



BioBacta

Journal of Medical and Life Science
www.jmals.journals.ekb.eg

Celecoxib effect on rivastigmine anti-Alzheimer activity against aluminum chloride-induced neurobehavioral deficits as a rat model of Alzheimer's disease; novel perspectives for an old drug

Raafat A. Abdel-Aal ^a, Ola A. Hussein ^b, Reham G. Elsaady ^c, Lobna A. Abdelzaher ^d

^{a,c,d} Department of Pharmacology, Faculty of Medicine, Assiut University, Assiut, Egypt.

^b Department of Histology and Cell Biology, Faculty of Medicine, Assiut University, Assiut, Egypt.

*Correspondence should be addressed to:

Lobna A. Abdelzaher

Department of Pharmacology, Faculty of Medicine, Assiut University, Assiut, Egypt.

Lobna@aun.edu.eg.

ORCID ID <http://orcid.org/0000-0001-8438-7924>

Celecoxib impact on rivastigmine anti-Alzheimer activity

DOI: [10.21608/JMALS.2021.210630](https://doi.org/10.21608/JMALS.2021.210630)

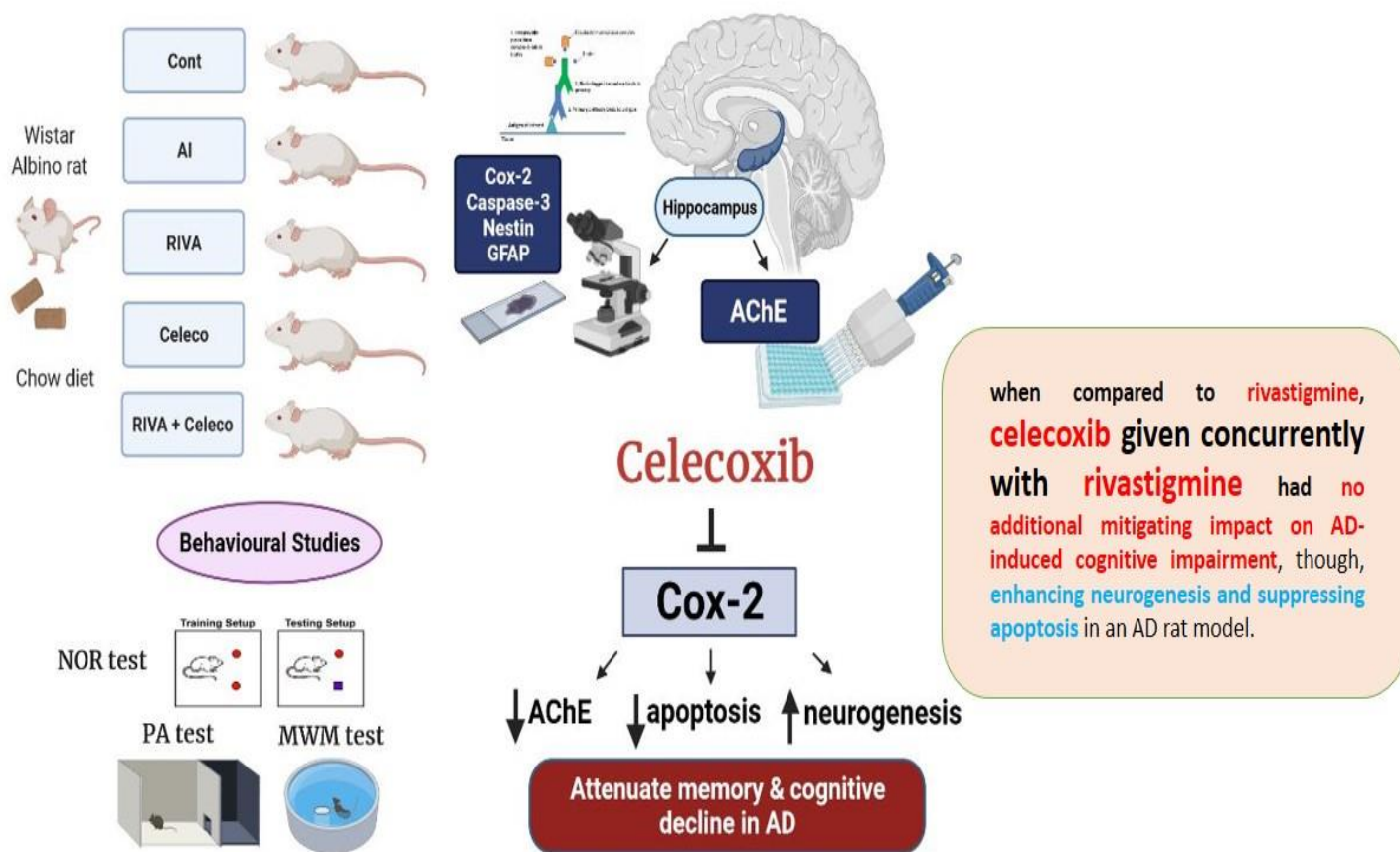
Abstract

Neuroinflammation plays a crucial role in Alzheimer's disease (AD) pathogenesis. Apoptosis, along with impaired neurogenesis, has been linked to AD neurodegenerative cell death, likely due to overexpression of cyclooxygenase-2 (COX-2). We investigated whether the concurrent administration of celecoxib, a selective COX-2 inhibitor, with rivastigmine, the standard anti-Alzheimer, would enhance rivastigmine anti-Alzheimer activity in the aluminum chloride (AlCl₃) Alzheimer's rat model. Male rats were randomly assembled into control (Cont), AlCl₃-treated (Al), rivastigmine-treated (RIVA), celecoxib-treated (Celeco), and combined rivastigmine and celecoxib-treated (RIVA+Celeco) groups. They were studied for memory, and cognitive skills, along with evaluating hippocampal acetylcholinesterase (AChE) activity. Hippocampal neuropathology, besides apoptosis, astroglial injury, and neurogenesis, were assessed by examining the expression of their related protein markers; activated caspase-3, glial fibrillary acidic protein (GFAP), and nestin. Celecoxib, rivastigmine, and their combination attenuated AlCl₃-induced intellectual impairment and the associated neurodegenerative changes. However, the combination therapy had no additional neuroprotective advantage over rivastigmine alone, except for the enhancement of neurogenesis and suppression of apoptosis in the AL-intoxicated rats. As compared to rivastigmine, the efficacy of celecoxib in combination with rivastigmine confers neuroprotection only at the cellular level, enhancement of neurogenesis, and suppression of apoptosis, without having a mitigating effect on Al-induced cognitive impairment.

Keywords: Alzheimer's disease; Celecoxib; Behavior; Caspase-3; Nestin.

Received November 10, 2021; Accepted December 18, 2021; Published December 24, 2021

Graphical abstract



1. Introduction

AD is the most prevailing cause of senile dementia, disability, and dependence among elderly over 65 years of age worldwide [1]. Acetylcholinesterase inhibitors (AChEIs), rivastigmine [2–4], and NMDA antagonist, memantine [5], are the currently FDA-approved anti-Alzheimer medications. They can, however, only offer symptomatic relief and could not delay the disease progression [6,7].

Amyloid- β ($A\beta$); the key component of senile plaque and neurofibrillary tangles (NFTs); the pathological insoluble aggregates of hyperphosphorylated tau proteins are the hallmark lesions of AD [8]. $A\beta$ -induced neuronal apoptosis

elicited by caspase-3 activation [9] besides impairment of adult hippocampal neurogenesis [10] mediates further cognitive decline.

COX-2 overexpression [11,12], activated microglia, astrocyte invasion, as well as cholinergic neuronal degeneration, have all been linked to AD neuroinflammatory reactions [13]. Neuroinflammation normally starts as host defense, then becomes detrimental with subsequent neuronal degeneration [14,15]. Celecoxib, a selective COX-2 inhibitor, improves the cognitive decline in APP/PS1 transgenic mice; the rare AD familial type [16]. However, its potential neuroprotective role in the sporadic, more

common AD type, which accounts for 95% of all cases [17], has not been thoroughly investigated [17,18].

In the sporadic AD type; $AlCl_3$ rat model, we investigated whether celecoxib, when given concurrently with rivastigmine, may enhance rivastigmine anti-Alzheimer activity.

The potential mitigating effect of celecoxib was investigated in the rivastigmine-treated Alzheimer's rat model by analyzing the rats' behavior, hippocampal histopathological changes, and AChE activity, as well as investigating the expression of inflammatory, neurogenesis, and apoptosis markers.

2. Materials and methods

2.1. Animals and experimental design

All the animal procedures were approved by the Faculty of Medicine Institutional Animal Care and Use Committee (IRB no:17100383) and can be given upon request. They were adopted following an update of the National Institute of Health Guide for the Care and Use of Laboratory Animals (NIH Publications No. 8023, revised 1978). Every effort has been made to minimize the number of animals used and their distress. Adult male Albino Wistar rats weighing 180–220 g were purchased from the animal house of the Faculty of Medicine, Assiut University. They were housed in stainless steel cages in a well-ventilated space under a 12-h light / dark cycle. Rats had access to water and food *ad libitum*. Rats were divided into five classes at random (n=10):

Group 1; Cont group received daily saline injections intraperitoneal (IP) for 60 days.

Group 2; Al group received $AlCl_3$ hexahydrate (qualikemes India), dissolved in saline, daily IP at a dose of 80 mg/Kg per injection for 60 days [19].

Group 3; RIVA group received $AlCl_3$ and rivastigmine (reference standard drug) (Beijing Mesochem Technology Co., Ltd.), dissolved in sterile water, daily IP at a dose of 1 mg/kg per injection for six weeks starting two weeks before $AlCl_3$ administration [20,21].

Group 4; Celeco group received $AlCl_3$ and celecoxib (Beijing Mesochem Technology Co., Ltd.), dissolved in saline, daily at a dose of 20 mg/kg per injection IP for four weeks concurrently with $AlCl_3$ [22,23].

Group 5; RIVA+Celeco group received $AlCl_3$, rivastigmine (1 mg/kg for six weeks starting two weeks before $AlCl_3$ administration), and celecoxib (20 mg/kg for four weeks concurrently with $AlCl_3$).

2.2. Behavioral studies

Cognitive performance was assessed using a battery of behavioral tests measuring episodic, aversive (emotional) conditioning, and visuospatial memory via NOR, PA, and MWM tests.

2.2.1. Novel Object Recognition (NOR) test

The test is designed to evaluate rodents' exploratory nature, memory, and object recognition based on their natural preference for exploring novel objects over familiar ones. The test was performed in a room with a uniformly dim light provided by a ceiling-mounted halogen lamp. The NOR test apparatus is a 60 x 60 x 40 cm square stainless steel open field box with black walls and floor. The NOR test objects were distinct in form and color, and they were made of heavily painted wood, which rats couldn't lift. They were about 15 cm in height. The test box and objects were wiped with 70% ethyl alcohol between trials to omit behavioral tasks linked to olfactory cues. Rats were kept in the experimental room for at least 30 min before testing. They were habituated to the empty test box for 10 min per day for two sequential days. Each rat was put in the test box with two identical objects for the amount of time needed to spend a total of 15 sec exploring these two objects to exclude the possibility of random preference. Exploration was considered when, within a cut-off period of 4 min, the rat was touching, looking at, or sniffing with its head within 2 cm of the object. Rats that had explored both objects for less than 15 sec were not included in the experiment. Following the learning phase, three assessment sessions were performed to evaluate the

short, moderate, and long-term memories after a retention period of 5 min, 2 hrs, and 24 hrs. The rats were permitted 3 min to explore one of the objects they had seen during the learning process as well as a novel object. Rats with a low level of object exploration, described as less than 5 sec spent exploring novel and familiar objects, were excluded from the study. Memory performance was evaluated through the recognition index (RI), which was calculated as (time spent exploring the novel object) / (total time spent exploring both familiar and novel objects)*100 [24].

2.2.2. *Passive avoidance (PA) test*

The PA test is a fear-motivated test used to evaluate learning and memory in rodent models of central nervous system (CNS) disorders. It requires a combination of an aversive stimulus, mild foot shock, and a particular environmental circumstance. The apparatus consists of two chambers separated by an 8 cm communicating hole within a separating wall. A single chamber was kept lit. The rats were put individually in the illuminated chamber on the first day, and an electrical shock was administered to their feet when they entered the dark chamber. Rats were put in the illuminated chamber 24 hrs later, and the step-through latency, the time lag till entry to the dark chamber, was recorded [19].

2.2.3. *Morris water maze (MWM) test*

The MWM test is a behavioral task that evaluates hippocampal-dependent learning and memory in rodents. MWM apparatus consists of a large circular pool (45 x 160 cm, filled to a depth of 30 cm with water at 28 ± 1 °C). Four colored light clues (lambs) were placed on each quadrant wall, dividing the tank into four equal quadrants. Since the light clues were used as a reference memory, they remained constant throughout the experiment. Each rat was exposed to four consecutive trials, considering changing the drop location for each trial, with a gap of 5 min. The rat was gently placed in the water between quadrants

facing the pool rim and given 120 sec to locate the platform, during which time the latency to reach the platform was measured. The rat was given 20 sec to remain on the platform. If it did not reach within 120 sec, it was tenderly directed to the platform and left there for 20 sec. Retention trials were conducted 24 hours after the initial trials to observe spatial reference memory. The latency to reach the target area was calculated as well [25].

2.3. **Acetylcholinesterase (AChE) activity**

The animal groups were sacrificed, and their brains were carefully dissected at the end of the experiment. Hippocampi were weighed, homogenized in phosphate buffer saline (PBS), centrifuged for 5 min at $5000 \times g$, and the supernatant was collected. The hippocampal AChE activity was determined using ELISA kits (Elabscience Co.) according to the manufacturer's protocol. After the enzyme-substrate reaction was completed, the optical density (OD) was calculated using a microplate reader at a wavelength of 450 nm. By comparing the OD of the samples to the standard curve, the AChE activity was assayed.

2.4. **Histopathological studies**

Rats were anesthetized with thiopental sodium (50 mg/kg) IP [26], their hearts were exposed and transcardially perfused with saline till getting clear flow, then finalized with 10% formalin. Dissected hippocampal tissues were processed for light microscopy and immunohistochemical staining techniques. Some sections were stained with hematoxylin and eosin (Hx& E) [27].

2.5. **Immunohistochemistry Studies**

The isolated hippocampi were fixed in 10% neutral formalin, dehydrated, cleared, and paraffin-embedded. Paraffin sections were incubated overnight at 4 °C with the following primary antibodies; rabbit COX-2 antibody (1:100) (Thermo Fisher Scientific, Fremont, CA 94538, USA), rabbit antimouse caspase-3 polyclonal antibody (1:100) (Chongqing Biospes co., Ltd. China), mouse monoclonal anti-nestin antibody

(Abcam, ab22035, UK) (1:100) and anti-GFAP; Ab-1 (Clone GA-5) mouse monoclonal antibody (1:100) (Thermo Fisher Scientific Co, Ferment, California, USA). After that, sections were stained with an avidin-biotin-peroxidase system with diaminobenzidine as the chromogen (DAKO (HRP; rabbit/mouse/goat (DAB+) code no. K0679; Dako Cytomation) according to the instructions enclosed in the Dako LSAB+ System-HRP. Hematoxylin was used to counterstain the studied sections. For negative control staining, some sections were incubated with PBS instead of the primary antibody.

2.6. Morphometric Studies

Morphometric studies were carried out with the help of Image J, a Java-based open-source image processing kit. Three non-overlapped fields/five sections/three rats from each group were used to calculate the tested parameters. The number of dark cells in cornu ammonis 1 (CA1) fields was determined in the Hx&E-stained sections. The number of caspase-3 (+ve) and the number of nestin (+ve) immunostained cells in CA1 areas were calculated using an X40 lens as well.

2.7. Electron Microscopy Studies

With the aid of a dissecting microscope, the extracted hippocampi were dissected out, fixed in glutaraldehyde, and processed for transmission electron microscopy. Toluidine blue was used to stain semi-thin sections (0.5-1 μm). For the selected areas in the semi-thin sections, ultrathin sections (500-800A) were contrasted with uranyl acetate and lead citrate, examined with the JEOL (JEM-100 CXII, Tokyo, Japan) transmission electron microscope (TEM), and photographed at 80 kV in the Assiut University-Electron Microscope Unit.

Statistical analysis

Data were expressed as mean \pm standard error of the mean (SEM). Statistical analysis was performed by multiple t-tests, one-way and two-way repeated-measures ANOVA, followed by post hoc Tukey's test

when appropriate. All statistical tests were directed using GraphPad Prism 7 software. The difference among groups was considered significant for $p < 0.05$.

3. Results

3.1 behavioral outcomes

3.1.1 Novel Object Recognition (NOR) test

The time spent exploring a novel object was significantly longer than that consumed for the familiar one in the Cont group at 5 min, 2 hrs, and 24 hrs. The AI group had poor memory performance, as shown by the lack of preference for a novel item compared to the Cont group ($p < 0.001$, 5 min; 2 hrs; $p < 0.01$, 24 hrs) (Fig. 1). RIVA group spent substantially more time exploring the novel object relative to the AI group ($p < 0.05$, 5 min; 2 hrs) (Fig. 1). At the given time points, the Celeco group spent more time investigating the novel object compared to the AI group ($p < 0.05$, 5 min; 2 hrs) (Fig. 1). However, its concurrent administration with rivastigmine could not significantly improve memory performance than rivastigmine-only therapy (Fig. 1).

3.1.2 Passive avoidance (PA) test

AI group showed marked cognitive deficit detected as a significant decrease in step-through latency when compared to the Cont group ($p < 0.001$) (Fig. 2). Rivastigmine and celecoxib improved learning and memory tasks as they significantly enhanced step-through latency of the AI-intoxicated rats ($p < 0.01$, $p < 0.05$). However, the retention latency time was not significantly different between the RIVA+Celeco and the RIVA groups (Fig. 2).

3.1.3 Morris water maze (MWM) test

The MWM test assessed spatial learning and memory through the acquisition and probe trials, respectively. AIC13 administration produced a significantly increased time to reach the platform in all four quadrants compared to the Cont group ($p < 0.001$) (table 1). RIVA, Celeco, and RIVA+Celeco groups showed significantly decreased time to reach the platform in all four quadrants when compared to the

Al group ($p < 0.05$) (table 1). In probe trials, the Al group had significantly longer escape latency to the platform site than the Cont group ($p < 0.01$). In contrast, the treated groups had significantly shorter escape latency compared to the Al group in all four quadrants ($p < 0.05$) (table 2). Therefore, celecoxib may have the ability to rescue the learning and the memory deficit induced by AIC13 in the AD rat model without any added benefits of the combination therapy over rivastigmine treatment.

3.2 Acetylcholinesterase (AChE) activity

AChE activity was evaluated in the hippocampi of the studied groups. Hippocampal AChE activity was significantly enhanced, an indication of cholinergic impairment, in the Al group when compared to the Cont group ($p < 0.001$) (Fig. 3). RIVA and Celeco groups dramatically reduced hippocampal AChE activity compared to the Al group ($p < 0.05$) (Fig. 3). RIVA+Celeco group showed a non-significant decrease in hippocampal AChE activity than the RIVA group (Fig. 3). Collectively, celecoxib could ameliorate cholinergic dysfunction, the hallmark of AD, through downregulating AChE activity; however, it could not confer additive effect when concurrently administered rivastigmine compared to rivastigmine only therapy.

