but may have serious clinical and medico-legal consequences.

In order to evaluate the importance of sub-concussive impact on aged brain, we reviewed published research data reporting induced mTBI on senescent animal (rodents). There are limitations in translating results from animals to humans but it is widely accepted that adequate rodent models may mimic human mTBI and will allow alignment of the mechanical impact with specific, measurable neuronal and behavioral consequences [22].
Method

Our research was performed under PRISMA recommendations [23]. We evaluated one publication database (PubMed) using a Boolean research strategy based on the following MeSH terminology: (((animals) AND (mice) OR (rats) OR (rodents)) AND (aging) AND ((mild traumatic brain injury) OR (concussion)) AND (head injury, closed))). 329 publications were initially found. 125 articles were eliminated as referring to humans, sport, blast injuries, military, treatment, paediatrics, protective medication or review articles. 204 articles (75 of them evaluating mice, 98 rats, 2 mice and rats and 30 rodents) were reviewed based on abstract. Finally, 41 articles were selected as reflecting the neurobehavioral consequences of a single, sub-concussive or mild brain trauma in adult or aged rodents. Articles were finally divided, based on published content, in main studies (responding to our research question) and associated studies (partially responding to our research question).

Results

Based on our specific selection criteria (single, mild brain trauma in senescent animals compared with a group of young animals) 17 research papers were selected by both investigators in the main review arm. 6 of them used a rat experimental model: 5 articles reported results after the use of lateral or medial fluid percussion (FP) (single trauma, 1-2 atm, craniotomy positive). 1 article reported closed head injury using weight drop (table 1). 5 evaluated histologic brain changes. In 5 articles, neuro-behavioral changes were analyzed as early as after 7 days after the impact.

11 articles evaluated mTBI in senescent mice vs young animals. 7 of these used controlled cortical impact (CCI) (craniotomy positive, 3-6 m/s, under 1 mm brain tissue deformation for a
time between 50-400ms) and 3 the closed head injury model (CHI) (craniotomy negative, CHIMERA model in 2 cases and weight drop in one case). One article evaluated the consequences of laser produced cerebral trauma in aged animals. 9 articles evaluated neurobehavioral consequences usually 1 month after the primary impact. Longest survival time was 8 months after the primary injury and reported long lasting cognitive-behavioral consequences (table 2).

24 articles were considered associated studies as research was performed on adult rodents and no young control group was described (table 3). 7 reports evaluated mTBI in rats and 17 and in mice. Force varied from extremely mild (under 1 atm, less than 5 m/s or weight under 15 grms in mice, under 450 grms in rats) to moderate (under 2 atm, more than 5 m/s or weight of more than 15 grms). Longest survival time was 24 months after the primary impact (reflecting aging after single mTBI and showing cognitive and behavioral deficits). One article was selected as it evaluated the structure of the normal aging in young and adult brain (cortical structure and ventricular volumes) comparing cortex and ventricular dimensions using MRI and histology measurements.

Discussion

Mild brain trauma is not a trivial disease. The notion of mild is referring only to the intensity of traumatic event and the immediate clinical picture. mTBI has an uncomplicated evolution in more than 80% of cases. In a minority of patients, evolution can be complicated by long-lasting neurobehavioral symptomatology [24]. In rare cases, chronic invalidating complications may develop. Between these complications, CTE was recently included. CTE was initially described
as an occupational injury (military or sport related) of young people that suffered repeated mild head trauma over their lifetime. It was demonstrated that mTBI complications can affect seniors as well, usually in relation with same-level (low intensity) falls. In elderslies, the central dogma of CTE (summation of multiple mild hits) is under scrutiny as it was observed that similar histopathological picture (neuronal tissue chronic alteration) may develop even after one single mild brain trauma [25]. It is a recognized connection between mTBI and dementia [26] but early phases of dementia (mild cognitive impairment) are difficult to diagnose and to tie causally with mTBI and falls [27].

Our research hypothesis is that a single, sub-concussive head trauma may generate specific CTE histopathological and neuro-behavioral changes in senescent subjects, when pre-existing pathology exists.

We evaluated published research data in order to find histopathological and neuro-behavioral effects of a single, very mild (sub-concussive) head trauma in aged rodents in comparison with the neuro-behavioral picture generated by similar forces in young animals. The difficulty of this review study resides in accommodating and analyzing data coming from a large number of impact models (five), a well-recognized challenge that reflects the complexity of the head trauma pathology research. We deliberately eliminated the blast mechanism as not frequently seen in geriatric populations.