3.3 Histopathological studies

Examination of the Hx&E-stained sections of the Cont group brain revealed the normal structure of the rat hippocampus. It was made up of the outer (Ob) and inner (Ib) blades of the dentate gyrus (DG), as well as the CA1, CA2, and cornu ammonis 3 (CA3) fields of Ammon's horn (AH) (Fig. 4a). Cont CA1 field showed stratum oriens (SO), stratum pyramidal (SP), and stratum radiatum (SR) that extended from the alveus to the hippocampal fissure (HF) (Fig. 4a). The pyramidal neurons (P), the principal cell type in the CA1 field, were characterized by their triangular perikarya, medium-sized round vesicular nuclei (N), and basophilic cytoplasm. Their processes extended to

stratum radiatum (SR) (Fig. 4b). The Al group showed multiple intensely stained irregular pyramidal neurons surrounded by empty spaces (Fig. 4c). Most pyramidal neurons (P) appeared normal with round vesicular nuclei (N), yet few cells still seemed irregular and deeply stained in the RIVA group (Fig. 4d). Celco group showed some pyramidal neurons (P) with a regular appearance and round vesicular nuclei (Fig. 4e). However, most pyramidal neurons (P) seemed regularly shaped with round vesicular nuclei in the RIVA+ Celeco group (Fig. 4f). Our morphometric results revealed a significant increase in the number of dark cells ($p < 0.0001$) in the Al group compared to the Cont group. A significant decrease in their numbers, however, was observed in the treated groups compared to the Al group ($p < 0.0001$) (Fig. 4g). Remarkably, the RIVA+Celeco group had a significantly lower number of dark cells compared to the RIVA group, indicating the additive neuroprotective effect of celecoxib when combined with rivastigmine ($p < 0.05$) (Fig. 4g).

3.4 Immunohistochemical Studies

3.4.1 COX-2 expression (hippocampus; CA1 field)

Cont group revealed few COX-2 (+ve) immunostained cells (Fig. 5a). A marked increase in COX-2 (+ve) immunostained cells was noticed in the Al group compared to the Cont group (Fig. 5b). RIVA, Celeco, and RIVA+ Celeco groups showed a marked reduction in COX-2 (+ve) immunostained cells compared to the Al group (Figs. 5c-5e). Morphometric results revealed a significant increase in COX-2 (+ve) immunostained cells in the Al group compared to the Cont group ($p < 0.0001$). In contrast, treated groups showed a significant reduction in COX-2 (+ve) immunostained cells compared to the Al group ($p < 0.0001$). There was also a substantial reduction in COX-2 (+ve) immunostained cells in the RIVA+Celeco group relative to the RIVA group ($p < 0.001$) (Fig. 5f), suggesting that the combination

therapy has a stronger anti-inflammatory effect compared to the rivastigmine only therapy.

3.4.2 Active caspase-3 expression (hippocampus; CA1 field)

Caspase-3 is one of the key proteases of the caspase cascade that is involved in apoptosis. Cont group revealed few caspase-3 (+ve) immunostained cells (Fig. 6a). A marked increase in caspase-3 (+ve) immunostained cells was noticed in the Al group compared to the Cont group (Fig. 6b). RIVA, Napro, and RIVA+Napro groups showed a marked reduction in caspase-3 (+ve) immunostained cells compared to the Al group (Figs. 6c-6e). Morphometric results revealed a significant increase in caspase-3 (+ve) immunostained cells in the Al group compared to the Cont group ($p < 0.0001$). Treated groups, however, showed a significant reduction in caspase-3 (+ve) immunostained cells compared to the Al group ($p < 0.0001$). Furthermore, the RIVA+Celeco group had a significantly lower number of caspase-3 (+ve) immunostained cells than the RIVA group ($p < 0.05$) (Fig. 6f), indicating that the combination therapy has an additive antiapoptotic effect.

3.4.3 Nestin expression (hippocampus; CA1 & DG fields)

Decreased nestin (+ve) immunostained cells were observed in the Al group (Fig. 7b) relative to the Cont group (Fig. 7a), indicating impaired neurogenesis. RIVA, Celeco, and RIVA+ Celeco groups showed increased nestin (+ve) immunostained cells (Figs. 7c-7e) compared to the Al group indicating enhanced neurogenesis. Statistically, a significant reduction in nestin (+ve) immunostained cells was observed in the Al group compared to the Cont group ($p < 0.05$). The RIVA, the Celeco and the RIVA+Celeco groups showed a substantial increase in nestin (+ve)

immunostained cells ($p < 0.01$, $p < 0.001$, $p < 0.05$) relative to the Al group (Fig. 7f). However, the combination therapy had enhanced nestin expression compared to the RIVA group ($p < 0.05$). Immunohistochemically stained sections of DG fields revealed few nestin (+ve) immunostained cells in the Al and the RIVA groups (Figs. 7h&7i). In contrast, the RIVA+Celeco group showed a marked increase of nestin (+ve) immunostained cells (Fig.7j) compared to the RIVA group. As a result, when celecoxib is combined with rivastigmine, neurogenesis in the hippocampal CA1 and DG fields is likely to be enhanced relative to rivastigmine alone.

3.4.4 GFAP expression (hippocampus; CA1 field)

Immunostained sections of the CA1 field revealed few GFAP (+ve) immunostained cells in the Cont group (Fig. 8a). Al group revealed an increased GFAP (+ve) immunostained cells compared to the Cont group (Fig. 8b). The RIVA, the Celeco, and the RIVA+Celeco groups showed a persistent increase of GFAP (+ve) immunostained cells (Figs.8c-8e).

3.5 Electron microscopy studies

Electron microscopic examination of stratum pyramidale (SP) of the CA1 field of the Cont group revealed large pyramidal cells with large oval euchromatic nuclei. Their voluminous electrolucent cytoplasm contained multiple rER, mitochondria, and ribosomes (Fig. 9a). The majority of the cells had degenerated rarified cytoplasm with small involute nuclei and heterochromatin in the Al group (Fig. 9b). Some cells tended to be electron-dense (Fig. 9b). Ultrastructure examination of the RIVA, the Celeco, and the RIVA+Celeco groups showed that most pyramidal cells were more or less similar to the Cont group as they appeared electrolucent with oval euchromatic nuclei (Figs. 9c-9e).

Table 1 Effect of rivastigmine, celecoxib, and their combination on the acquisition trials of the Morris Water Maze (MWM) test

Groups	Quadrant 1 (latency in s)	Quadrant 2 (latency in s)	Quadrant 3 (latency in s)	Quadrant 4 (latency in s)
Cont	14.9 ± 3.6	18.3 ± 3.5	8.9 ± 1	9.3 ± 1
Al	74.3 ± 18.4 ^{***}	91.8 ± 12.4 ^{****}	31.3 ± 6.3 ^{***}	69.7 ± 14.3 ^{****}
RIVA	21.2 ± 4 ^{##}	19.1 ± 3.4 ^{####}	14.3 ± 1.9 [#]	18 ± 2.5 ^{####}
Celeco	26 ± 6.1 ^{##}	25.1 ± 3.9 ^{####}	14.5 ± 2.2 ^{##}	10.6 ± 1.2 ^{####}
RIVA+ Celeco	24.4 ± 3.3 ^{##}	26.6 ± 6.6 ^{####}	13.5 ± 3.3 ^{##}	10.3 ± 1.6 ^{####}

The data are expressed as mean ± SEM. Statistical analysis was performed by one-way repeated-measures ANOVA followed by a post hoc Tukey's test. All statistical tests were directed using GraphPad Prism 7 software. ^{***}*P*<0.001 compared with Cont; ^{****}*P*<0.0001 compared with Cont; [#]*P*<0.05 compared with Al; ^{##}*P*<0.01 compared with Al; ^{####}*P*<0.0001 compared with Al.

Table 2 Effect of rivastigmine, celecoxib, and their combination on the probe trials of the Morris Water Maze (MWM) test

Groups	Quadrant 1 (latency in s)	Quadrant 2 (latency in s)	Quadrant 3 (latency in s)	Quadrant 4 (latency in s)
Cont	13.7 ± 3.1	8.7 ± 1.6	9.4 ± 2.6	8.8 ± 0.7
Al	72 ± 18.9 ^{***}	40.9 ± 5.7 ^{****}	45.6 ± 14.4 ^{**}	41.2 ± 7.5 ^{****}
RIVA	29.3 ± 4.4 ^{##}	14.4 ± 1.9 ^{###}	13 ± 2.8 [#]	9.9 ± 0.7 ^{####}
Celeco	34.9 ± 7.3 [#]	19.8 ± 1.9 ^{###}	18 ± 4.3 [#]	11.7 ± 2.3 ^{####}
RIVA+ Celeco	28.3 ± 2.9 ^{##}	17.3 ± 2.8 ^{###}	12 ± 1.1 ^{##}	9.1 ± 0.4 ^{####}

The data are expressed as mean ± SEM. Statistical analysis was performed by one-way repeated-measures ANOVA followed by a post hoc Tukey's test. All statistical tests were directed using GraphPad Prism 7 software. ^{**}*P*<0.01 compared with Cont; ^{***}*P*<0.001 compared with Cont; ^{****}*P*<0.0001 compared with Cont; [#]*P*<0.05 compared with Al; ^{##}*P*<0.01 compared with Al; ^{###}*P*<0.001 compared with Al; ^{####}*P*<0.0001 compared with Al.

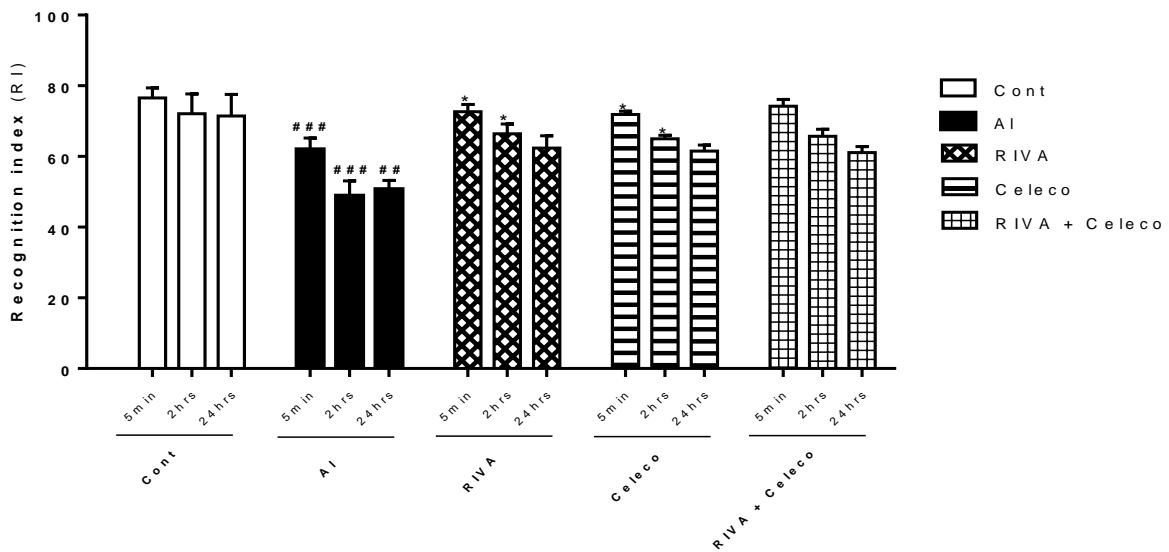


Fig 1. Effect of rivastigmine, celecoxib, and their combination on the recognition index (RI) of the novel object recognition (NOR) test in the A β 1-3-induced Alzheimer's in rats. The data are expressed as mean \pm SEM. * p < 0.05, ## p < 0.01, ### p < 0.001. #, ###: a significant difference from the Cont group. *: a significant difference from the AI group.

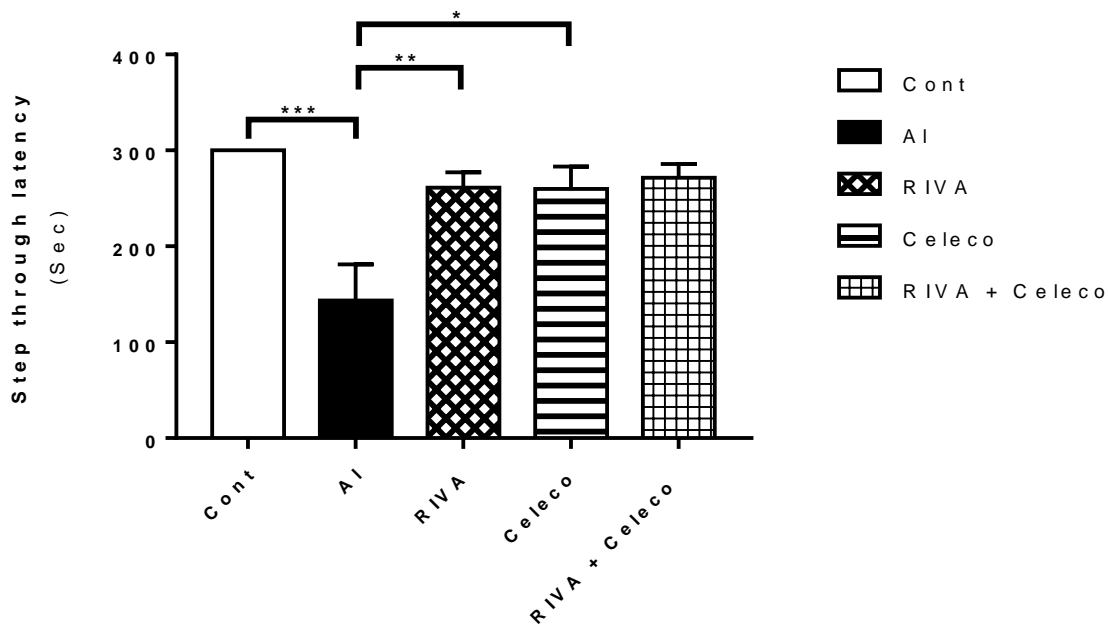


Fig 2. Effect of rivastigmine, celecoxib, and their combination on the step-through latency of passive avoidance (PA) test in the A β 1-3-induced Alzheimer's in rats. The data are expressed as mean \pm SEM. * p < 0.05, ** p < 0.01, *** p < 0.001.

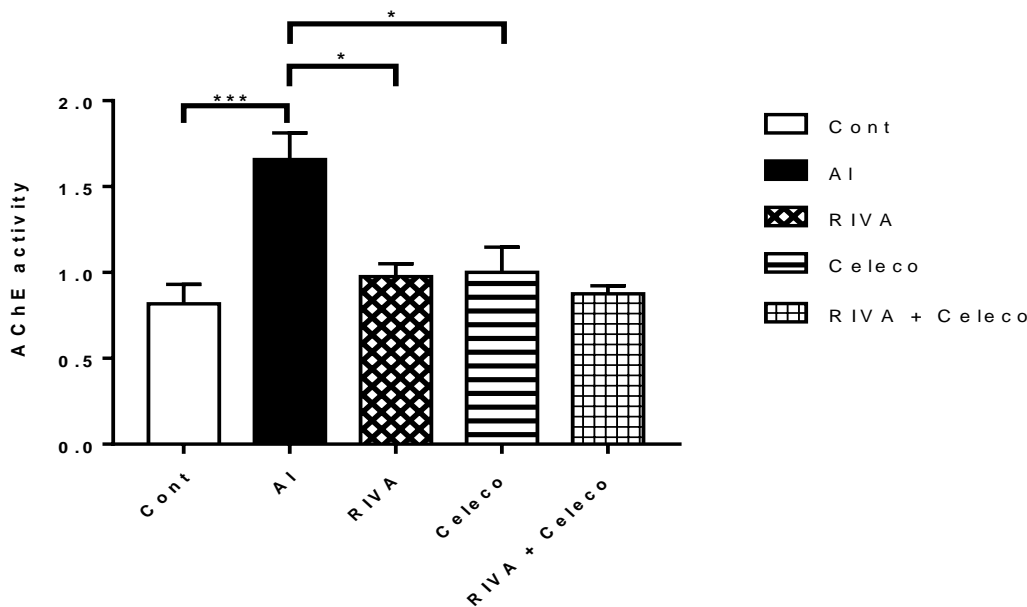


Fig 3. Effect of rivastigmine, celecoxib, and their combination on the acetylcholinesterase (AChE) in the AIC13-induced Alzheimer’s in rats. The data are expressed as mean ± SEM. * $p < 0.05$, *** $p < 0.001$.

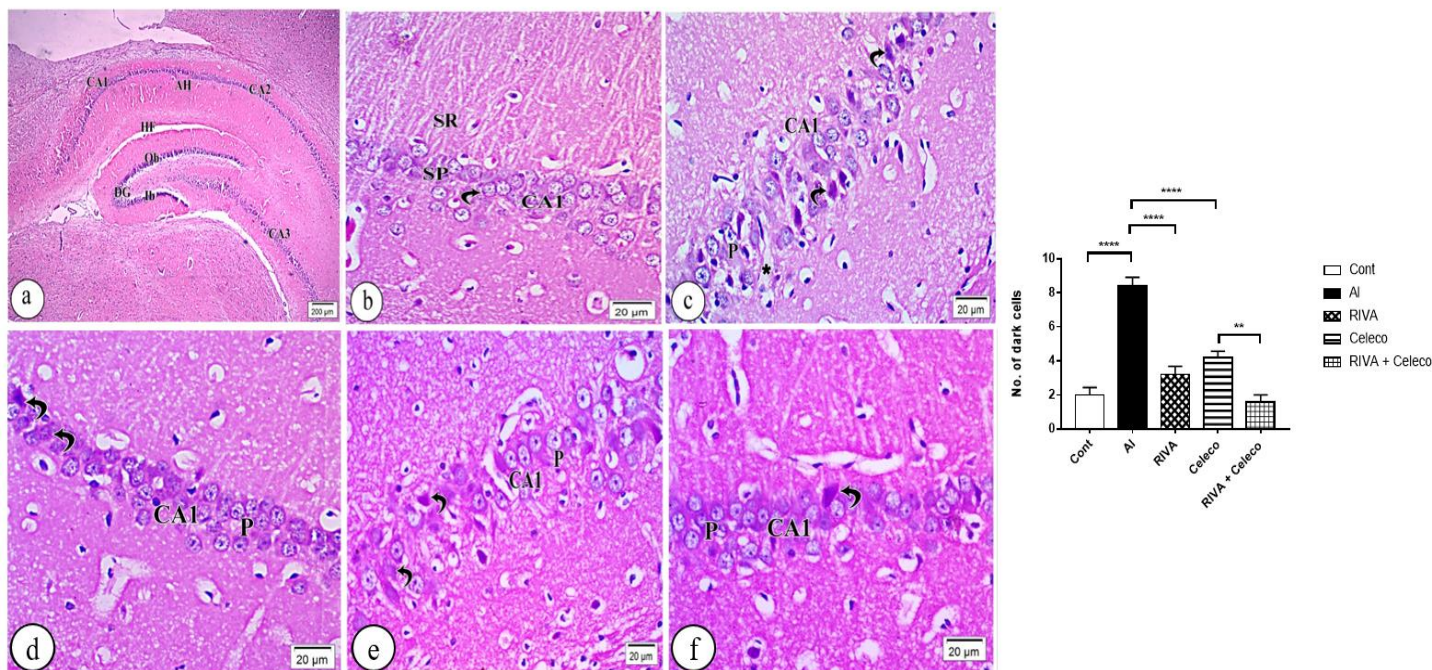


Fig 4. Photomicrographs of Hx&E stained sections of rat hippocampus (CA1 field). a) Cont hippocampus; showing V-shaped structure, inner blade (Ib) and outer blade (Ob) of the dentate gyrus (DG), CA1 of Ammon’s horn (AH), and hippocampal fissure (Hf) x40. b) Cont CA1; showing stratum pyramidal (SP), medium-sized pyramidal cells (curved arrow), round vesicular nuclei, and their processes extend to stratum radiatum (SR) x400. e) AI CA1; much irregular, dark shrunken pyramidal cells (curved arrows) surrounded by empty spaces (*). Few of them appear pale stained (P) x400. d) RIVA CA1; multiple pale stained pyramidal (P) cells with round vesicular nuclei, few of which are still dark (curved arrows) x400. e) Celeco CA1; pyramidal cells with round vesicular nuclei (P), shrunken and deeply stained cells are seen (curved arrows) x400. f) RIVA+Celeco CA1; most pyramidal cells (P) are well arranged and have round vesicular nuclei, dark cells (curved arrow) x400. g) Statistical analysis of the number of dark cells in CA1 fields. The data are expressed as mean ± SEM. * $p < 0.05$, **** $p < 0.0001$.

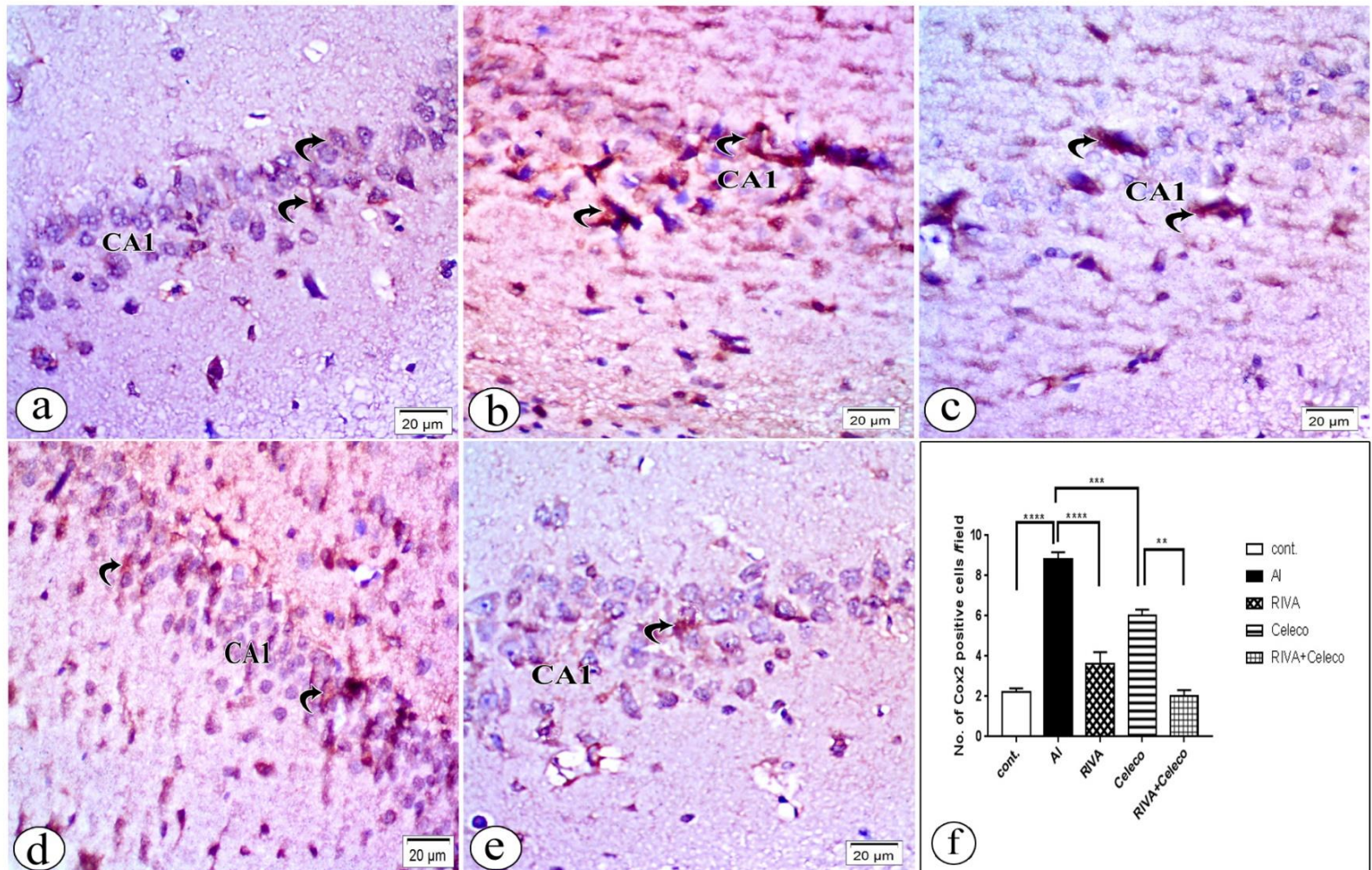


Fig 5. Photomicrographs of Cox-2 immunostained sections of the rat hippocampus (CA1 field). a) Cont CA1; few Cox-2 immunostained cells (curved arrows) x400. b) AI CA1; multiple Cox-2 immunostained cells (curved arrows) x400. c) RIVA CA1; some Cox-2 immunostained cells (curved arrows). d) Celeco CA1; few Cox-2 immunostained cells (curved arrows) x400. e) RIVA+Celeco CA1; few Cox-2 immunostained cells (curved arrow) x400. f) Statistical analysis of the number of Cox-2 immunostained cells in CA1 fields. The data are expressed as mean \pm SEM. *** $p < 0.001$, **** $p < 0.0001$.

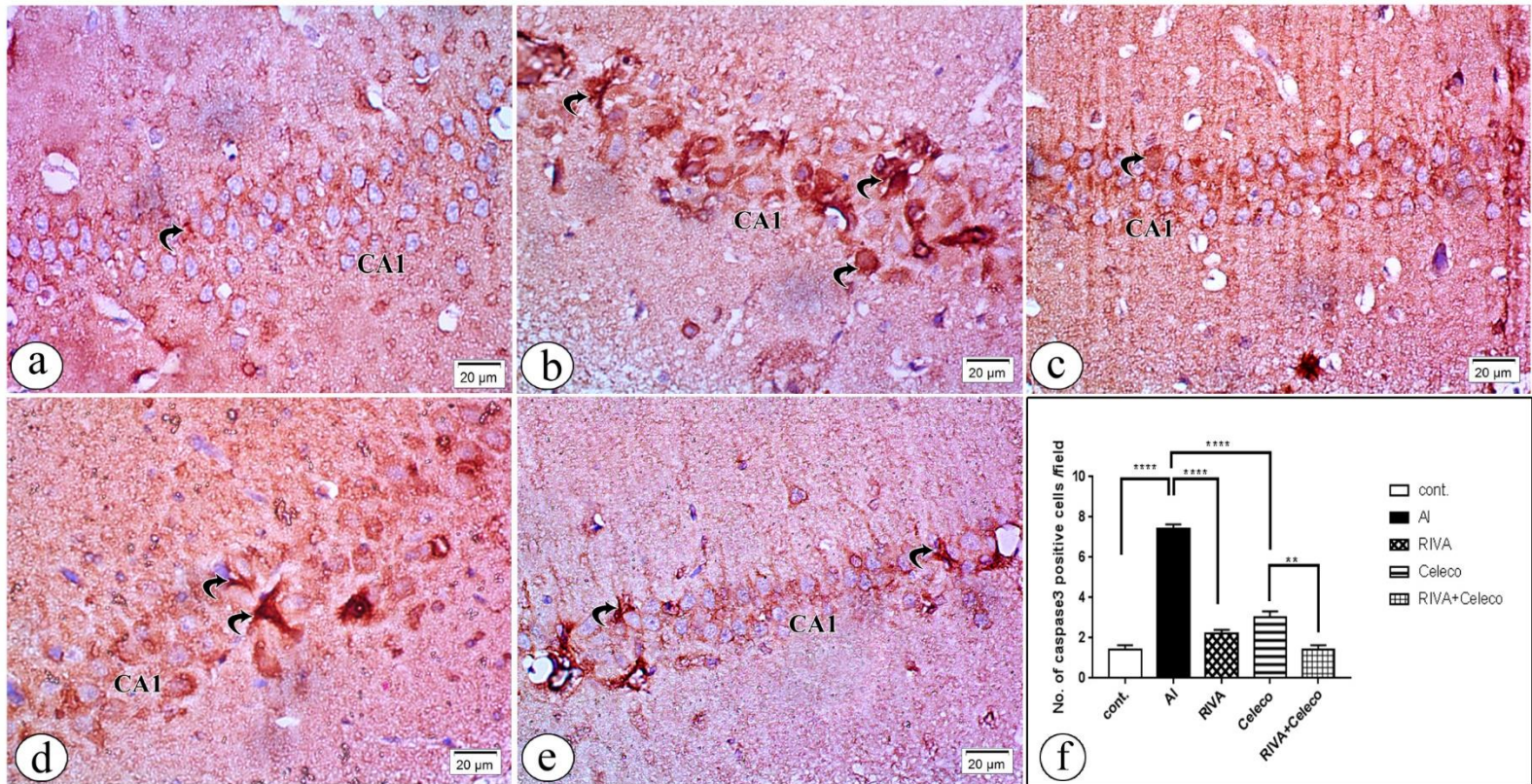


Fig 6. Photomicrographs of active caspase-3 immunostained sections of the rat hippocampus (CA1 field). a) Cont CA1; few caspase-3 immunostained cells (↑) x400. b) AI CA1; multiple caspase-3 immunostained cells (↑) x400. c) RIVA CA1; few caspase-3 immunostained cells (↑) x400. d) Celeco CA1; few caspase-3 immunostained cells (↑) x400. e) RIVA+Celeco CA1; few caspase-3 immunostained cells (↑) x400. f) Statistical analysis of the number of caspase-3 immunostained cells in CA1 fields. The data are expressed as mean ± SEM. * $p < 0.05$, **** $p < 0.0001$.

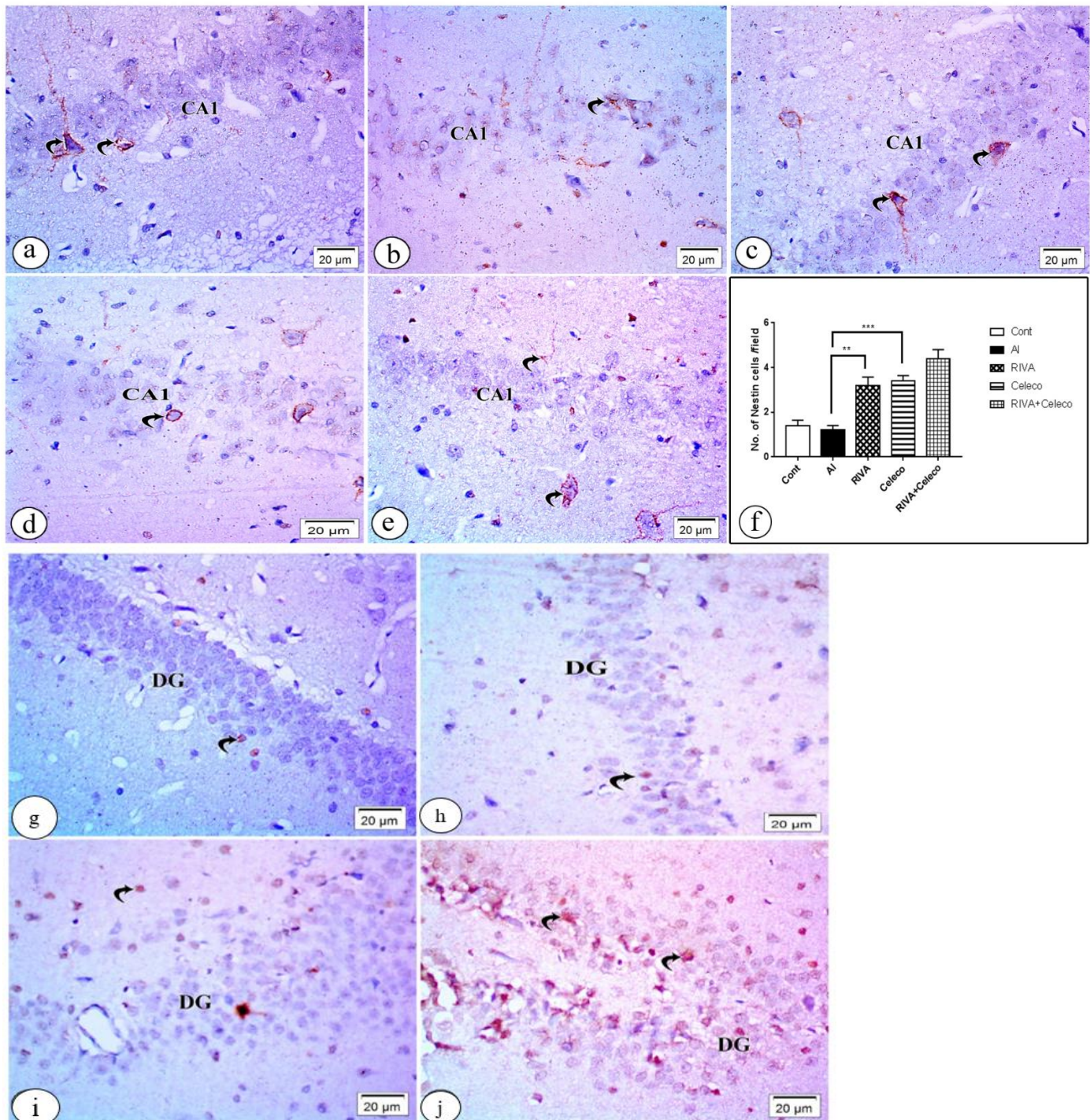


Fig 7. Photomicrographs of nestin immunostained sections of the rat hippocampus (CA1 & DG fields). **a)** Cont CA1; nestin immunostained cells, especially in glial cells (curved arrows) x400. **b)** AI CA1; few nestin immunostained cells (curved arrows) x400. **c)** RIVA CA1; multiple nestin immunostained cells (curved arrows) x400. **d)** Celeco CA1; multiple nestin immunostained cells (curved arrows) x400. **e)** RIVA+Celeco CA1; multiple nestin immunostained cells (curved arrows) x400. **f)** Statistical analysis of the number of nestin immunostained cells in CA1 fields. The data are expressed as mean \pm SEM. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$. **g)** Cont DG; nestin immunostained cells (curved arrows) x400. **h)** AI DG; few nestin immunostained cells (curved arrows) x400. **i)** RIVA DG; few nestin immunostained cells (curved arrows) x400. **j)** RIVA+Celeco DG; multiple nestin immunostained cells (curved arrows) x400.

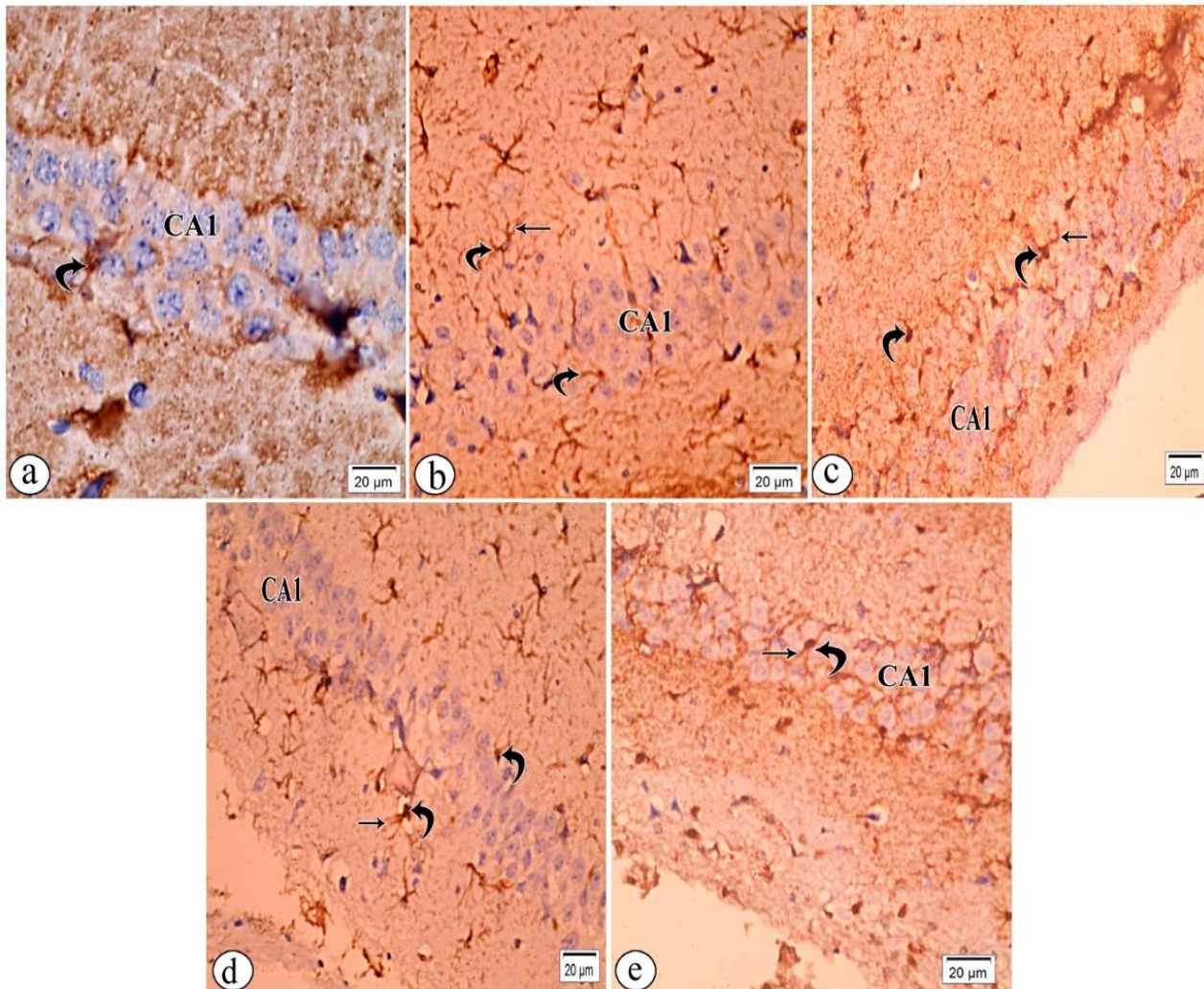


Fig 8. Photomicrographs of GFAP immunostained sections of the rat hippocampus (CA1 field). a) Cont CA1; few immunostained cells (curved arrows) x400. b) Al CA1; multiple GFAP immunostained cells (curved arrows) with long processes (↑) x400. c) RIVA CA1; multiple GFAP immunostained cells (curved arrows) x400. d) Celeco CA1; multiple GFAP immunostained cells (curved arrows) with long processes (↑) x400. e) RIVA+Celeco CA1; multiple GFAP immunostained cells (curved arrows) x400.