There are several ways to produce brain trauma in rodents. The open-head injury model (craniotomy+) includes FP model (median or lateral) and CCI model. The external force (fluid or mechanical) is applied against intact dura. The impact is a combination of speed, depth and time. The closed-head injury (CHI) models use either gravity (classical weight drop – “Marmarou”
model) or an external mechanical force impacting the intact skull. In most models, head is blocked in a stereotaxic frame. One external closed head mechanical model (Closed-Head Impact Model of Engineered Rotational Acceleration - CHIMERA) allows a free movement of the head in the moment of the impact.

In closed head models, the intensity of head injury is judged based on general reported outcomes like well-being of the animal (blood pressure, spontaneous respirations or recovery of free movements) or recognized measurables like the righting reflex [28]. All animals that had an open-skull surgery were allowed to recover after trauma. Animals age was judged based on published average lifespan (36 months for rats and 24 months for mice) [29].

Three rat studies evaluated inflammatory and apoptotic gene expressions, early after primary impact (PMID: 24385964, 23238576, 31039431). All senescent mice studies performed extensive neuronal tissue inflammatory biomarkers evaluation (chemokines, cytokines) and gene expression in support of histopathological observation (immunohistology). In young animals, after mild trauma proinflammatory cytokines had an early increase followed by rapid decrease. In aged animals, same proteins increased slower and did not decreased even after one month after the primary impact. Specific markers (p16inka) doubled in aged animals after trauma; p21cip1a increased in old animals but not in relation with mTBI (30904769). Other apoptotic related proteins (hippocalcin, LANP, Heat Shock Protein 27) were detected significantly increased in aged brains in precise areas (dentate gyrus) after very mild trauma (32290848).

In young/adult rodents, very mild trauma generated an acute reactive astrogliosis and activated microglia in cortex and evidence of axonal injury in the corpus callosum. Changes were
discovered in specific brain regions and were proportional with trauma intensity (22245525, 21704658, 17174280). Transitory neuro-behavioral changes were reported in adult rodents, mainly in relation with very mild trauma, in response to white matter (axonal) alterations (29993324, 24550885 and 29376093). Long-term cognitive and behavioral changes were observed after 4 weeks after mild to moderate injuries (26774527, 28910378).

In senescent animals, 15 studies reported similar histopathological changes. Increased apoptosis and mild axonal injury were associated with maladaptive astrogliosis and microglial activation. Edema and neuronal tissular destruction were significantly stronger in aged animal groups and persisted over time (8 months). Several studies (22952778, 32290848, 30904769) reported similar microglia senescent changes and increased, exacerbated secondary neuro-inflammatory response (“inflamming”). Trauma intensity was responsible for the location of lesions: very mild trauma affected only hippocampus and corpus callosum of aged animals (2328576, 1335138). 30486287 reported an increased complement activation with specific local lesion evolution (lesion peaked and associated cavitation increased), permanent hippocampus involvement and persistent neuronal cognitive and behavioral changes in aged mice.

12 studies evaluated neuronal (open field, rotarod and balance beam), cognitive (passive avoidance, mazes, novel objects avoidance) and behavioral changes (social, memory). Specific hippocampal inflammation and white matter destruction were associated with alteration of cognitive and executive functions (30904769, 29848996 and 22952778). 30998995 evaluated executive function in adult rats after mTBI and observed that age and not traumatic intensity was responsible for long term changes (12 months).
Several studies evaluated the consequences of mild trauma in specific circumstances. 29269117 observed “lifelong” neuro-behavioral consequences in mutated mice (APP/PS KI) after very mild brain trauma. Using similar mutated APP/PS KI mice, 25904805 reported a delayed but persistent inflammatory response that involved both astrocytes and microglia and generated very long-lasting cognitive deficits (3 months) after trauma. 31262044 reported destruction of blood brain barrier and persistent neuro-inflammatory response after very mild trauma in spontaneous hypertensive rats, also associated with chronic cognitive impairment. 23953759, using a different open-skull injury technique (laser injuries in young, adult and aged mice) observed significant maladaptive response of microglia (increased soma and reduced processes) after injuries in senescent animals. 29808778 showed metabolic long-time changes in aged injured brain that worsen in time. Study 32641073 reported an increased number of microbleeds, cortical thinning and increased ventricular volume in aged, non-injured animals. 31039431 reported that, in aged animals, an associated external trauma (limb fractures) may exacerbate neuroinflammatory response.