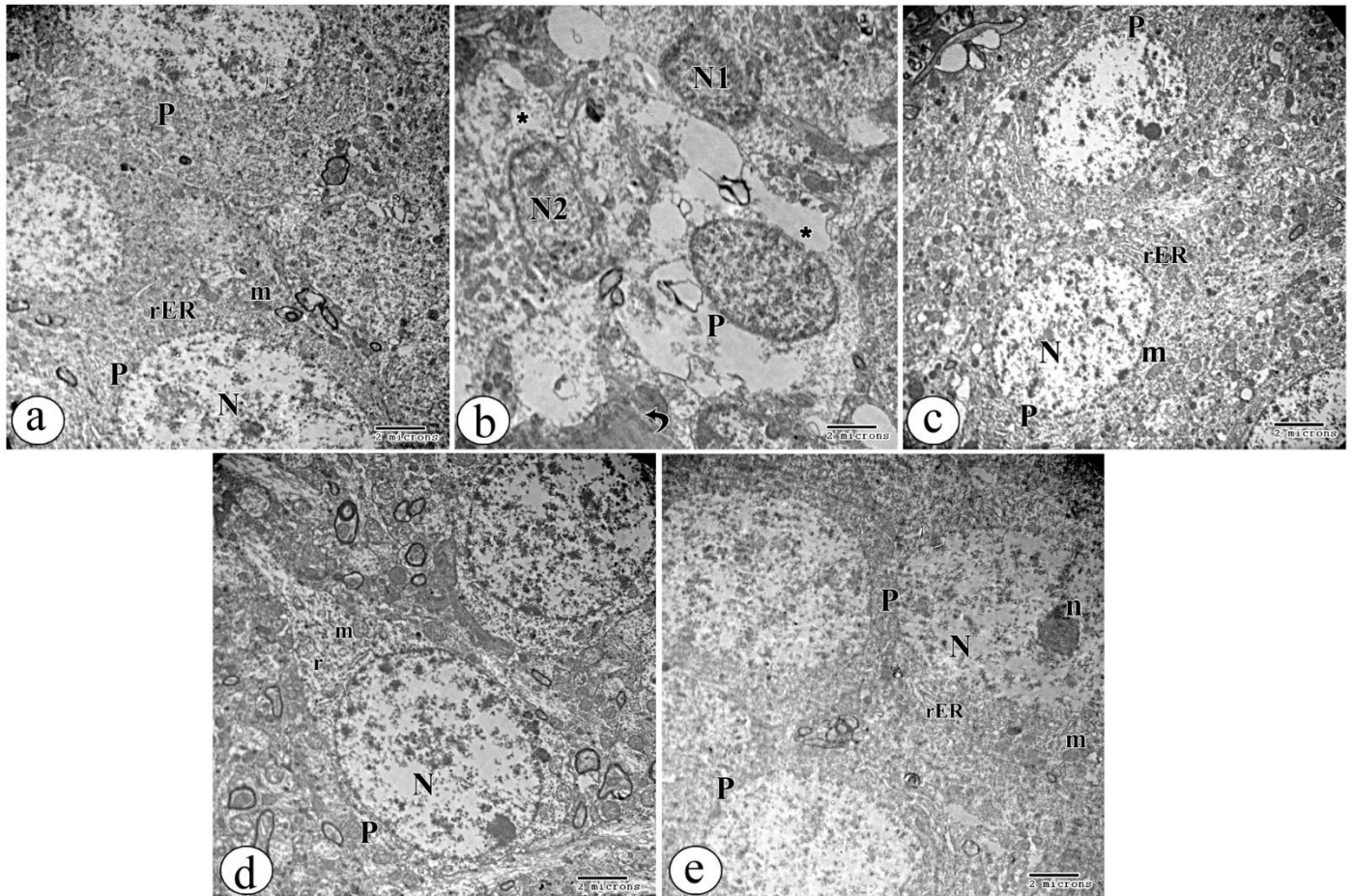


Fig 9. Transmission electron micrographs (TEM) of the rat hippocampus (CA1 fields; pyramidal cells). a) Cont pyramidal cells; multiple medium-sized pyramidal cells (P) with large oval nuclei (N) and abundant cytoplasm that contains mitochondria (m) and rER x3600. b) Al pyramidal cells; multiple pyramidal cells (P) with small involute oval nuclei (N1&N2) contain heterochromatin and rarefied cytoplasm (*). Electron-dense cells can be detected x3600. c) RIVA pyramidal cells; a group of pyramidal cells (P) with euchromatic oval or rounded nuclei (N), abundant electrolucent cytoplasm containing mitochondria (m) and rER x2900. d) Celecox pyramidal cells; electrolucent pyramidal cell (P), nuclei (N), mitochondria (m), and ribosomes (r) x3600. e) RIVA+Celecox pyramidal cells; a group of electrolucent pyramidal cells (P) with oval nuclei (N), mitochondria (m), and rER x3600.

Highlights

- AIC13 Alzheimer's rat model represents the sporadic type, the most common.
- Rivastigmine, a cholinesterase inhibitor, is the standard anti-Alzheimer drug.
- Celecoxib, a selective COX-2 inhibitor, ameliorated cholinergic dysfunction and mitigated behavioral insults, and hippocampal neuropathology in the Alzheimer's rat model.
- When given concurrently with rivastigmine, celecoxib improved rivastigmine anti-Alzheimer activity at the cellular level, enhanced neurogenesis & suppresses apoptosis.
- Celecoxib given concurrently with rivastigmine did not have an extra mitigating effect on Alzheimer's rat cognitive impairment compared to rivastigmine-only therapy.

4. Discussion

The current study was the first to display the celecoxib additive neuroprotective impact when combined with rivastigmine, the standard anti-Alzheimer drug, in the sporadic AD; A β 13-induced Alzheimer's rat model. Celecoxib enhanced the anti-Alzheimer activity of rivastigmine by restoring the AD-disrupted neuronal turnover through suppressing apoptosis, caspase-3 downregulation, and enhancing neurogenesis; nestin upregulation consequent to its anti-inflammatory effect. Even though celecoxib reduced cognitive deficits and enhanced memory efficiency in the AD rat model, it could not confer an additive impact when paired with rivastigmine compared to rivastigmine alone therapy, implying that it should be given earlier or for a longer period.

AD is a common irreversible, progressive neurodegenerative disorder manifested by gradual loss of memory, judgment, visuospatial skill, and executive functions [28,29]. Complex behavioral and psychological problems develop as the illness progresses, posing a significant burden on the patients, their caregivers, and society [30]. Collectively, AD typically progresses slowly in three general stages: (1) the asymptomatic "preclinical" stage, (2) mild cognitive impairment, which manifests as changes in mood commonly associated with confusion, and some memory loss, and (3) dementia, which exhibits multiple cognitive domains significant enough to cause loss of function [31–33]. The currently available medications merely ease the AD-associated symptoms. Curative therapies, therefore, are yet to be discovered.

AD pathology often begins several years preceding the onset of the clinical signs [34,35] and is already irreversible at diagnosis [17]. Extracellular senile plaques formed from deposits of hydrophobic A β peptides and intracellular aggregates of NFTs are the disease hallmark [36]. A β is the cleavage product of amyloid precursor protein (APP) via α -, β -, and γ -

secretases [37]. The accumulated β -amyloid plaques function as a toxin for neurons, disrupting neuron-neuron contact [38]. Furthermore, tau tangles cause poor nutrient transport triggering AD-associated neuronal loss [38].

Synaptic dysfunction and eventually neurodegeneration, particularly within the hippocampus, the entorhinal, the polymodal association cortices, and the basal forebrain [39], are common AD associations. Early in the disease, cholinergic neuron loss, particularly within the basal forebrain, occurs and is a major contributor to cognitive impairment [40–42], besides being strongly linked to the severity of dementia [43,44].

According to the cholinergic hypothesis, the onset of AD symptoms is primarily caused by structural changes in cholinergic synapses, the loss of particular subtypes of acetylcholine (ACh) receptors, the death of ACh-producing neurons with consequent weakening of cholinergic neurotransmission [45]. As a result, the ACh-hydrolyzing enzyme acetylcholinesterase (AChE) accumulates. Therefore, the enzyme activity can be used as a marker for the central cholinergic status [46–55]. Jha et al. [56] showed a marked decrease in AChE activity with increasing age. However, intense AChE activity appears in the neuritic plaques and neurofibrillary tangles [57]. AChE can promote A β assembly [58] and form stable complexes with A β fibrils [59] that are more toxic than A β fibrils alone [60]. A significant correlation between AChE inhibition and cognitive improvement in AD patients was observed [51]. Therefore, AChEIs [2] are significant of the currently approved AD medications. Rivastigmine is a unique cholinesterase inhibitor (ChEIs) with both AChE and butyrylcholinesterase inhibitory activity [61,62]. It displays specific activity for the central AChE over the peripheral one [63]. Rivastigmine tends to have the same effectiveness in inhibiting cholinesterases (ChEs) in plaques and tangles as it

does in neurons and axons [64]. It provides only symptomatic relief and a moderate disease-modifying effect as it can dramatically reduce agitated behavior and improve cognitive tasks in patients with dementia [65].

Al-induced neurotoxicity and its link to AD pathophysiology have been reported [66–70]. Al is considered the third most abundant element in the earth's crust, with high human exposure anticipated by cooking tools, food antacids, and various industrial applications [71] to produce rubber, lubricants, paints, wood preservatives, pesticides, antiperspirants, and pharmaceuticals. Al is a potentially dangerous neurotoxic metal that can pass through the blood-brain barrier through the iron-binding protein transferrin and is widely distributed in different brain areas [43]. Higher levels of Al have been found in the brains of AD patients, especially in the hippocampus, indicating that it may play a role in the disease progression [72,73]. Al has been found in senile plaques and NFT-bearing neurons [74] and is particularly abundant in areas rich in cholinergic synapses [75]. It has a robust ability to induce epigenetic changes [76] due to its strong affinity for interaction with biological molecules, including DNA, RNA, and proteins [77]. Al can induce changes in APP gene expression and cause misfolding of cytoskeleton protein, which contributes to increased APP transcription and enhanced A β plaque and neurofibrillary tangles deposition in the brain [78–81]. Collectively, Al has been linked to oxidative brain damage, neuronal death, cholinergic neuron degradation, and amyloid deposition, all of which have been associated with intellectual disability in AD cases [82–84].

Al is considered a potent cholinotoxin, causing increased ChE activities [85,86] with consequent impairment of the brain cholinergic transmission. After being exposed to low levels of Al, various mouse brain regions showed a rise in AChE activity [87]. AICl₃ Alzheimer rat model may experimentally

simulate the critical aspects of AD pathology and disease processes [17]; hence, it is widely accepted that AICl₃ can be used to induce neurodegenerative changes in animals to mimic AD [88].

Our findings revealed a substantial increase in hippocampal AChE activity in the AD rat, consistent with other studies [89,90] indicating significant cognitive deficits. According to Gulya et al. [91], an increase in AChE activity after Al exposure is due to the allosteric interaction between cation (Al⁺³) and anionic sites of AChE, which leads to alteration in its secondary structure and activity. The second proposed mechanism [92] is Al's ability to facilitate the accumulation of insoluble A β (1-42) [93], which has a direct inhibitory effect on nicotinic acetylcholine receptors (nAChR) with consequent enhancement of AChE activity. On the other hand, other research found decreased AChE activity in the cerebellum and hippocampus on long-term exposure to Al [94]. The conflicting results could be attributed to the various molecular forms of AChE or the Al biphasic influence on the AChE activity [94] mediated through the formation of reversible/irreversible Al complex [94]. Rivastigmine inhibited AICl₃-enhanced hippocampal AChE activity, resulting in a partial enhancement in cholinergic neurotransmission, which is in line with previous studies [95–97]. However, in advanced cases, rivastigmine works by inhibiting butyrylcholinesterase (BuChE) activity [98,99].

Our study showed that Al-intoxicated rats had a weak exploratory preference, impaired spatial and retention memory, and shortened step-through latency as assessed by NOR, MWM, and PA tests, consistent with previous literature [25]. AICl₃-treated animals have displayed increased retention latency and decreased RI percentage [100], indicating reduced short, intermediate, and long-term memory [101,102]. Intracerebral AICl₃ administration caused learning deficits in rabbits (93). Rivastigmine improved spatial and retrieval memory impairments caused by AICl₃

administration, which is in line with previous studies (94). It has reversed the scopolamine-induced spatial learning deficits as well [103]. RIVA group spent considerably more time investigating the novel object (5 min & 2 hrs). It exhibited a significant increase in step-through latency than the AI group reflecting improvement in learning and memory tasks. Decreased escape latency and increased discrimination index reported in the streptozotocin rat model of AD have been attenuated through rivastigmine administration [104]. Moreover, rivastigmine exerted positive effects on learning and memory in the chronic d-galactose-induced accelerated aging rodent model [105].

The hippocampus is considered the neurobiological root [106] of spatial learning, working, and episodic memory skills, which deteriorate with age progression [107]. DG, CA3, and CA1 are the three distinct subregions of the hippocampus. Hippocampal hyperactivity has been identified in people who have a genetic or familial risk of AD [108,109] and asymptomatic and minimally impaired older individuals with A β deposition [110]. Its degree correlated with memory decline [111] and was previously thought to be a compensatory reaction for worsening neuronal circuitry [108]. New research, however, indicates that hippocampal hyperactivity may be a sign of neuronal excitotoxicity, a pathological mechanism in which neurons are destroyed by excessive activation. Since CA1 neurons are highly sensitive to excitotoxicity and more vulnerable to loss in AD, they are perhaps the most studied of all the hippocampal subregions in rodents [112]. Our findings revealed that AI-intoxicated rats had a substantial increase in degenerated pyramidal cells within the CA1 region, which appeared shrunken with ill-defined organelles and nuclei, which is in line with previous research [90,113,114]. The RIVA group revealed a rise in the number of regenerating pyramidal cells in line with

other research [115] confirming the rivastigmine mitigating impact on AIC13-induced oxidative damage and neurotoxicity.

Recent studies have uncovered the role of neuroinflammation and aberrant gliosis in AD [15]. When neuronal loss is still absent, pathological neuroinflammatory reactions, astrogliosis, and microglial activation were observed [116–118]. Microglia react more quickly to injury or pathological insult than astrocytes [119]. Once pathologic A β deposition begins, microglia activation or reactive astrogliosis may be required to improve clearance of excess toxic amyloid [120], thereby limiting A β plaques build-up [121,122] through the expression of type III intermediate filament (IF) protein; GFAP (a marker for astrogliosis) [123,124]. However, as plaques form, microglia and astrocyte stimulation can further worsen the disease [125,126]. They are known to release reactive oxygen species (ROS), nitric oxide (NO), and inflammatory cytokines that encourage low-grade neuroinflammation [127–129], enhance APP, A β production [130–132], and tau hyperphosphorylation. Microglia and astrocytes' role, therefore, in AD is still controversial. Nonetheless, the degree of astrogliosis is often linked to cognitive decline in AD [133–135].

In AIC13-intoxicated rats, we detected an enhanced hippocampal GFAP immunoreactivity, indicating extreme astrocytic activation, probably, induced by AI uptake. Our findings are in line with those of other studies [136–139], which found increased GFAP immunolabelling in the hippocampus after chronic AI intoxication in rats and rabbits [140]. However, unchanged GFAP immunoreactivity was still reported [141]. In contrast, a decrease in GFAP immunoreactivity was detected, reflecting astrocytes' vulnerability to AI-induced neurotoxicity [114,136]. The RIVA group showed enhanced GFAP immunoreactivity indicating persistent astrogliosis, which contradicts other studies [142] that displayed

reduced immunoreactivity by 45–50%. Rivastigmine has also been shown to reduce memory loss in T2DM-AD model mice by suppressing gliosis [143]. The diversity of the AD models could lie behind the conflicting outcomes.

NSAIDs and cyclooxygenase (COX) inhibitors have been studied to reduce the risk and help delay the progression of AD [144–149] by suppressing the associated inflammatory response [150–152]. They can lower the production of A β in senile plaques and phosphorylated tau in NFTs [120], signifying COX activity's involvement in the cascade of AD development. Both COX isoforms, COX-1 and COX-2, were found to be constitutively expressed in the brain [153,154]. COX-2 expression is the highest in the hippocampus and cerebral cortex, suggesting the enzyme's role in the regulation of the plasticity mechanisms that sustain learning and memory processes [155,156]. Preclinical studies have shown that treatment with specific COX-2 inhibitors impairs cognitive functions in hippocampal-dependent paradigms [157].

On the other hand, a piece of evidence has indicated that COX-2 aberrant activation under pathological circumstances may cause the associated cognitive dysfunction [158,159]. COX-2 expression is rapidly upregulated in response to a variety of neurotoxic stimuli [155]. Therefore, it has been linked to the pathogenesis of neurodegenerative diseases like multiple sclerosis, Parkinson's disease, traumatic brain injury, ischemia-induced neuronal damage, epileptogenesis, and AD [160].

COX-2 is induced by various inflammatory molecules such as IL-1, IL-2, and TNF- α and is expressed differently in different stages of AD [161]. Previous literature has reported an association between COX-2 induction and the development of amyloid plaques [162] and neurodegeneration [163]. COX-2 overexpression decreased the learning ability of Tg mice by increasing the production of A β [164–

168]. It is considered the rate-limiting enzyme in the synthesis of prostaglandins (PGs) such as PGE₂, PGD₂, PGI₂, PGF₂, and thromboxane A₂ (TXA₂), which are important components in brain cell damage associated with AD [169]. Activated microglia have been identified as the major producers of the prostaglandin PGE₂ via the COX-2 pathway [170–172]. COX-2 and PGE₂ regulate the expression of APP, α -, β -, and γ -secretases [120,173]. PGD₂ has been described as the most abundant eicosanoid in the brain [174,175]. Activation of the DP₂ receptor in astrocytes or Th₂ cells enhanced inflammatory cytokines production [176,177], which is crucial in the progression of AD [120].

COX-2 expression was significantly higher in the CA1 area of the AI group, which is consistent with other studies [137,178] that revealed elevated hippocampal COX-2 mRNA [179] and protein expression in correlation with amyloid plaque density [12]. COX-2 has been shown to have high expression within pyramidal neurons [11,180–184] in the early stages of AD, which could be related to regeneration and cell cycle regulation [167,185,186], suggesting that it may be used as a functional indicator of clinical dementia [180]. Whereas, as the disease progresses, the expression of COX-2 is observed to decrease, most likely due to selective degeneration of COX-2-expressing neurons and loss of synaptic activity [161]. These findings suggest that COX-2 may play a role in the early stages of AD but is unlikely to play a role later, emphasizing the importance of early intervention.

RIVA group showed a substantial decrease in COX-2 expression compared to the AI group, which is consistent with other studies showing a reduction of COX-1 and COX-2 mRNA and proteins expression within macrophages fluoride-influenced model [187] that may be attributed to rivastigmine anti-inflammatory properties. Its anti-inflammatory activity can be explained by its effect on nAChRs and

transmitter-gated ion channels involved in the cognitive processes [188–190].

The most appropriate AD prevention seems to be achieved by the specific inhibitors of COX-2, namely celecoxib and rofecoxib [191]. Selective COX-2 inhibitors can attenuate amyloid- β -mediated suppression of memory and synaptic plasticity by preventing PGE2 response at synapses [192]. Restoration of memory in Tg2576 mice overexpressing APP by selective COX-2 inhibitors was previously detected as well [192]. Patients with AD who were given celecoxib for 18 months and 4 to 5 years had their learning ability preserved [193–195]. However, in large-scale clinical trials, selective COX-2 inhibitors did not reveal any benefit in Alzheimer's patients [196–198]. A one-year regimen with celecoxib did not slow cognitive deterioration in 285 patients with AD [199].

Moreover, celecoxib has been reported to raise the level of the A β 1-42 segment in cell culture studies and the brains of transgenic and non-transgenic mice [200]. These conflicting results have recently been attributed to the hypothesis that the effects of COX-2 inhibitors may vary depending on the stage of AD development. Celecoxib is ineffective once the A β deposition begins. It may be detrimental due to its inhibitory effect on the chronically activated microglia, which may slow down A β clearance (38), accelerating the disease process [201]. Failure of selective COX-2 inhibitors to treat advanced stages of AD may be explained by the fact that the expression of the COX-2 enzyme in AD hippocampal tissue correlates with the disease severity, with increased immunoreactivity in early AD and decreased activity in advanced stages [155]. Our study revealed that the RIVA+Celeco group displayed significant suppression of COX-2 compared to the RIVA group. The concurrent administration of celecoxib with rivastigmine, therefore, can provide a further anti-

inflammatory effect that could probably enhance rivastigmine anti-Alzheimer activity.

Persistently enhanced hippocampal GFAP immunoreactivity was detected in the RIVA, Celeco, and RIVA+Celeco groups. On the contrary, other studies showed that celecoxib had reduced the number of activated astrocytes and the percentage of GFAP immunostaining, thus attenuating LPS-induced brain inflammation in neonatal rats [23,202]. The variations in the model and treatment period may be to blame for the conflicting results.

Celecoxib has been shown to inhibit the enzyme AChE in a non-competitive manner, possibly due to its aromatic structural motives [203]. In agreement with a previous study [204], our study showed that celecoxib suppressed AChE activity previously enhanced in Al-intoxicated rats that ensure its mitigating effect on learning and memory deficit [205]. When combined with rivastigmine, celecoxib had no additional AChE inhibitory effect relative to rivastigmine alone. Other studies [203], on the other hand, found that the celecoxib AChE inhibitory effect is too weak to trigger major AChE inhibition and that it can be used as an additive inhibitor to a standard medication such as rivastigmine.

Celecoxib attenuated the Al-induced cognitive deficit, which is in line with previous research [193,194]. Importantly, the Celeco group showed a decreased time to reach the platform, whereas increased step-through latency and time exploring novel objects implied enhanced spatial and retention memory in the AD rat model. Celecoxib did, however, impair memory acquisition and retrieval [206], probably mediated through late short-term celecoxib therapy, which increased A β -42 deposition. Importantly, our study found that the simultaneous administration of celecoxib with rivastigmine had no other mitigating impact on the memory and cognitive decline of the Al-intoxicated rats. Still, it did effectively attenuate Al-induced hippocampal

neurodegeneration compared to rivastigmine medication alone.

AD neuronal cell death can be attributed to apoptosis and DNA fragmentation [207–210]. Caspase-3 is considered the final executor of apoptosis [211] that mediates cytoskeletal and nuclear proteins [212]. Increased A β is likely to result in a higher likelihood of oligomer formation, intermediate assemblies, as soluble [213] as well as insoluble aggregates, that possess toxic effects [214–216] through several canonical apoptotic pathways, including caspase-3 [217]. Studies have revealed the apoptogenic role of AChE [218], which may lie behind the beneficial role of AChEIs in the early stages of AD [219]. Furthermore, PGE₂ and PGF₂ α have been shown to promote cortical neuronal apoptosis and damage (135, 136) during the late stages [120].

Al is thought to cause cell death by triggering apoptotic pathways mediated by stress processes in the mitochondria or endoplasmic reticulum [220]. In Al-intoxicated rats, the protein level of activated caspase-3 was significantly increased relative to the Cont group, consistent with other studies [137,221–226]. The RIVA group displayed a significant reduction of the activated caspase-3 expression compared to the AL group, consistent with other research findings [227,228]. Similarly, Al-mediated caspase-3 overexpression was significantly attenuated by celecoxib administration, which agrees with previous studies that showed celecoxib's ability to inhibit the A β -induced apoptosis of neurons through inhibiting PGD₂-induced apoptosis in AD [16]. Moreover, celecoxib was found to be neuroprotective against ethanol-induced neurodegeneration [229] and renoprotective against gentamicin-induced nephrotoxicity [230]. Caspase inhibition is recognized as a Cox-independent anti-inflammatory mechanism for NSAID drugs with a consequent decrease in cell

death and pro-inflammatory cytokine production [231].

On the contrary, celecoxib has been studied to have an antitumor and chemo-sensitizing effect [232,233] by enhancing caspase-3 activity. The conflicting results may be related to the variability in the tissue type, the nature of the experiment, the dose, and the duration of administration. Celecoxib decreased Al-induced activated caspase-3 overexpression significantly when combined with rivastigmine compared to rivastigmine alone, indicating that celecoxib may improve rivastigmine anti-Alzheimer role.

Recent studies have suggested the implication of neurogenesis in neurodegenerative disorders [234–237]. Deficits in adult neurogenesis may contribute to tau hyperphosphorylation in new neurons, compromised hippocampal circuitry, and cognitive impairments in AD [238]. Therefore, through pharmacological and genetic approaches, the induction of neurogenesis can slow down disease progression [239]. The neurogenesis process has been well acknowledged in two brain regions; the subventricular zone (SVZ) of the lateral ventricle and the subgranular zone (SGZ) of the hippocampus DG [234]. Several studies have shown that impaired neurogenesis within the hippocampus is linked to the development of AD [240,241]. The disturbance of the equilibrium between neurogenesis and neurotoxicity caused by COX-2 and PG expression hastens the development of AD [126]. Conversely, It has been reported that the proliferation of neuroprogenator stem cells (NPCs) is increased in the hippocampus of AD patients [242] early in the disease as a compensatory mechanism for A β -induced apoptosis [243]. Therefore, enhancing neurogenesis in the earliest stages of AD could provide a potentially powerful disease-modifying treatment strategy for AD [244].

Nestin, a type VI intermediate filament, is considered an important component of the

cytoskeleton and is thought to be a marker for neural stem cells; neurogenesis [245]. Our study showed a significant reduction of nestin immunoreactivity in the AI group, indicating impaired cellular proliferation compared to the Cont group which is consistent with other studies [246–248]. However, concomitant administration of rivastigmine increased nestin immunoreactivity, presumably, due to the compensation for the cholinergic deficits [249]. Similarly, other research results [250] showed enhanced expression of the brain nestin gene by 65.2% through rivastigmine administration relative to the untreated AD population. Similarly, celecoxib administration-induced nestin protein overexpression may represent a therapeutic option to restore adult neurogenesis in AD patients. Other studies, similarly, revealed celecoxib's ability to enhance neurogenesis and reduce apoptosis in APP/PS1 transgenic mice [16] and alleviate A β -reduced neurogenesis in the transgenic Alzheimer mouse model [16]. Previous studies [251] showed that celecoxib increased the number of neural stem cells in the lesion zone in spontaneous intracerebral hemorrhage. When combined with rivastigmine, celecoxib significantly increased nestin expression in AI-intoxicated rats relative to rivastigmine alone, suggesting that celecoxib can boost neurogenesis and possibly enhance rivastigmine anti-Alzheimer activity.

Conclusion

AD is the primary cause of dementia in the middle-aged and elderly worldwide. Rivastigmine, the standard anti-Alzheimer drug, can only provide symptomatic relief and has a moderate disease-modifying effect. COX-2 overexpression has been related to the formation of senile amyloid plaques and subsequent neurodegeneration, highlighting the role of neuroinflammation in AD pathogenesis. Our study displayed that celecoxib, a selective COX-2 inhibitor, has attenuated cognitive deficits and improved memory performance in the AIC13-induced

Alzheimer's rat model. On the other hand, celecoxib was unable to boost rivastigmine anti-Alzheimer activity when given concurrently, despite being able to restore AD-disrupted neuronal turnover via suppressing apoptosis and enhancing neurogenesis relative to rivastigmine alone medication. More study is needed to reveal whether celecoxib, when given over a longer period or earlier in the course of the disease, may increase the efficacy of rivastigmine in treating Alzheimer's disease.

Limitations of the study

Our study has several limitations. First, we had better study other signaling mechanisms that may be implicated in celecoxib neuroprotective effect as peroxisome proliferator-activated receptor γ (PPAR γ), γ -secretase enzyme, NF- κ B, and mTOR. Second, to test its comparable efficacy, we should have a study group that receives celecoxib prophylactically in a rivastigmine-treated Alzheimer's rat model.

Conflict of interest

The authors declare that they have no conflict of interest.

Acknowledgments

This work was supported by Assiut Medical School Grants Office (grant code 20171121002R1) at Assiut University, Assiut, Egypt.

References

- [1] A.P. Reddy, J. Ravichandran, N. Carkaci-Salli, Neural regeneration therapies for Alzheimer's and Parkinson's disease-related disorders, *Biochim. Biophys. Acta - Mol. Basis Dis.* 1866 (2020) 165506.
- [2] T.C. Russ, J.R. Morling, Cholinesterase inhibitors for mild cognitive impairment, *Cochrane Database Syst. Rev.* 2012 (2012).
- [3] B. Ray, D.K. Lahiri, Neuroinflammation in Alzheimer's disease: different molecular targets and potential therapeutic agents including curcumin, *Curr. Opin. Pharmacol.* 9

- (2009) 434–444.
- [4] A. Nazem, R. Sankowski, M. Bacher, Y. Al-Abed, Rodent models of neuroinflammation for Alzheimer's disease, *J. Neuroinflammation*. 12 (2015) 74.
- [5] R. Dingledine, K. Borges, D. Bowie, S.F. Traynelis, The Glutamate Receptor Ion Channels, *Pharmacol. Rev.* 51 (1999) 7–61.
- [6] M. Citron, Alzheimer's disease: strategies for disease modification, *Nat. Rev. Drug Discov.* 9 (2010) 387–398.
- [7] E. Alteri, L. Guizzaro, Be open about drug failures to speed up research, *Nature*. 563 (2018) 317–319.
- [8] V.L. Villemagne, S. Burnham, P. Bourgeat, B. Brown, K.A. Ellis, O. Salvado, C. Szoëke, S.L. Macaulay, R. Martins, P. Maruff, D. Ames, C.C. Rowe, C.L. Masters, Amyloid β deposition, neurodegeneration, and cognitive decline in sporadic Alzheimer's disease: A prospective cohort study, *Lancet Neurol.* 12 (2013) 357–367.
- [9] A. Awasthi, Y. Matsunaga, T. Yamada, Amyloid-beta causes apoptosis of neuronal cells via caspase cascade, which can be prevented by amyloid-beta-derived short peptides, *Exp. Neurol.* 196 (2005) 282–289.
- [10] T. Toda, S.L. Parylak, S.B. Linker, F.H. Gage, The role of adult hippocampal neurogenesis in brain health and disease, *Mol. Psychiatry*. 24 (2019) 67–87.
- [11] G.M. Pasinetti, P.S. Aisen, Cyclooxygenase-2 expression is increased in frontal cortex of Alzheimer's disease brain, *Neuroscience*. 87 (1998) 319–24.
- [12] L. Ho, C. Pieroni, D. Winger, D.P. Purohit, P.S. Aisen, G.M. Pasinetti, Regional distribution of cyclooxygenase-2 in the hippocampal formation in Alzheimer's disease, *J Neurosci Res*. 57 (1999) 295–303.
- [13] M.G. Giovannini, C. Scali, C. Prosperi, A. Bellucci, G. Pepeu, F. Casamenti, Experimental brain inflammation and neurodegeneration as model of Alzheimer's disease: protective effects of selective COX-2 inhibitors, *Int J Immunopathol Pharmacol.* 16 (2003) 31–40.
- [14] E.G. McGeer, P.L. McGeer, The importance of inflammatory mechanisms in Alzheimer disease, *Exp Gerontol.* 33 (1998) 371–8.
- [15] B.P. Imbimbo, V. Solfrizzi, F. Panza, Are NSAIDs useful to treat Alzheimer's disease or mild cognitive impairment?, *Front. Aging Neurosci.* 2 (2010) 19.
- [16] J.W. Guo, P.P. Guan, W.Y. Ding, S.L. Wang, X.S. Huang, Z.Y. Wang, P. Wang, Erythrocyte membrane-encapsulated celecoxib improves the cognitive decline of Alzheimer's disease by concurrently inducing neurogenesis and reducing apoptosis in APP/PS1 transgenic mice, *Biomaterials*. 145 (2017) 106–127.
- [17] X. Li, X. Bao, R. Wang, Experimental models of Alzheimer's disease for deciphering the pathogenesis and therapeutic screening (Review), *Int. J. Mol. Med.* 37 (2016) 271–283.
- [18] P.A. Evrard, C. Ragusi, G. Boschi, R.K. Verbeeck, J.M. Scherrmann, Simultaneous microdialysis in brain and blood of the mouse: extracellular and intracellular brain colchicine disposition., *Brain Res.* 786 (1998) 122–127.
- [19] R.A. Abdel-Aal, A.A.A. Assi, B.B. Kostandy, Memantine prevents aluminum-induced cognitive deficit in rats, *Behav. Brain Res.* 225 (2011) 31–38.
- [20] L. Zhou, S. Tan, Y.-L. Shan, Y.-G. Wang, W. Cai, X.-H. Huang, X.-Y. Liao, H.-Y. Li, L. Zhang, B.-J. Zhang, Z.-Q. Lu, Baicalein improves behavioral dysfunction induced by Alzheimer's disease in rats, *Neuropsychiatr.*

- Dis. Treat. 12 (2016) 3145–3152.
- [21] M.F. Ismail, A.N. Elmeshad, N.A.-H. Salem, Potential therapeutic effect of nano based formulation of rivastigmine on rat model of Alzheimer's disease., *Int. J. Nanomedicine*. 8 (2013) 393–406.
- [22] Y. Yang, L. Gao, Celecoxib Alleviates Memory Deficits by Downregulation of COX-2 Expression and Upregulation of the BDNF-TrkB Signaling Pathway in a Diabetic Rat Model, *J Mol Neurosci*. 62 (2017) 188–198.
- [23] A. Kaizaki, L.-T. Tien, Y. Pang, Z. Cai, S. Tanaka, S. Numazawa, A.J. Bhatt, L.-W. Fan, Celecoxib reduces brain dopaminergic neuronal dysfunction, and improves sensorimotor behavioral performance in neonatal rats exposed to systemic lipopolysaccharide., *J. Neuroinflammation*. 10 (2013) 45.
- [24] M. Antunes, G. Biala, The novel object recognition memory: Neurobiology, test procedure, and its modifications, *Cogn. Process*. 13 (2012) 93–110.
- [25] B.V.S. Lakshmi, M. Sudhakar, K.S. Prakash, Protective Effect of Selenium Against Aluminum Chloride-Induced Alzheimer's Disease: Behavioral and Biochemical Alterations in Rats, *Biol. Trace Elem. Res*. 165 (2015) 67–74.
- [26] H. Abdi-Azar, S. Maleki, Comparison of the anesthesia with thiopental sodium alone and their combination with Citrus aurantium L. (Rutaseae) essential oil in male rat, *Bull. Environ. Pharmacol. Life Sci*. 3 (2014) 37–44.
- [27] J.D. Bancroft, M. Gamble, Theory and Practice of Histological Techniques, 6th Edition, *J. Neuropathol. Exp. Neurol*. 67 (2008) 633.
- [28] B. BF, Mild cognitive impairment associated with underlying Alzheimer's disease versus Lewy body disease, *Park. Relat Disord*. 18 (2012) S41–4.
- [29] F. Sá, P. Pinto, C. Cunha, R. Lemos, L. Letra, M. Simões, I. Santana, Differences between early and late-onset Alzheimer's disease in neuropsychological tests, *Front Neurol*. 3 (2012) 81.
- [30] J. Hort, J.T. O'Brien, G. Gainotti, T. Pirttila, B.O. Popescu, I. Rektorova, S. Sorbi, P. Scheltens, EFNS guidelines for the diagnosis and management of Alzheimer's disease, *Eur J Neurol*. 17 (2010) 1236–48.
- [31] H. Förstl, A. Kurz, Clinical features of Alzheimer's disease, *Eur Arch Psychiatry Clin Neurosci*. 249 (1999) 288–90.
- [32] C.R.J. Jr, D.S. Knopman, W.J. Jagust, L.M. Shaw, P.S. Aisen, M.W. Weiner, R.C. Petersen, J.Q. Trojanowski, Hypothetical model of dynamic biomarkers of the Alzheimer's pathological cascade, *Lancet Neurol*. 9 (2010) 119–28.
- [33] R.A. Sperling, P.S. Aisen, L.A. Beckett, D.A. Bennett, S. Craft, A.M. Fagan, T. Iwatsubo, C.R.J. Jr, J. Kaye, T.J. Montine, D.C. Park, E.M. Reiman, C.C. Rowe, E. Siemers, Y. Stern, K. Yaffe, M.C. Carrillo, B. Thies, M. Morrison-Bogorad, M. V Wagster, C.H. Phelps, Toward defining the preclinical stages of Alzheimer's disease: recommendations from the National Institute on Aging-Alzheimer's Association workgroups on diagnostic guidelines for Alzheimer's disease, *Alzheimers Dement*. 7 (2011) 280–92.
- [34] R.J. Bateman, C. Xiong, T.L.S. Benzinger, A.M. Fagan, A. Goate, N.C. Fox, D.S. Marcus, N.J. Cairns, X. Xie, T.M. Blazey, D.M. Holtzman, A. Santacruz, V. Buckles, A. Oliver, K. Moulder, P.S. Aisen, B. Ghetti, W.E. Klunk, E. McDade, R.N. Martins, C.L. Masters, R. Mayeux, J.M. Ringman, M.N. Rossor, P.R. Schofield, R.A. Sperling, S.