The complexity of brain trauma mechanism is well recognized. Closed head injuries alter brain tissue in a two steps mechanism. The primary impact will produce direct (primary) brain tissue destruction that will be followed by a cascade of pathophysiologic and neurochemical (secondary) events that can be highly variable among patients [29]. The association between neuronal death (produced either by apoptosis or necrosis), neuro-inflammation and white matter, axonal alterations in different brain areas is probably related with neuro-behavioral changes observed in humans and animals after mild brain trauma.
Most of the brain trauma studies performed on animal models used young or adult subjects and evaluated the consequences of mild or moderate impacts. It was not clear if a single, very mild trauma (sub-concussive) can persistently alter cognition and behavior in aged animals. Our literature research revealed that a single, sub-concussional trauma will induce only minimal, transitory changes in young or adult animals. These lesions can also “prime” the brain for further traumatic events allowing CTE development as a summation of trauma over time. In senescent animals, maladaptive neuro-inflammatory response with increased sensibility to apoptosis, a weakened blood brain barrier, an increased propensity to brain edema formation, the presence of microbleeds and altered local metabolism with increased reactive oxygen species generation are responsible for the development of a serious histopathologic picture associated with long term neuro-cognitive consequences.

Our study has several limitations. Our literature review was performed on only one, English speaking, database. The low number of true long-lasting senescent rodent studies was perceived as a serious limitation. There are several models that are currently used for modelling consequences of mTBI in rodents, reflecting both the complexity of the brain trauma and specific research interests. As there are not accepted guidelines for mTBI research design, there are not many similitudes between research centers. In our review, we did not separate mTBI consequences based on subjects’ sex but in aged humans there were reported serious differences. Translation of the results from rodents to humans is a well-recognized basic science challenge, mainly when cognition and behavior are evaluated [30].

In conclusion, apoptosis and inflammation were contributing factors for neuro-cognitive alterations discovered after very mild brain trauma in rodent models. Chronic inflammation
status (“inflammaging”), coexistence of peripheric traumatic pathology, the summative effects of repeated head trauma, abnormal brain protein deposits and associated cardio-vascular pathology were aggravating factors. In senescent animals, neuro-cognitive changes were observed long time after the primary mild impact. As translation of research results from animal models to humans is challenging, further studies are needed before to conclude that very mild brain trauma may be involved in the development of mild cognitive impairment in senescent patients.

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Table 1

<table>
<thead>
<tr>
<th>PMID</th>
<th>1st Author, year, Reference</th>
<th>Age_1</th>
<th>Age_2</th>
<th>Method</th>
<th>No. Hits</th>
<th>Survival</th>
<th>Cranio tomy</th>
<th>Histo (*)</th>
<th>Neuro</th>
<th>Behave</th>
<th>Obs.</th>
</tr>
</thead>
<tbody>
<tr>
<td>24385964</td>
<td>Sun_D, 2013 [31]</td>
<td>1 mo</td>
<td>24 mo</td>
<td>LFP 2 atm</td>
<td>1</td>
<td>2 d</td>
<td>Yes</td>
<td>Yes, proteomics + Dentate gyrus</td>
<td>NA</td>
<td>NA</td>
<td>Increased apoptosis in aged group,</td>
</tr>
<tr>
<td>23238576</td>
<td>Titus 2013 [32]</td>
<td>3 mo</td>
<td>19 mo</td>
<td>LFP mild 1.5 and moderate 1.9 atm</td>
<td>1</td>
<td>1-3 d</td>
<td>Yes</td>
<td>Yes, proteomics + hippocamp</td>
<td>NA</td>
<td>Yes</td>
<td>mTBI lowered cAMP in aged animals hippocamp, proportional cu impact (low impact affects just aged, High impact, both groups)</td>
</tr>
<tr>
<td>31039431</td>
<td>Sun_M 2018 [33]</td>
<td>3 mo</td>
<td>12 mo</td>
<td>LFP 2 atm</td>
<td>1</td>
<td>7 d</td>
<td>Yes</td>
<td>Yes hippocamp MRI</td>
<td>Yes</td>
<td>Yes</td>
<td>Bone fracture may exacerbate brain inflammation worse behavioral</td>
</tr>
<tr>
<td>27449121</td>
<td>Rowe, 2016 [34]</td>
<td>Rats from &lt;1 mo to 6 mo</td>
<td>6 mo</td>
<td>MidFP 1.5-2 atm (age dependent)</td>
<td>1</td>
<td>6 mo max</td>
<td>Yes</td>
<td>NA</td>
<td>Yes</td>
<td>No depression, anxiety if mTBI in adulthood</td>
<td>5 groups, aim was to evaluate age at injury and aging with mTBI using acute and chronic behavior batteries</td>
</tr>
<tr>
<td>22374222</td>
<td>Itoh, 2013 [35]</td>
<td>2 1/2 mo</td>
<td>24 mo</td>
<td>CHI</td>
<td>1</td>
<td>1,3,7 d</td>
<td>No</td>
<td>Yes</td>
<td>NA</td>
<td>Yes</td>
<td>Cognitive Impaired in old free radicals increased in aged group</td>
</tr>
<tr>
<td>1335138</td>
<td>Hamm, 1993 [36]</td>
<td>3 mo</td>
<td>20 mo</td>
<td>LFP, medium 1.7-1.8 atm</td>
<td>1</td>
<td>5 d motor, 12 d cognitive</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Selective cognitive deficits hippocampus related. No motor or cognitive in young group</td>
</tr>
</tbody>
</table>

Table 1. Single mild brain trauma in senescent rats. (*): axonal destruction, apoptosis, activation of astrogliaosis and microgliosis, inflammation, LFP: lateral fluid percussion, MidFP: midline fluid percussion, CHI: closed head injury, d: days, mo: months. Histo: histology; Neuro: neurologic; Behave: Behavioral
Table 2.

<table>
<thead>
<tr>
<th>PMID</th>
<th>1st Author, Year, Reference</th>
<th>Age_1</th>
<th>Age_2</th>
<th>Method</th>
<th>No.</th>
<th>Survival</th>
<th>Cranio</th>
<th>Histo (*)</th>
<th>Neuro</th>
<th>Behave</th>
<th>Obs.</th>
</tr>
</thead>
<tbody>
<tr>
<td>32290848</td>
<td>Early, 2020 [37]</td>
<td>Adult 4mo</td>
<td>Old 18 mo</td>
<td>CCI, 4m/s, 0.9, 30ms</td>
<td>1</td>
<td>1, 3, 7 d</td>
<td>Y</td>
<td>Y</td>
<td>NA</td>
<td>NA</td>
<td>aged increased altered edema, inflammation and astrocytes function (aquaporin 4)</td>
</tr>
<tr>
<td>30904769</td>
<td>Ritzel, 2019 [38]</td>
<td>Adult 3mo</td>
<td>Old 18 mo</td>
<td>CCI, 6m/s, 1mm, 50 ms</td>
<td>1</td>
<td>3 d</td>
<td>Y (sham no crano)</td>
<td>Y</td>
<td>Y</td>
<td>Anxiety Microglial sensitivity and peripheral immune dysfunction</td>
<td></td>
</tr>
<tr>
<td>23273602</td>
<td>Kumar, 2013 [39]</td>
<td>3 mo</td>
<td>24 mo</td>
<td>CCI 6m/s, 2mm</td>
<td>1</td>
<td>1, 7 d</td>
<td>Y</td>
<td>Y</td>
<td>NA</td>
<td>NA</td>
<td>Highly reactive macrophage and microglia, hippocamp</td>
</tr>
<tr>
<td>27090212</td>
<td>Morganti, 2016 [40]</td>
<td>3 mo</td>
<td>23 mo</td>
<td>CCI 4, 0.95, 300ms</td>
<td>1</td>
<td>1 d</td>
<td>Y</td>
<td>Y</td>
<td>NA</td>
<td>NA</td>
<td>Peripheral CCR2 macro recruit increased in aged brains</td>
</tr>
<tr>
<td>29848996</td>
<td>Chou, 2018 [41]</td>
<td>3-6 mo</td>
<td>20-25 mo</td>
<td>CCI 4m/s, 0.95, 300ms</td>
<td>1</td>
<td>4, 7, 28d</td>
<td>Y</td>
<td>Y</td>
<td>NA</td>
<td>NA</td>
<td>Y (spatial learning and memory) CCR2/5 response increased in aged reducing behavioral recovery</td>
</tr>
<tr>
<td>30486287</td>
<td>Krukowski, 2018 [42]</td>
<td>NA</td>
<td>19mo</td>
<td>CCI 4m/s, 0.95, 300ms</td>
<td>1</td>
<td>1 mo</td>
<td>Y</td>
<td>Y</td>
<td>Complement C1q, C3, CR3</td>
<td>Y (spatial learn and memory)</td>
<td>Complement activation, loss of synapses, increased macrophages MRI-T2: lesion size peaked, cavitation size increase, hippocamp CD11 increase</td>
</tr>
<tr>
<td>22952778</td>
<td>Timaru-Kast, 2012 [43]</td>
<td>2mo</td>
<td>21mo</td>
<td>CCI, 8m/s, 1mm, 150 m/s</td>
<td>1</td>
<td>1h, 24h, 3 d</td>
<td>Y</td>
<td>Y, IL1, IL6 TNF</td>
<td>Y</td>
<td>Y</td>
<td>Age effect on Brain Edema Formation, Secondary Brain Damage and Inflammatory Response inflammaging = increase of IL1, IL6, TNF x2 in aged animals</td>
</tr>
<tr>
<td>Study ID</td>
<td>Author Year</td>
<td>Age (mo)</td>
<td>Age (mo)</td>
<td>Model Details</td>