- Salloway, J.C. Morris, Clinical and biomarker changes in dominantly inherited Alzheimer's disease, *N Engl J Med.* 367 (2012) 795–804.
- [35] A.S. Fleisher, K. Chen, Y.T. Quiroz, L.J. Jakimovich, M.G. Gomez, C.M. Langois, J.B.S. Langbaum, N. Ayutyanont, A. Roontiva, P. Thiyyagura, W. Lee, H. Mo, L. Lopez, S. Moreno, N. Acosta-Baena, M. Giraldo, G. Garcia, R.A. Reiman, M.J. Huentelman, K.S. Kosik, P.N. Tariot, F. Lopera, E.M. Reiman, Flortetapir PET analysis of amyloid- β deposition in the presenilin 1 E280A autosomal dominant Alzheimer's disease kindred: a cross-sectional study, *Lancet Neurol.* 11 (2012) 1057–65.
- [36] J. KA, B. C, Neuropathology of Alzheimer's disease: a critical update, *J Neural Transm.* 54 (1998) 77–95.
- [37] R.J. O'Brien, P.C. Wong, Amyloid precursor protein processing and Alzheimer's disease, *Annu Rev Neurosci.* 34 (2011) 185-204.
- [38] T. Ali, G.H. Yoon, S.A. Shah, H.Y. Lee, M.O. Kim, Osmotin attenuates amyloid beta-induced memory impairment, tau phosphorylation and neurodegeneration in the mouse hippocampus, *Sci. Rep.* 5 (2015) 1–17.
- [39] A. Serrano-Pozo, M.P. Frosch, E. Masliah, B.T. Hyman, Neuropathological alterations in Alzheimer disease, *Cold Spring Harb Perspect Med.* 1 (2011) a006189.
- [40] D.J. Selkoe, Alzheimer's disease is a synaptic failure, *Science* (80-.). 298 (2002) 789–791.
- [41] T.P. Wong, T. Debeir, K. Duff, A.C. Cuello, Reorganization of cholinergic terminals in the cerebral cortex and hippocampus in transgenic mice carrying mutated presenilin-1 and amyloid precursor protein transgenes, *J. Neurosci.* 19 (1999) 2706–2716.
- [42] S. Chandra, M. Jana, K. Pahan, Aspirin Induces Lysosomal Biogenesis and Attenuates Amyloid Plaque Pathology in a Mouse Model of Alzheimer's Disease via PPAR α , *J. Neurosci.* 38 (2018) 6682–6699.
- [43] E. Elmorsy, E. Elsharkawy, F.A. Alhumaydhi, M. Salama, The protective effect of Indian Catechu methanolic extract against aluminum chloride-induced neurotoxicity, A rodent model of Alzheimer's disease, *Heliyon.* 7 (2021) e06269.
- [44] H. Hampel, M.-M. Mesulam, A.C. Cuello, A.S. Khachaturian, A. Vergallo, M.R. Farlow, P.J. Snyder, E. Giacobini, Z.S. Khachaturian, Revisiting the Cholinergic Hypothesis in Alzheimer's Disease: Emerging Evidence from Translational and Clinical Research, *J Prev Alzheimers Dis.* 6 (2019) 2–15.
- [45] G.D. Stanciu, A. Luca, R.N. Rusu, V. Bild, S.I.B. Chiriac, C. Solcan, W. Bild, D.C. Ababei, Alzheimer's Disease Pharmacotherapy in Relation to Cholinergic System Involvement, *Biomolecules.* 10 (2020) 40.
- [46] R.H. Perry, I.D. Wilson, M.J. Bober, J. Atack, G. Blessed, B.E. Tomlinson, E.K. Perry, Plasma and erythrocyte acetylcholinesterase in senile dementia of Alzheimer type, *Lancet.* 1 (1982) 174–5.
- [47] J.R. Atack, E.K. Perry, R.H. Perry, I.D. Wilson, M.J. Bober, G. Blessed, B.E. Tomlinson, Blood acetyl- and butyrylcholinesterases in senile dementia of Alzheimer type, *J Neurol Sci.* 70 (1985) 1–12.
- [48] J. Sirviö, R. Kutvonen, H. Soininen, P. Hartikainen, P.J. Riekkinen, Cholinesterases in the cerebrospinal fluid, plasma, and erythrocytes of patients with Alzheimer's disease, *J Neural Transm.* 75 (1989) 119–27.
- [49] T. Darreh-Shori, O. Almkvist, Z.Z. Guan, A. Garlind, B. Strandberg, A.-L. Svensson, H. Soreq, E. Hellström-Lindahl, A. Nordberg, Sustained cholinesterase inhibition in AD

- patients receiving rivastigmine for 12 months, *Neurology*. 59 (2002) 563–72.
- [50] Y. Yamamoto, S. Nakano, S. Kawashima, S. Nakamura, K. Urakami, T. Kato, M. Kameyama, Plasma and serum G4 isoenzyme of acetylcholinesterase in patients with Alzheimer-type dementia and vascular dementia, *Ann Clin Biochem*. 27 (1990) 321–6.
- [51] D.G. Wilkinson, P.T. Francis, E. Schwam, J. Payne-Parrish, Cholinesterase inhibitors used in the treatment of Alzheimer's disease: the relationship between pharmacological effects and clinical efficacy, *Drugs Aging*. 21 (2004) 453–78.
- [52] L. Parnetti, D. Chiasserini, U. Andreasson, M. Ohlson, C. Hüls, H. Zetterberg, L. Minthon, A.K. Wallin, N. Andreasen, V.N. Talesa, K. Blennow, Changes in CSF acetyl- and butyrylcholinesterase activity after long-term treatment with AChE inhibitors in Alzheimer's disease, *Acta Neurol Scand*. 124 (2011) 122–9.
- [53] A. Alkalay, G.D. Rabinovici, G. Zimmerman, N. Agarwal, D. Kaufer, B.L. Miller, W.J. Jagust, H. Soreq, Plasma acetylcholinesterase activity correlates with intracerebral β -amyloid load, *Curr Alzheimer Res*. 10 (2013) 48–56.
- [54] P. Davidsson, K. Blennow, N. Andreasen, B. Eriksson, L. Minthon, C. Hesse, Differential increase in cerebrospinal fluid-acetylcholinesterase after treatment with acetylcholinesterase inhibitors in patients with Alzheimer's disease, *Neurosci Lett*. 300 (2001) 157–60.
- [55] S. Amici, A. Lanari, R. Romani, C. Antognelli, V. Gallai, L. Parnetti, Cerebrospinal fluid acetylcholinesterase activity after long-term treatment with donepezil and rivastigmine, *Mech Ageing Dev*. 122 (2001) 2057–62.
- [56] R. Jha, S.I. Rizvi, Age-dependent decline in erythrocyte acetylcholinesterase activity: correlation with oxidative stress, *Biomed Pap Med Fac Univ Palacky Olomouc Czech Repub*. 153 (2009) 195–8.
- [57] M.M. Mesulam, M.A. Morán, Cholinesterases within neurofibrillary tangles related to age and Alzheimer's disease, *Ann Neurol*. 22 (1987) 223–8.
- [58] N.C. Inestrosa, A. Alvarez, C.A. Pérez, R.D. Moreno, M. Vicente, C. Linker, O.I. Casanueva, C. Soto, J. Garrido, Acetylcholinesterase accelerates assembly of amyloid-beta-peptides into Alzheimer's fibrils: possible role of the peripheral site of the enzyme, *Neuron*. 16 (1996) 881–891.
- [59] A. Alvarez, C. Opazo, R. Alarcón, J. Garrido, N.C. Inestrosa, Acetylcholinesterase promotes the aggregation of amyloid-beta-peptide fragments by forming a complex with the growing fibrils, *J Mol Biol*. 272 (1997) 348–361.
- [60] A. Alvarez, R. Alarcón, C. Opazo, E.O. Campos, F.J. Muñoz, F.H. Calderón, F. Dajas, M.K. Gentry, B.P. Doctor, F.G. De Mello, N.C. Inestrosa, Stable complexes involving acetylcholinesterase and amyloid-beta peptide change the biochemical properties of the enzyme and increase the neurotoxicity of Alzheimer's fibrils, *J Neurosci*. 18 (1998) 3213–23.
- [61] M.L. Onor, M. Trevisiol, E. Aguglia, Rivastigmine in the treatment of Alzheimer's disease: an update., *Clin. Interv. Aging*. 2 (2007) 17–32.
- [62] R.A. Abdel-Aal, A.A.A. Assi, B.B. Kostandy, Rivastigmine reverses aluminum-induced behavioral changes in rats, *Eur. J. Pharmacol*. 659 (2011) 169–176.
- [63] R.A. Hansen, G. Gartlehner, A.P. Webb, L.C. Morgan, C.G. Moore, D.E. Jonas, Efficacy and

- safety of donepezil, galantamine, and rivastigmine for the treatment of Alzheimer's disease: a systematic review and meta-analysis, *Clin Interv Aging*. 3 (2008) 211–25.
- [64] M.F. Eskander, N.G. Nagykerly, E.Y. Leung, B. Khelghati, C. Geula, Rivastigmine is a potent inhibitor of acetyl- and butyrylcholinesterase in Alzheimer's plaques and tangles, *Brain Res*. 1060 (2005) 144–52.
- [65] R. Mahlberg, S. Walther, U. Eichmann, F. Tracik, D. Kunz, Effects of rivastigmine on actigraphically monitored motor activity in severe agitation related to Alzheimer's disease: a placebo-controlled pilot study, *Arch Gerontol Geriatr*. 45 (2007) 19–26.
- [66] D.R. McLachlan, C. Bergeron, J.E. Smith, D. Boomer, S.L. Rifat, Risk for neuropathologically confirmed Alzheimer's disease and residual aluminum in municipal drinking water employing weighted residential histories, *Neurology*. 46 (1996) 401–405.
- [67] P. Altmann, J. Cunningham, U. Dhanesha, M. Ballard, J. Thompson, F. Marsh, Disturbance of cerebral function in people exposed to drinking water contaminated with aluminium sulphate: retrospective study of the Camelford water incident, *BMJ*. 319 (1999) 807–811.
- [68] E. Gauthier, I. Fortier, F. Courchesne, P. Pepin, J. Mortimer, D. Gauvreau, Aluminum Forms in Drinking Water and Risk of Alzheimer's Disease, *Environ. Res*. 84 (2000) 234–246.
- [69] T. Peder Flaten, Aluminium as a risk factor in Alzheimer's disease, with emphasis on drinking water, *Brain Res. Bull*. 55 (2001) 187–196.
- [70] C. Exley, M.M. Esiri, Severe cerebral congophilic angiopathy coincident with increased brain aluminium in a resident of Camelford, Cornwall, UK, *J. Neurol. Neurosurg. Psychiatry*. 77 (2006) 877–879.
- [71] T. Singh, R.K. Goel, Neuroprotective effect of *Allium cepa* L. in aluminium chloride induced neurotoxicity, *Neurotoxicology*. 49 (2015) 1–7.
- [72] V. Rondeau, H. Jacqmin-Gadda, D. Commenges, C. Helmer, J.-F. Dartigues, Aluminum and silica in drinking water and the risk of Alzheimer's disease or cognitive decline: findings from 15-year follow-up of the PAQUID cohort, *Am J Epidemiol*. 169 (2009) 489–96.
- [73] A. Kaur, K.D. Gill, Possible peripheral markers for chronic aluminium toxicity in Wistar rats, *Toxicol Ind Heal*. 22 (2006) 39–46.
- [74] J.M. Candy, A.E. Oakley, J. Klinowski, T.A. Carpenter, R.H. Perry, J.R. Atack, E.K. Perry, G. Blessed, A. Fairbairn, J.A. Edwardson, Aluminosilicates and senile plaque formation in Alzheimer's disease, *Lancet*. 1 (1986) 354–7.
- [75] P.T. Francis, A.M. Palmer, M. Snape, G.K. Wilcock, The cholinergic hypothesis of Alzheimer's disease: A review of progress, *J Neurol Neurosurg Psychiatry*. 66 (1999) 137–147.
- [76] R. Liang, Cross Talk Between Aluminum and Genetic Susceptibility and Epigenetic Modification in Alzheimer's Disease, *Adv Exp Med Biol*. 1091 (2018) 173–191.
- [77] M.-H. Yang, S.-C. Chen, Y.-F. Lin, Y.-C. Lee, M.-Y. Huang, K.-C. Chen, H.-Y. Wu, P.-C. Lin, I. Gozes, Y.-C. Tyan, Reduction of aluminum ion neurotoxicity through a small peptide application - NAP treatment of Alzheimer's disease, *J Food Drug Anal*. 27 (2019) 551–564.
- [78] N.A. Singh, A.K.A. Mandal, Z.A. Khan, Inhibition of Al(III)-Induced A β 42 Fibrillation

- and Reduction of Neurotoxicity by Epigallocatechin-3-Gallate Nanoparticles, *J Biomed Nanotechnol.* 14 (2018) 1147–1158.
- [79] X.J. Yang, Y.Z. Yuan, Q. Niu, Association between serum aluminium level and methylation of amyloid precursor protein gene in workers engaged in aluminium electrolysis, *Zhonghua Lao Dong Wei Sheng Zhi Ye Bing Za Zhi.* 34 (2016) 255–8.
- [80] A. Campbell, A. Kumar, F.G. La Rosa, K.N. Prasad, S.C. Bondy, Aluminum increases levels of beta-amyloid and ubiquitin in neuroblastoma but not in glioma cells, *Proc Soc Exp Biol Med.* 223 (2000) 397–402.
- [81] M. Kawahara, M. Kato, Y. Kuroda, Effects of aluminum on the neurotoxicity of primary cultured neurons and on the aggregation of beta-amyloid protein, *Brain Res Bull.* 55 (2001) 211–7.
- [82] E. Bjertness, J.M. Candy, A. Torvik, P. Ince, F. McArthur, G.A. Taylor, S.W. Johansen, J. Alexander, J.K. Grønnesby, L.S. Bakkeiteig, J.A. Edwardson, Content of brain aluminum is not elevated in Alzheimer disease, *Alzheimer Dis Assoc Disord.* 10 (1996) 171–4.
- [83] C. Germano, G.J. Kinsella, Working memory and learning in early Alzheimer's disease, *Neuropsychol Rev.* 15 (2005) 1–10.
- [84] A.C. Miu, O. Benga, Aluminum, and Alzheimer's disease: a new look, *J Alzheimers Dis.* 10 (2006) 179–201.
- [85] P. Zatta, T. Kiss, M. Suwalsky, G. Berthon, Aluminium (III) as a promoter of cellular oxidation, *Coord. Chem. Rev.* 228 (2002) 271–284.
- [86] V. Kakkar, I.P. Kaur, Evaluating potential of curcumin loaded solid lipid nanoparticles in aluminium induced behavioural, biochemical and histopathological alterations in mice brain, *Food Chem Toxicol.* 49 (2011) 2906–13.
- [87] R.R. Kaizer, M.C. Corrêa, R.M. Spanevello, V.M. Morsch, C.M. Mazzanti, J.F. Gonçalves, M.R.C. Schetinger, Acetylcholinesterase activation and enhanced lipid peroxidation after long-term exposure to low levels of aluminum on different mouse brain regions, *J Inorg Biochem.* 99 (2005) 1865–70.
- [88] J.R. Walton, M.-X. Wang, APP expression, distribution and accumulation are altered by aluminum in a rodent model for Alzheimer's disease, *J Inorg Biochem.* 103 (2009) 1548–54.
- [89] H. Zheng, M.B.H. Youdim, M. Fridkin, Site-Activated Multifunctional Chelator with Acetylcholinesterase and Neuroprotective-Neurorestorative Moieties for Alzheimer's Therapy, *J. Med. Chem.* 52 (2009) 4095–4098.
- [90] M.M. Said, M.M.A. Rabo, Neuroprotective effects of eugenol against aluminium induced toxicity in the rat brain, *Arh Hig Rada Toksikol.* 68 (2017) 27–37.
- [91] K. Gulya, Z. Rakonczay, P. Kása, Cholinotoxic effects of aluminum in rat brain, *J Neurochem.* 54 (1990) 1020–6.
- [92] L.R. Fodero, S.S. Mok, D. Losic, L.L. Martin, M.I. Aguilar, C.J. Barrow, B.G. Livett, D.H. Small, Alpha7-nicotinic acetylcholine receptors mediate an Aβ(1-42)-induced increase in the level of acetylcholinesterase in primary cortical neurones, *J Neurochem.* 88 (2004) 1186–93.
- [93] P. Nayak, Aluminum: impacts and disease, *Env. Res.* 89 (2002) 101–15.
- [94] R.R. Kaizer, M.C. Corrêa, L.R.S. Gris, C.S. da Rosa, D. Bohrer, V.M. Morsch, M.R. Chitolina, Schetinger, Effect of long-term exposure to aluminum on the acetylcholinesterase activity in the central nervous system and erythrocytes, *Neurochem Res.* 33 (2008) 2294–2301.
- [95] M. Nampoothiri, J. John, N. Kumar, J.

- Mudgal, G.K. Nampurath, M.R. Chamallamudi, Modulatory Role of Simvastatin against Aluminium Chloride-Induced Behavioural and Biochemical Changes in Rats, *Behav. Neurol.* 2015 (2015) 210169.
- [96] M.L. Onor, M. Trevisiol, E. Aguglia, Rivastigmine in the treatment of Alzheimer's disease: an update, *Clin. Interv. Aging.* 2 (2007) 17–32.
- [97] M. Nampoothiri, N. Kumar, G.V. Ramalingayya, N.G. Kutty, N. Krishnadas, C. Mallikarjuna, Rao, Effect of insulin on spatial memory in aluminum chloride-induced dementia in rats, *Neuroreport.* 28 (2017) 540–544.
- [98] N.H. Greig, T. Utsuki, Q. Yu, X. Zhu, H.W. Holloway, T. Perry, B. Lee, D.K. Ingram, D.K. Lahiri, A new therapeutic target in Alzheimer's disease treatment: attention to butyrylcholinesterase, *Curr. Med. Res. Opin.* 17 (2001) 159–165.
- [99] E. Giacobini, R. Spiegel, A. Enz, A.E. Veroff, N.R. Cutler, Inhibition of acetyl- and butyrylcholinesterase in the cerebrospinal fluid of patients with Alzheimer's disease by rivastigmine: correlation with cognitive benefit, *J. Neural Transm.* 109 (2002) 1053–1065.
- [100] A. Jangra, P. Kasbe, S.N. Pandey, S. Dwivedi, S.S. Gurjar, M. Kwatra, M. Mishra, A.K. Venu, K. Sulakhiya, R. Gogoi, N. Sarma, B.K. Bezbaruah, M. Lahkar, Hesperidin and Silibinin Ameliorate Aluminum-Induced Neurotoxicity: Modulation of Antioxidants and Inflammatory Cytokines Level in Mice Hippocampus, *Biol. Trace Elem. Res.* 168 (2015) 462–471.
- [101] M.F. Ikram, S.M. Farhat, A. Mahboob, S. Baig, A. Yaqinuddin, T. Ahmed, Expression of DnMTs and MBDs in AICl₃-Induced Neurotoxicity Mouse Model, *Biol. Trace Elem. Res.* (2020).
- [102] H.N. Mustafa, Neuro-amelioration of cinnamaldehyde in aluminum-induced Alzheimer's disease rat model, *J. Histotechnol.* 43 (2020) 11–20.
- [103] S. Deiana, C.R. Harrington, C.M. Wischik, G. Riedel, Methylthionium chloride reverses cognitive deficits induced by scopolamine: comparison with rivastigmine, *Psychopharmacol.* 202 (2009) 53–65.
- [104] A. Akhtar, M. Bishnoi, S.P. Sah, Sodium orthovanadate improves learning and memory in intracerebroventricular-streptozotocin rat model of Alzheimer's disease through modulation of brain insulin resistance induced tau pathology, *Brain Res. Bull.* 164 (2020) 83–97.
- [105] B. Chogtu, A. Arivazhahan, S.K. Kunder, A. Tilak, R. Sori, A. Tripathy, Evaluation of Acute and Chronic Effects of D-Galactose on Memory and Learning in Wistar Rats, *Clin. Psychopharmacol. Neurosci.* 16 (2018) 153–160.
- [106] I.A. Wilson, M. Gallagher, H. Eichenbaum, H. Tanila, Neurocognitive aging: prior memories hinder new hippocampal encoding, *Trends Neurosci.* 29 (2006) 662–670.
- [107] M. Gallagher, P.R. Rapp, The use of animal models to study the effects of aging on cognition, *Annu Rev Psychol.* 48 (1997) 339–70.
- [108] M.W. Bondi, W.S. Houston, L.T. Eyler, G.G. Brown, fMRI evidence of compensatory mechanisms in older adults at genetic risk for Alzheimer disease, *Neurology.* 64 (2005) 501–8.
- [109] Y.T. Quiroz, A.E. Budson, K. Celone, A. Ruiz, R. Newmark, G. Castrillón, F. Lopera, C.E.

- Stern, Hippocampal hyperactivation in presymptomatic familial Alzheimer's disease, *Ann Neurol.* 68 (2010) 865–75.
- [110] R.A. Sperling, B.C. Dickerson, M. Pihlajamaki, P. Vannini, P.S. LaViolette, O. V Vitolo, T. Hedden, J.A. Becker, D.M. Rentz, D.J. Selkoe, K.A. Johnson, Functional alterations in memory networks in early Alzheimer's disease, *Neuromolecular Med.* 12 (2010) 27–43.
- [111] S.Y. Bookheimer, M.H. Strojwas, M.S. Cohen, A.M. Saunders, M.A. Pericak-Vance, J.C. Mazziotta, G.W. Small, Patterns of brain activation in people at risk for Alzheimer's disease, *N Engl J Med.* 343 (2000) 450–6.
- [112] M.J. West, C.H. Kawas, L.J. Martin, J.C. Troncoso, The CA1 region of the human hippocampus is a hot spot in Alzheimer's disease, *Ann N Y Acad Sci.* 908 (2000) 255–9.
- [113] F.H. Bazzari, D.M. Abdallah, H.S. El-abhar, Chenodeoxycholic Acid Ameliorates A β 1-Induced Alzheimer's Disease Neurotoxicity and Cognitive Deterioration via Enhanced Insulin Signaling in Rats, *Molecules.* 24 (2019) 1992.
- [114] A.F.S. Junior, M.S.S. Aguiar, O.S.C. Junior, L. de N.S. Santana, E.C.S. Franco, R.R. Lima, N.V.M. de Siqueira, R.A. Feio, L.R.F. Faro, W. Gomes-Leal, Hippocampal neuronal loss, decreased GFAP immunoreactivity and cognitive impairment following experimental intoxication of rats with aluminum citrate, *Brain Res.* 1491 (2013) 23–33.
- [115] K. Mahdy, O. Shaker, H. Wafay, Y. Nassar, H. Hassan, A. Hussein, Effect of some medicinal plant extracts on the oxidative stress status in Alzheimer's disease induced in rats, *Eur. Rev. Med. Pharmacol. Sci.* 16 (2012) 31–42.
- [116] C. Laurent, G. Dorothée, S. Hunot, E. Martin, Y. Monnet, M. Duchamp, Y. Dong, F.P. Légeron, A. Leboucher, S. Burnouf, E. Faivre, K. Carvalho, R. Caillierez, N. Zommer, D. Demeyer, N. Jouy, V. Sazdovitch, S. Schraen-Maschke, C. Delarasse, L. Buée, D. Blum, Hippocampal T cell infiltration promotes neuroinflammation and cognitive decline in a mouse model of tauopathy, *Brain.* 140 (2017) 184–200.
- [117] A. Bellucci, A.J. Westwood, E. Ingram, F. Casamenti, M. Goedert, M.G. Spillantini, Induction of inflammatory mediators and microglial activation in mice transgenic for mutant human P301S tau protein, *Am. J. Pathol.* 165 (2004) 1643–1652.
- [118] Y. Yoshiyama, M. Higuchi, B. Zhang, S.-M. Huang, N. Iwata, T.C. Saido, J. Maeda, T. Suhara, J.Q. Trojanowski, V.M.-Y. Lee, Synapse Loss and Microglial Activation Precede Tangles in a P301S Tauopathy Mouse Model, *Neuron.* 53 (2007) 337–351.
- [119] F. González-Scarano, G. Baltuch, Microglia as mediators of inflammatory and degenerative diseases, *Annu Rev Neurosci.* 22 (1999) 219–40.
- [120] P.P. Guan, P. Wang, Integrated communications between cyclooxygenase-2 and Alzheimer's disease, *FASEB J.* 33 (2019) 13–33.
- [121] M. Olabarria, H.N. Noristani, A. Verkhratsky, J.J. Rodríguez, Concomitant astroglial atrophy and astrogliosis in a triple transgenic animal model of Alzheimer's disease, *Glia.* 58 (2010) 831–838.
- [122] L. Hou, Y. Liu, X. Wang, H. Ma, J. He, Y. Zhang, C. Yu, W. Guan, Y. Ma, The effects of amyloid- β 42 oligomer on the proliferation and activation of astrocytes in vitro, *Vitr. Cell. Dev. Biol. - Anim.* 47 (2011) 573–580.
- [123] U. Wilhelmsson, E.A. Bushong, D.L. Price, B.L. Smarr, V. Phung, M. Terada, M.H.

- Ellisman, M. Pekny, Redefining the concept of reactive astrocytes as cells that remain within their unique domains upon reaction to injury, PNAS. 103 (2006) 17513–17518.
- [124] W. Kamphuis, C. Mamber, M. Moeton, L. Kooijman, J.A. Sluijs, A.H.P. Jansen, M. Verveer, L.R. de Groot, V.D. Smith, S. Rangarajan, J.J. Rodríguez, M. Orre, E.M. Hol, GFAP isoforms in adult mouse brain with a focus on neurogenic astrocytes and reactive astrogliosis in mouse models of Alzheimer disease, PLoS One. 7 (2012) e42823.
- [125] M. Erta, A. Quintana, J. Hidalgo, Interleukin-6, a major cytokine in the central nervous system, Int J Biol Sci. 8 (2012) 1254–66.
- [126] R.G. Biringer, The Role of Eicosanoids in Alzheimer's Disease, Int J Env. Res Public Heal. 16 (2019) 2560.
- [127] P. Teismann, J.B. Schulz, Cellular pathology of Parkinson's disease: astrocytes, microglia and inflammation, Cell Tissue Res. 318 (2004) 149–61.
- [128] S.A. Liddelow, K.A. Guttenplan, L.E. Clarke, F.C. Bennett, C.J. Bohlen, L. Schirmer, M.L. Bennett, A.E. Münch, W.S. Chung, T.C. Peterson, D.K. Wilton, A. Frouin, B.A. Napier, N. Panicker, M. Kumar, M.S. Buckwalter, D.H. Rowitch, V.L. Dawson, T.M. Dawson, B. Stevens, B.A. Barres, Neurotoxic reactive astrocytes are induced by activated microglia, Nature. 541 (2017) 481–487.
- [129] M. Reale, C. Iarlori, A. Thomas, D. Gambi, B. Perfetti, M. Di Nicola, M. Onofri, Peripheral cytokines profile in Parkinson's disease, Brain Behav Immun. 23 (2009) 55–63.
- [130] M. Zhu, X. Wang, L. Sun, M. Schultzberg, E. Hjorth, Can inflammation be resolved in Alzheimer's disease?, Ther. Adv. Neurol. Disord. 11 (2018) PMC6088473.
- [131] J. Hu, K.T. Akama, G.A. Krafft, B.A. Chromy, L.J. Van Eldik, Amyloid- β peptide activates cultured astrocytes: Morphological alterations, cytokine induction and nitric oxide release, Brain Res. 785 (1998) 195–206.
- [132] H.S. Nhan, K. Chiang, E.H. Koo, The multifaceted nature of amyloid precursor protein and its proteolytic fragments: friends and foes, Acta Neuropathol. 129 (2015) 1–19.
- [133] T.G. Beach, E.G. McGeer, Lamina-specific arrangement of astrocytic gliosis and senile plaques in Alzheimer's disease visual cortex, Brain Res. 463 (1988) 357–361.
- [134] M.L. Kashon, G.W. Ross, J.P. O'Callaghan, D.B. Miller, H. Petrovitch, C.M. Burchfiel, D.S. Sharp, W.R. Markesbery, D.G. Davis, J. Hardman, J. Nelson, L.R. White, Associations of cortical astrogliosis with cognitive performance and dementia status, J. Alzheimer's Dis. 6 (2004) 595–604.
- [135] R.E. Mrak, J.G. Sheng, W.S. Griffin, Correlation of astrocytic S100 beta expression with dystrophic neurites in amyloid plaques of Alzheimer's disease, J. Neuropathol. Exp. Neurol. 55 (1996) 273–279.
- [136] S.X. Guo-ross, E.Y. Yang, T.J. Walsh, S.C. Bondy, Decrease of Glial Fibrillary Acidic Protein in Rat Frontal Cortex Following Aluminum Treatment, J. Neurochem. 73 (1999) 1609–1614.
- [137] A. Justin-Thenmozhi, M.D. Bharathi, R. Kiruthika, T. Manivasagam, A. Borah, M.M. Essa, Attenuation of Aluminum Chloride-Induced Neuroinflammation and Caspase Activation Through the AKT / GSK-3 β Pathway by Hesperidin in Wistar Rats, Neurotox. Res. 34 (2018) 463–476.
- [138] H. Erazi, W. Sansar, S. Ahboucha, H. Gamrani, Aluminum affects glial system and behavior of rats, C. R. Biol. 333 (2010) 23–27.
- [139] X.-B. Li, H. Zheng, Z.-R. Zhang, M. Li, Z.-Y.

- Huang, H.J. Schluesener, Y.-Y. Li, S.-Q. Xu, Glia activation induced by peripheral administration of aluminum oxide nanoparticles in rat brains, *Nanomedicine*. 5 (2009) 473–479.
- [140] R.A. Yokel, J.P.O. Callaghan, An Aluminum-Induced Increase in GFAP Is Attenuated by Some Chelators, *Neurotoxicol. Teratol.* 20 (1998) 55–60.
- [141] B. Platt, G. Fiddler, G. Riedel, Z. Henderson, Aluminium toxicity in the rat brain: histochemical and immunocytochemical evidence, *Brain Res. Bull.* 55 (2001) 257–267.
- [142] L.A. Mohamed, J.N. Keller, A. Kaddoumi, Role of P-glycoprotein in mediating rivastigmine effect on amyloid- β brain load and related pathology in Alzheimer's disease mouse model, *Biochim. Biophys. Acta.* 1862 (2016) 778–787.
- [143] T. Matsuda, T. Hisatsune, Cholinergic Modification of Neurogenesis and Gliosis Improves the Memory of A β PPswe/PSEN1dE9 Alzheimer's Disease Model Mice Fed a High-Fat Diet, *J Alzheimers Dis.* 56 (2017) 1–23.
- [144] P.L. McGeer, M. Schulzer, E.G. McGeer, Arthritis and anti-inflammatory agents as possible protective factors for Alzheimer's disease: a review of 17 epidemiologic studies, *Neurology*. 47 (1996) 425–432.
- [145] B.A. in t' Veld, A. Ruitenber, A. Hofman, L.J. Launer, C.M. van Duijn, T. Stijnen, M.M. Breteler, B.H. Stricker, Nonsteroidal antiinflammatory drugs and the risk of Alzheimer's disease, *N Engl J Med.* 345 (2001) 1515–21.
- [146] P.L. McGeer, E.G. McGeer, NSAIDs and Alzheimer disease: epidemiological, animal model and clinical studies, *Neurobiol. Aging*. 28 (2007) 639–647.
- [147] J.C. Breitner, The role of anti-inflammatory drugs in the prevention and treatment of Alzheimer's disease, *Annu Rev Med.* 47 (1996) 401–11.
- [148] B.A. in 't Veld, L.J. Launer, A.W. Hoes, A. Ott, A. Hofman, M.M. Breteler, B.H. Stricker, NSAIDs and incident Alzheimer's disease. The Rotterdam Study, *Neurobiol Aging*. 19 (1998) 607–11.
- [149] C. Zhang, Y. Wang, D. Wang, J. Zhang, F. Zhang, NSAID Exposure and Risk of Alzheimer's Disease: An Updated Meta-Analysis From Cohort Studies, *Front Aging Neurosci.* 10 (2018) 83.
- [150] G.P. Lim, F. Yang, T. Chu, P. Chen, W. Beech, B. Teter, T. Tran, O. Ubeda, K.H. Ashe, S.A. Frautschy, G.M. Cole, Ibuprofen suppresses plaque pathology and inflammation in a mouse model for Alzheimer's disease, *J. Neurosci.* 20 (2000) 5709–5714.
- [151] G.P. Lim, F. Yang, T. Chu, E. Gahtan, O. Ubeda, W. Beech, J.B. Overmier, K. Hsiao-Ashec, S.A. Frautschy, G.M. Cole, Ibuprofen effects on Alzheimer pathology and open field activity in APPsw transgenic mice, *Neurobiol. Aging*. 22 (2001) 983–991.
- [152] A.C. McKee, I. Carreras, L. Hossain, H. Ryu, W.L. Klein, S. Oddo, F.M. LaFerla, B.G. Jenkins, N.W. Kowall, A. Dedeoglu, Ibuprofen reduces Abeta, hyperphosphorylated tau and memory deficits in Alzheimer mice, *Brain Res.* 207 (2008) 225–36.
- [153] M. Cuendet, A.D. Mesecar, D.L. DeWitt, J.M. Pezzuto, An ELISA method to measure inhibition of the COX enzymes, *Nat Protoc.* 1 (2006) 1915–21.
- [154] K. Yamagata, K.I. Andreasson, W.E. Kaufmann, C.A. Barnes, P.F. Worley, Expression of a mitogen-inducible cyclooxygenase in brain neurons: regulation by

- synaptic activity and glucocorticoids, *Neuron*. 11 (1993) 371–86.
- [155] E. Mhillaj, M.G. Morgese, P. Tucci, A. Furiano, L. Luongo, M. Bove, S. Maione, V. Cuomo, S. Schiavone, L. Trabace, Celecoxib Prevents Cognitive Impairment and Neuroinflammation in Soluble Amyloid β -treated Rats, *Neuroscience*. 372 (2018) 58–73.
- [156] W.E. Kaufmann, P.F. Worley, J. Pegg, M. Bremer, P. Isakson, COX-2, a synaptically induced enzyme, is expressed by excitatory neurons at postsynaptic sites in rat cerebral cortex, *Proc Natl Acad Sci U S A*. 93 (1996) 2317–21.
- [157] L.A. Teather, M.G. Packard, N.G. Bazan, Post-training cyclooxygenase-2 (COX-2) inhibition impairs memory consolidation, *Learn Mem*. 9 (2002) 41–7.
- [158] X. Liang, L. Wu, Q. Wang, T. Hand, M. Bilak, L. McCullough, K. Andreasson, Function of COX-2 and prostaglandins in neurological disease, *J Mol Neurosci*. 33 (2007) 94–9.
- [159] T. Wyss-Coray, J. Rogers, Inflammation in Alzheimer disease—a brief review of the basic science and clinical literature, *Cold Spring Harb Perspect Med*. 2 (2012) a006346.
- [160] H. Yang, C. Chen, Cyclooxygenase-2 in Synaptic Signaling, *Curr Pharm Des*. 14 (2008) 1443–51.
- [161] J.J.M. Hoozemans, J.M. Rozemuller, E.S. van Haastert, R. Veerhuis, P. Eikelenboom, Cyclooxygenase-1 and -2 in the different stages of Alzheimer’s disease pathology, *Curr Pharm Des*. 14 (2008) 1419–27.
- [162] W.F. Stewart, C. Kawas, M. Corrada, E.J. Metter, Risk of Alzheimer’s disease and duration of NSAID use, *Neurology*. 48 (1997) 626–32.
- [163] S. Miettinen, F.R. Fusco, J. Yrjänheikki, R. Keinänen, T. Hirvonen, R. Roivainen, M. Närhi, T. Hökfelt, J. Koistinaho, Spreading depression and focal brain ischemia induce cyclooxygenase-2 in cortical neurons through N-methyl-D-aspartic acid- receptors and phospholipase A2, *Proc Natl Acad Sci U S A*. 94 (1997) 6500–6505.
- [164] K.I. Andreasson, A. Savonenko, S. Vidensky, J.J. Goellner, Y. Zhang, A. Shaffer, W.E. Kaufmann, P.F. Worley, P. Isakson, A.L. Markowska, Age-dependent cognitive deficits and neuronal apoptosis in cyclooxygenase-2 transgenic mice, *J Neurosci*. 21 (2001) 8198–209.
- [165] K. Kadoyama, Y. Takahashi, H. Higashida, T. Tanabe, T. Yoshimoto, Cyclooxygenase-2 stimulates production of amyloid beta-peptide in neuroblastoma x glioma hybrid NG108-15 cells, *Biochem Biophys Res Commun*. 281 (2001) 483–90.
- [166] T. Melnikova, A. Savonenko, Q. Wang, X. Liang, T. Hand, L. Wu, W.E. Kaufmann, A. Vehmas, K.I. Andreasson, Cyclooxygenase-2 activity promotes cognitive deficits but not increased amyloid burden in a model of Alzheimer’s disease in a sex-dimorphic pattern, *Neuroscience*. 141 (2006) 1149–62.
- [167] Z. Xiang, L. Ho, J. Valdellon, D. Borchelt, K. Kelley, L. Spielman, P.S. Aisen, G.M. Pasinetti, Cyclooxygenase (COX)-2 and cell cycle activity in a transgenic mouse model of Alzheimer’s disease neuropathology, *Neurobiol Aging*. 23 (2002) 327–34.
- [168] Z. Xiang, L. Ho, S. Yemul, Z. Zhao, W. Qing, P. Pompl, K. Kelley, A. Dang, W. Qing, D. Teplow, G.M. Pasinetti, Cyclooxygenase-2 promotes amyloid plaque deposition in a mouse model of Alzheimer’s disease neuropathology, *Gene Expr*. 10 (2002) 271–8.
- [169] N.G. Bazan, V. Colangelo, W.J. Lukiw, Prostaglandins and other lipid mediators in