<td>Force (g)</td>
<td>Speed (m/s)</td>
<td>Duration (days)</td>
<td>Y (Proteomics)</td>
<td>Y (Impact)</td>
<td>Y (Histo)</td>
<td>Notes</td>
</tr>
<tr>
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</tr>
<tr>
<td>29269117</td>
<td>Cheng 2018</td>
<td>3</td>
<td>13</td>
<td>CHIMERA, 50 g, 0.5 m/s (0.6 J)</td>
<td>2</td>
<td>14 d - 8 mo</td>
<td>No</td>
<td>Y</td>
<td>Y</td>
<td>Y</td>
<td>APP vs WT. In WT, microgliosis and axonal injury may persist 8 mo after injury.</td>
</tr>
<tr>
<td>30636629</td>
<td>Cheng 2018</td>
<td>3 mo</td>
<td>13 mo</td>
<td>CHIMERA, 0.1 - 0.7 J</td>
<td>1</td>
<td>14 d</td>
<td>No</td>
<td>Y</td>
<td>Y impact &gt;0.5 J</td>
<td>Y</td>
<td>threshold increase of Iba and GFAP in corpus callosum.</td>
</tr>
<tr>
<td>25904805</td>
<td>Webster 2015</td>
<td>APP1 (8 mo)</td>
<td>WT</td>
<td>CHI 5 m/s, 1 mm, 100 ms</td>
<td>1</td>
<td>9h, 1d, 7d, 1 mo 3 mo</td>
<td>No</td>
<td>Y</td>
<td>Y</td>
<td>Proteomics+</td>
<td></td>
</tr>
<tr>
<td>23953759</td>
<td>Hefendehl 2014</td>
<td>3 mo, 12 mo, 27 mo</td>
<td>Laser (910 nm, 15 s)</td>
<td>21 d</td>
<td>Yes</td>
<td>Y</td>
<td>NA</td>
<td>NA</td>
<td>Microglia soma increased, process length decreased, low responsiveness, no homogenous distrib. (photon microscopy)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>PMI</th>
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<th>Method</th>
<th>Number Hits</th>
<th>Survival</th>
<th>Craniotomy</th>
<th>Histo (*)</th>
<th>Neuro</th>
<th>Behave</th>
<th>Obs</th>
</tr>
</thead>
<tbody>
<tr>
<td>22245525</td>
<td>Shultz 2012</td>
<td>Adult rat (300 grm)</td>
<td>NA</td>
<td>LFP, very mild (0.5 atm)</td>
<td>1</td>
<td>4days/1month</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Increased microglial/macros and reactive astrogliosis</td>
</tr>
<tr>
<td>21704658</td>
<td>Shultz 2011</td>
<td>Adult rat</td>
<td>NA</td>
<td>LFP 1 atm</td>
<td>1</td>
<td>24h/1 mo</td>
<td>Yes</td>
<td>Yes</td>
<td>NA</td>
<td>Cognitive, anxiolytic Astrogliosis and microgliosis in corpus callosum</td>
<td></td>
</tr>
<tr>
<td>21933013</td>
<td>Shultz 2012</td>
<td>Adult rat (repeated)</td>
<td>NA</td>
<td>LFP 1 atm</td>
<td>1-3-5</td>
<td>5d/2 mo</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Apoptosis, cortex and hippocamp</td>
</tr>
<tr>
<td>8155285</td>
<td>Hamm 1993</td>
<td>Adult rats</td>
<td>NA</td>
<td>LFP 2 atm</td>
<td>1</td>
<td>7d/12d</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Selective hippocampal modification with neurobehavioral changes of the environment and a flexible response to it</td>
</tr>
<tr>
<td>30998995</td>
<td>Arulsamy 2019</td>
<td>Rats 12mo</td>
<td>NA</td>
<td>WD (450 gr, 0.75 m)</td>
<td>1</td>
<td>12 mo</td>
<td>No</td>
<td>Yes Proteomics +</td>
<td>Yes</td>
<td>Yes</td>
<td>Age and not mTBI intensity affect executive function</td>
</tr>
<tr>
<td>28855139</td>
<td>Collins-Praino 2013</td>
<td>Rats 3 mo</td>
<td>NA</td>
<td>WD</td>
<td>1</td>
<td>3 mo</td>
<td>No</td>
<td>Yes IL1, IL6, TNF (9 cytokines)</td>
<td>No</td>
<td>Yes</td>
<td>mTBI induces changes when microglial priming exists</td>
</tr>
<tr>
<td>20528171</td>
<td>Spain 2010</td>
<td>Adult mice</td>
<td>NA</td>
<td>LFP, 0.9 atm</td>
<td>1</td>
<td>4h – 1½ mo</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Learning</td>
<td>PT changes unmyelinated axons in both ipsi and contra</td>
</tr>
<tr>
<td>26774527</td>
<td>Muccigrosso 2016</td>
<td>Adult mice</td>
<td>NA</td>
<td>MidPF 1.2 atm</td>
<td>1</td>
<td>1 mo</td>
<td>Yes IL-1ß, CCL2, TNFα</td>
<td>Yes</td>
<td>Yes, at 1 mo Hippocampal Learning after 1-week, anterograde learning after 1 mo</td>
<td></td>
<td></td>
</tr>
<tr>
<td>28910378</td>
<td>Taib 2016</td>
<td>Adult mice</td>
<td>NA</td>
<td>CCI, 3.5 m/s, 1mm, 50ms</td>
<td>1</td>
<td>1w-3 mo</td>
<td>Yes</td>
<td>IL1b and CD11b increased max at 3 days + demyelination at 3 mo in CC Proteomics+</td>
<td>Signific. at 3 mo</td>
<td>Yes, + at 3 mo Neuro-inflammation (microglia) and WMI. Myelin sheath defects in corpus callosum at 3 months (em)</td>
<td></td>
</tr>
<tr>
<td>24289885</td>
<td>Fenn 2014</td>
<td>3 mo, mice</td>
<td>NA</td>
<td>LPF 2 atm</td>
<td>1</td>
<td>7d - 1 mo</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Motor</td>
<td>Primed microglia reduced motility and increased soma</td>
</tr>
<tr>
<td>24223856</td>
<td>Tajiri 2013</td>
<td>Adult mice</td>
<td>NA</td>
<td>CCI4.0 m/s, 1 mm, 150 ms</td>
<td>1</td>
<td>1 ½ mo</td>
<td>Yes</td>
<td>Increased Abeta deposits</td>
<td>Significant errors</td>
<td>Tg2576 APP mice, more Abeta in both cortex and hippocam</td>
<td></td>
</tr>
<tr>
<td>29993324</td>
<td>Mouzon 2019</td>
<td>3 mo, mice</td>
<td>NA</td>
<td>CH1 5m/s, 1mm, 200 ms</td>
<td>1 or 5</td>
<td>12 mo</td>
<td>No</td>
<td>Yes CA1/CA3 hippocamp</td>
<td>Yes</td>
<td>Yes, Cognitive impair at single impact</td>
<td>hTau minimal after 1 impact, WMI present</td>
</tr>
<tr>
<td>Study ID</td>
<td>Year</td>
<td>Species</td>
<td>Age</td>
<td>Gender</td>
<td>Impact Type</td>
<td>Impact Parameters</td>
<td>Outcome 1</td>
<td>Outcome 2</td>
<td>Outcome 3</td>
<td>Outcome 4</td>
<td>Outcome 5</td>
</tr>
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</tr>
<tr>
<td>Mouzon 2017</td>
<td>2017</td>
<td>mice</td>
<td>3</td>
<td>NA</td>
<td>CHI</td>
<td>5 m/s, 1 mm, 200 ms</td>
<td>24 mo</td>
<td>No</td>
<td>Yes</td>
<td>Increased risk behavior at 24 mo</td>
<td>Iba-1, GFAP+</td>
</tr>
<tr>
<td>Lyons 2018</td>
<td>2018</td>
<td>mice</td>
<td>4</td>
<td>NA</td>
<td>CHI</td>
<td>5 m/s, 1, 100 ms</td>
<td>3d, 28d</td>
<td>No</td>
<td>Yes</td>
<td>NA</td>
<td>Learning &amp; spatial memory</td>
</tr>
<tr>
<td>Zohar 2003</td>
<td>2003</td>
<td>mice</td>
<td>2</td>
<td>NA</td>
<td>WD</td>
<td>20,25,30 grms</td>
<td>7d, 1 mo, 2 mo, 3 mo</td>
<td>No</td>
<td>Yes</td>
<td>No differe</td>
<td>Learning and spatial memory</td>
</tr>
<tr>
<td>Rachman 2013</td>
<td>2013</td>
<td>mice</td>
<td>2</td>
<td>NA</td>
<td>WD</td>
<td>30 grms</td>
<td>3 d, 7 d, 1 mo</td>
<td>No</td>
<td>Yes, diffuse</td>
<td>Yes</td>
<td>cell culture, p53</td>
</tr>
<tr>
<td>Rachman 2012</td>
<td>2012</td>
<td>mice</td>
<td>2</td>
<td>NA</td>
<td>WD</td>
<td>30 grms</td>
<td>24h, 3 d, 7 d</td>
<td>No</td>
<td>Yes, diffuse</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Tashlykov 2006</td>
<td>2006</td>
<td>mice</td>
<td>2</td>
<td>NA</td>
<td>WD</td>
<td>5-30 grms</td>
<td>3 d</td>
<td>No</td>
<td>Yes, cingulate</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>Tashlykov 2008</td>
<td>2008</td>
<td>mice</td>
<td>2</td>
<td>NA</td>
<td>WD</td>
<td>5-30 grms</td>
<td>3 d</td>
<td>No</td>
<td>Yes p53 and Bcl-2</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>Baratz 2015</td>
<td>2015</td>
<td>mice</td>
<td>2</td>
<td>NA</td>
<td>WD</td>
<td>50 grms</td>
<td>1-18h, 72 h, 7 days</td>
<td>No</td>
<td>Yes, GFAP</td>
<td>NA</td>
<td>Cognitive impairment</td>
</tr>
<tr>
<td>Luo 2014</td>
<td>2014</td>
<td>mice</td>
<td>3</td>
<td>NA</td>
<td>CHI</td>
<td>3-5 m/s, 200ms, 0.9mm</td>
<td>1 d, 6 mo</td>
<td>No</td>
<td>Yes, CREB</td>
<td>No change after repeated</td>
<td>Cognitive impair after repeated</td>
</tr>
<tr>
<td>Taylor 2020</td>
<td>2020</td>
<td>mice</td>
<td>3</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>Cerebral microbleeds (MRI in-vivo and ex-vivo + histo) without previous trauma.</td>
</tr>
<tr>
<td>Aungst 2020</td>
<td>2020</td>
<td>rats</td>
<td>3</td>
<td>NA</td>
<td>LFP</td>
<td>1.1 atm</td>
<td>28 days</td>
<td>Yes</td>
<td>neuronal cell loss increased numbers of activated microglia</td>
<td>Yes</td>
<td>Yes</td>
</tr>
</tbody>
</table>

Table 3. Associated studies related to mTBI in adult and senescent rats (*) axonal destruction, apoptosis, activation of astrogliosis and microgliosis, inflammation; LFP: lateral fluid percussion, MFP: midline fluid percussion, WD: weight drop, CHI: closed head injury, NA - not available, PT: posttraumatic, MRI: magnetic resonance imaging, em: electronic microscopy, CC: corpus callosum

Histo: histology; Neuro: neurologic; Behave: Behavioral
References


41. Chou A, Krukowski K, Morganti JM et al. Persistent Infiltration and Impaired Response of Peripherally-Derived Monocytes after Traumatic Brain


50. Shultz SR, Bao F, et al. Repeated mild lateral fluid percussion brain injury in the rat causes cumulative long-term behavioral impairments, neuroinflammation,