- Alzheimer's disease, Prostaglandins Other Lipid Mediat. 68–69 (2002) 197–210.
- [170] K. Yui, G. Imataka, H. Nakamura, N. Ohara, Y. Naito, Eicosanoids Derived From Arachidonic Acid and Their Family Prostaglandins and Cyclooxygenase in Psychiatric Disorders, *Curr Neuropharmacol.* 13 (2015) 776–85.
- [171] D. Zhang, X. Hu, L. Qian, B. Wilson, C. Lee, P. Flood, R. Langenbach, J.-S. Hong, Prostaglandin E2 released from activated microglia enhances astrocyte proliferation in vitro, *Toxicol Appl Pharmacol.* 238 (2009) 64–70.
- [172] M. Sastre, T. Klockgether, M.T. Heneka, Contribution of inflammatory processes to Alzheimer's disease: molecular mechanisms, *Int J Dev Neurosci.* 24 (2006) 167–76.
- [173] R.K. Lee, S. Knapp, R.J. Wurtman, Prostaglandin E2 stimulates amyloid precursor protein gene expression: inhibition by immunosuppressants, *J Neurosci.* 19 (1999) 940–7.
- [174] J.S.B. Shaik, T.M. Miller, S.H. Graham, M.D. Manole, S.M. Poloyac, Rapid and simultaneous quantitation of prostanoids by UPLC-MS/MS in rat brain, *J Chromatogr B Anal. Technol Biomed Life Sci.* 945–946 (2014) 207–16.
- [175] T. Ogorochi, S. Narumiya, N. Mizuno, K. Yamashita, H. Miyazaki, O. Hayaishi, Regional distribution of prostaglandins D2, E2, and F2 alpha and related enzymes in postmortem human brain, *J Neurochem.* 43 (1984) 71–82.
- [176] H. Anninos, G. Andrikopoulos, S. Pastromas, D. Sakellariou, G. Theodorakis, P. Vardas, Triflusal: an old drug in modern antiplatelet therapy. Review of its action, use, safety and effectiveness, *Hell. J Cardiol.* 50 (2009) 199–207.
- [177] D. Sredni-Kenigsbuch, TH1/TH2 cytokines in the central nervous system, *Int J Neurosci.* 112 (2002) 665–703.
- [178] H.H. Ahmed, A.M. Salem, G.M. Sabry, A.A. Husein, S.E. Kotob, New insights in the horizon for the treatment of Alzheimer's Disease: A proposal based on experimental study, *Der Pharm. Lett.* 7 (2015) 165–182.
- [179] Y. Jun-Qing, L. Bei-Zhong, H. Bai-Cheng, Z. Qi-Qin, Protective effects of meloxicam on aluminum overload-induced cerebral damage in mice, *Eur J Pharmacol.* 547 (2006) 52–8.
- [180] L. Ho, D. Purohit, V. Haroutunian, J.D. Luteran, F. Willis, J. Naslund, J.D. Buxbaum, R.C. Mohs, P.S. Aisen, G.M. Pasinetti, Neuronal cyclooxygenase 2 expression in the hippocampal formation as a function of the clinical progression of Alzheimer disease, *Arch Neurol.* 58 (2001) 487–92.
- [181] J.J. Hoozemans, A.J. Rozemuller, I. Janssen, C.J. De Groot, R. Veerhuis, P. Eikelenboom, Cyclooxygenase expression in microglia and neurons in Alzheimer's disease and control brain, *Acta Neuropathol.* 101 (2001) 2–8.
- [182] J.J.M. Hoozemans, M.K. Brückner, A.J.M. Rozemuller, R. Veerhuis, P. Eikelenboom, T. Arendt, Cyclin D1 and cyclin E are co-localized with cyclo-oxygenase 2 (COX-2) in pyramidal neurons in Alzheimer disease temporal cortex, *J Neuropathol Exp Neurol.* 61 (2002) 678–88.
- [183] K. Yasojima, C. Schwab, E.G. McGeer, P.L. McGeer, Distribution of cyclooxygenase-1 and cyclooxygenase-2 mRNAs and proteins in human brain and peripheral organs, *Brain Res.* 830 (1999) 226–36.
- [184] H. Braak, E. Braak, Neuropathological staging of Alzheimer-related changes, *Acta*

- Neuropathol. 82 (1991) 239–59.
- [185] R.W. Chen, Y. Zhang, M.E. Rose, S.H. Graham, Cyclooxygenase-2 activity contributes to neuronal expression of cyclin D1 after anoxia/ischemia in vitro and in vivo, *Brain Res Mol Brain Res.* 132 (2004) 31–7.
- [186] J.J.M. Hoozemans, R. Veerhuis, A.J.M. Rozemuller, T. Arendt, P. Eikelenboom, Neuronal COX-2 expression and phosphorylation of pRb precede p38 MAPK activation and neurofibrillary changes in AD temporal cortex, *Neurobiol Dis.* 15 (2004) 492–9.
- [187] M. Goschorska, I. Baranowska-Bosiacka, I. Gutowska, M. Tarnowski, K. Piotrowska, E. Metryka, K. Safranow, D. Chlubek, Effect of acetylcholinesterase inhibitors donepezil and rivastigmine on the activity and expression of cyclooxygenases in a model of the inflammatory action of fluoride on macrophages obtained from THP-1 monocytes, *Toxicology.* 406–407 (2018) 9–20.
- [188] S. Jones, S. Sudweeks, J.L. Yakel, Nicotinic receptors in the brain: correlating physiology with function, *Trends Neurosci.* 22 (1999) 555–61.
- [189] A. Nordberg, Human nicotinic receptors--their role in aging and dementia, *Neurochem Int.* 25 (1994) 93–7.
- [190] D. Paterson, A. Nordberg, Neuronal nicotinic receptors in the human brain, *Prog Neurobiol.* 61 (2000) 75–111.
- [191] M. Ferencik, M. Novak, J. Rovensky, I. Rybar, Alzheimer's disease, inflammation and non-steroidal anti-inflammatory drugs., *Bratisl. Lek. Listy.* 102 (2001) 123–132.
- [192] L.A. Kotilinek, M.A. Westerman, Q. Wang, K. Panizzon, G.P. Lim, A. Simonyi, S. Lesne, A. Falinska, L.H. Younkin, S.G. Younkin, M. Rowan, J. Cleary, R.A. Wallis, G.Y. Sun, G. Cole, S. Frautschy, R. Anwyl, K.H. Ashe, Cyclooxygenase-2 inhibition improves amyloid-beta-mediated suppression of memory and synaptic plasticity, *Brain.* 131 (2008) 651–64.
- [193] C.-Y. Chen, N.-S. Tzeng, Y.-C. Chen, Maintenance therapy of celecoxib for major depression with mimicking neuropsychological dysfunction, *Gen Hosp Psychiatry.* 32 (2010) 647.e7–9.
- [194] G.W. Small, P. Siddarth, D.H.S. Silverman, L.M. Ercoli, K.J. Miller, H. Lavretsky, S.Y. Bookheimer, S.-C. Huang, J.R. Barrio, M.E. Phelps, Cognitive and cerebral metabolic effects of celecoxib versus placebo in people with age-related memory loss: randomized controlled study, *Am J Geriatr Psychiatry.* 16 (2008) 999–1009.
- [195] J.-M.S. Leoutsakos, B.O. Muthen, J.C.S. Breitner, C.G. Lyketsos, Effects of non-steroidal anti-inflammatory drug treatments on cognitive decline vary by phase of pre-clinical Alzheimer disease: findings from the randomized controlled Alzheimer's Disease Anti-inflammatory Prevention Trial, *Int J Geriatr Psychiatry.* 27 (2012) 364–74.
- [196] P.S. Aisen, K.A. Schafer, M. Grundman, E. Pfeiffer, M. Sano, K.L. Davis, M.R. Farlow, S. Jin, R.G. Thomas, L.J. Thal, Effects of rofecoxib or naproxen vs placebo on Alzheimer disease progression: a randomized controlled trial, *JAMA.* 289 (2003) 2819–26.
- [197] S.A. Reines, G.A. Block, J.C. Morris, G. Liu, M.L. Nessly, C.R. Lines, B.A. Norman, C.C. Baranak, Rofecoxib: no effect on Alzheimer's disease in a 1-year, randomized, blinded, controlled study, *Neurology.* 62 (2004) 66–71.
- [198] L.J. Thal, S.H. Ferris, L. Kirby, G.A. Block, C.R. Lines, E. Yuen, C. Assaid, M.L. Nessly, B.A. Norman, C.C. Baranak, S.A. Reines, A

- randomized, double-blind, study of rofecoxib in patients with mild cognitive impairment, *Neuropsychopharmacology*. 30 (2005) 1204–15.
- [199] S.M. Sainati, D.M. Ingram, S. Talwalker, G. Geis, Results of a double-blind, randomized, placebo-controlled study of celecoxib in the treatment of progression of Alzheimer's disease., 6th Int. Stock. Symp. Adv. Alzheimer Ther. Stock. Sweden, 180. (2000).
- [200] T. Kukar, M.P. Murphy, J.L. Eriksen, S.A. Sagi, S. Weggen, T.E. Smith, T. Ladd, M.A. Khan, R. Kache, J. Beard, M. Dodson, S. Merit, V. V Ozols, P.Z. Anastasiadis, P. Das, A. Fauq, E.H. Koo, T.E. Golde, Diverse compounds mimic Alzheimer disease-causing mutations by augmenting Abeta42 production, *Nat Med*. 11 (2005) 545–50.
- [201] B.P. Imbimbo, An update on the efficacy of non-steroidal anti-inflammatory drugs in Alzheimer's disease, *Expert Opin Investig Drugs*. 18 (2009) 1147–68.
- [202] L.-W. Fan, A. Kaizaki, L.-T. Tien, Y. Pang, S. Tanaka, S. Numazawa, A.J. Bhatt, Z. Cai, Celecoxib attenuates systemic lipopolysaccharide-induced brain inflammation and white matter injury in the neonatal rats, *Neuroscience*. 240 (2013) 27–38.
- [203] M. Pohanka, Celecoxib is an inhibitor of enzyme acetylcholinesterase, *Neuro Endocrinol Lett*. 37 (2016) 118–122.
- [204] M. Pohanka, Celecoxib is an inhibitor of enzyme acetylcholinesterase, in *Neuroendocrinol. Lett.*, Maghira and Maas Publications, 2016: pp. 118–122.
- [205] T.H. Ferreira-Vieira, I.M. Guimaraes, F.R. Silva, F.M. Ribeiro, Alzheimer's disease: Targeting the Cholinergic System, *Curr. Neuropharmacol*. 14 (2016) 101–115.
- [206] B. Sharma, N. Singh, M. Singh, Modulation of celecoxib- and streptozotocin-induced experimental dementia of Alzheimer's disease by pitavastatin and donepezil, *J. Psychopharmacol*. 22 (2008) 162–171.
- [207] P.N. Pompl, S. Yemul, Z. Xiang, L. Ho, V. Haroutunian, D. Purohit, R. Mohs, G.M. Pasinetti, Caspase gene expression in the brain as a function of the clinical progression of Alzheimer disease, *Arch Neurol*. 60 (2003) 369–76.
- [208] R.E. Tanzi, R.D. Moir, S.L. Wagner, Clearance of Alzheimer's Aβ peptide: the many roads to perdition, *Neuron*. 43 (2004) 605–8.
- [209] J.H. Su, A.J. Anderson, B.J. Cummings, C.W. Cotman, Immunohistochemical evidence for apoptosis in Alzheimer's disease, *Neuroreport*. 5 (1994) 2529–33.
- [210] Y.-J. Chang, N.H. Linh, Y.-H. Shih, H.-M. Yu, M.S. Li, Y.-R. Chen, Alzheimer's Amyloid-β Sequesters Sequesters Caspase-3 in vitro via its C-terminal Tail, *ACS Chem. Neurosci*. 7 (2016) 1097–1106.
- [211] A. Ashkenazi, V.M. Dixit, Death receptors: signaling and modulation, *Science* (80-.). 281 (1998) 1305–8.
- [212] M. D'Amelio, M. Sheng, F. Cecconi, Caspase-3 in the central nervous system: beyond apoptosis, *Trends Neurosci*. 35 (2012) 700–9.
- [213] C.J. Pike, A.J. Walencewicz-Wasserman, J. Kosmoski, D.H. Cribbs, C.G. Glabe, C.W. Cotman, Structure-activity analyses of beta-amyloid peptides: contributions of the beta 25–35 region to aggregation and neurotoxicity, *J Neurochem*. 64 (1995) 253–65.
- [214] R. Bhatia, H. Lin, R. Lal, Fresh and globular amyloid beta protein (1-42) induces rapid cellular degeneration: evidence for Aβ channel-mediated cellular toxicity, *FASEB J*. 14 (2000) 1233–43.

- [215] D. Watson, E. Castaño, T.A. Kokjohn, Y.-M. Kuo, Y. Lyubchenko, D. Pinsky, E.S.C. Jr, C. Esh, D.C. Luehrs, W.B. Stine, L.M. Rowse, M.R. Emmerling, A.E. Roher, Physicochemical characteristics of soluble oligomeric Abeta and their pathologic role in Alzheimer's disease, *Neurol Res.* 27 (2005) 869–81.
- [216] C. Malaplate-Armand, S. Florent-Béchar, I. Youssef, V. Koziel, I. Sponne, B. Kriem, B. Leininger-Muller, J.-L. Olivier, T. Oster, T. Pillot, Soluble oligomers of amyloid-beta peptide induce neuronal apoptosis by activating a cPLA2-dependent sphingomyelinase-ceramide pathway, *Neurobiol Dis.* 23 (2006) 178–89.
- [217] C.-P. Chang, Y.-F. Liu, H.-J. Lin, C.-C. Hsu, B.-C. Cheng, W.-P. Liu, M.-T. Lin, S.-F. Hsu, L.-S. Chang, K.-C. Lin, Beneficial Effect of Astragaloside on Alzheimer's Disease Condition Using Cultured Primary Cortical Cells Under β -amyloid Exposure, *Mol Neurobiol.* 53 (2016) 7329–7340.
- [218] X.J. Zhang, D.S. Greenberg, Acetylcholinesterase involvement in apoptosis, *Front. Mol. Neurosci.* 5 (2012) 40.
- [219] D. Toiber, A. Berson, D. Greenberg, N. Melamed-Book, S. Diamant, H. Soreq, N-Acetylcholinesterase-Induced Apoptosis in Alzheimer's Disease, *PLoS One.* 3 (2008) e3108.
- [220] J. Savory, M.M. Herman, O. Ghribi, Intracellular mechanisms underlying aluminum-induced apoptosis in rabbit brain, *J Inorg Biochem.* 97 (2003) 151–4.
- [221] P. Kumar, A. Kumar, Protective effect of rivastigmine against 3-nitropropionic acid-induced Huntington's disease like symptoms: Possible behavioural, biochemical and cellular alterations, *Eur J Pharmacol.* 615 (2009) 91–101.
- [222] J. Greilberger, C. Koidl, M. Greilberger, M. Lamprecht, K. Schroecksnadel, F. Leblhuber, D. Fuchs, K. Oettl, Malondialdehyde, carbonyl proteins and albumin-disulphide as useful oxidative markers in mild cognitive impairment and Alzheimer's disease, *Free Radic Res.* 42 (2008) 633–638.
- [223] A.P. Porsteinsson, G.T. Grossberg, J. Mintzer, J.T. Olin, M.M.-M.-12 S. Group, Memantine treatment in patients with mild to moderate Alzheimer's disease already receiving a cholinesterase inhibitor: a randomized, double-blind, placebo-controlled trial, *Curr Alzheimer Res.* 5 (2008) 83–89.
- [224] H. Attia, S. Albuhayri, S. Alaraidh, A. Alotaibi, H. Yacoub, R. Mohamad, M. Al-Amin, Biotin, coenzyme Q10, and their combination ameliorate aluminium chloride-induced Alzheimer's disease via attenuating neuroinflammation and improving brain insulin signaling, *J Biochem Mol Toxicol.* 34 (2020) e22519.
- [225] E.M. Al-Olayan, M.F. El-khadragy, A.E.A. Moneim, The protective properties of melatonin against aluminium-induced neuronal injury, *Int J Exp Pathol.* 96 (2015) 196–202.
- [226] S.H. Alawdi, E.S. El-Denshary, M.M. Safar, H. Eidi, M.-O. David, M.A. Abdel-Wahhab, Neuroprotective Effect of Nanodiamond in Alzheimer's Disease Rat Model: a Pivotal Role for Modulating NF- κ B and STAT3 Signaling, *Mol Neurobiol.* 54 (2017) 1906–1918.
- [227] A.K. Sachdeva, K. Chopra, Lycopene abrogates $A\beta(1-42)$ -mediated neuroinflammatory cascade in an experimental model of Alzheimer's disease, *J Nutr Biochem.* 26 (2015) 736–744.
- [228] G.A. Elmegeed, H.H. Ahmed, M.A. Hashash,

- M.M. Abd-Elhalim, D.S. El-kady, Synthesis of novel steroidal curcumin derivatives as anti-Alzheimer's disease candidates: Evidences-based on in vivo study, *Steroids*. 101 (2015) 78–89.
- [229] L.T. Al Kury, A. Zeb, Z.U. Abidin, N. Irshad, I. Malik, A.M. Alvi, A.A.K. Khalil, S. Ahmad, M. Faheem, A.-U. Khan, F.A. Shah, S. Li, Neuroprotective effects of melatonin and celecoxib against ethanol-induced neurodegeneration: a computational and pharmacological approach, *Drug Des Devel Ther*. 13 (2019) 2715–2727.
- [230] R.S. Abdelrahman, M.E. Abdelmageed, Renoprotective effect of celecoxib against gentamicin-induced nephrotoxicity through suppressing NF κ B and caspase-3 signaling pathways in rats, *Chem Biol Interact*. 315 (2020) 108863.
- [231] C.E. Smith, S. Soti, T.A. Jones, A. Nakagawa, D. Xue, H. Yin, NSAIDs are Caspase Inhibitors, *Cell Chem Biol*. 24 (2017) 281–292.
- [232] S. Atari-Hajipirloo, S. Nikanfar, A. Heydari, F. Noori, F. Kheradmand, The effect of celecoxib and its combination with imatinib on human HT-29 colorectal cancer cells: Involvement of COX-2, Caspase-3, VEGF and NF- κ B genes expression, *Cell Mol Biol*. 62 (2016) 68–74.
- [233] H. Janakiraman, R.P. House, S. Talwar, S.M. Courtney, E.S. Hazard, G. Hardiman, S. Mehrotra, P.H. Howe, V. Gangaraju, V. Palanisamy, Repression of caspase-3 and RNA-binding protein HuR cleavage by cyclooxygenase-2 promotes drug resistance in oral squamous cell carcinoma, *Oncogene*. 36 (2017) 3137–3148.
- [234] C. Zhao, W. Deng, F.H. Gage, Mechanisms and functional implications of adult neurogenesis, *Cell*. 132 (2008) 645–660.
- [235] F. Marxreiter, M. Regensburger, J. Winkler, Adult neurogenesis in Parkinson's disease, *Cell Mol Life Sci*. 70 (2013) 459–473.
- [236] O. Lazarov, M.P. Mattson, D.A. Peterson, S.W. Pimplikar, H. van Praag, When neurogenesis encounters aging and disease, *Trends Neurosci*. 33 (2010) 569–579.
- [237] Y. Mu, F.H. Gage, Adult hippocampal neurogenesis and its role in Alzheimer's disease, *Mol. Neurodegener*. 6 (2011) 85.
- [238] C. Hollands, M.K. Tobin, M. Hsu, K. Musaraca, T.-S. Yu, R. Mishra, S.G. Kernie, O. Lazarov, Depletion of adult neurogenesis exacerbates cognitive deficits in Alzheimer's disease by compromising hippocampal inhibition, *Mol Neurodegener*. 12 (2017) 64.
- [239] S.-R. Cho, A. Benraiss, E. Chmielnicki, A. Samdani, A. Economides, S.A. Goldman, Induction of neostriatal neurogenesis slows disease progression in a transgenic murine model of Huntington disease, *J Clin Invest*. 117 (2007) 2889–2902.
- [240] M. Demars, Y.-S. Hu, A. Gadadhar, O. Lazarov, Impaired neurogenesis is an early event in the etiology of familial Alzheimer's disease in transgenic mice, *J Neurosci Res*. 88 (2010) 2103–17.
- [241] O. Lazarov, R.A. Marr, Of mice and men: neurogenesis, cognition and Alzheimer's disease, *Front Aging Neurosci*. 5 (2013) 43.
- [242] Z. Nagy, M.M. Esiri, A.D. Smith, Expression of cell division markers in the hippocampus in Alzheimer's disease and other neurodegenerative conditions, *Acta Neuropathol*. 93 (1997) 294–300.
- [243] A. Shruster, E. Melamed, D. Offen, Neurogenesis in the aged and neurodegenerative brain, *Apoptosis*. 15 (2010) 1415–1421.
- [244] S.H. Choi, E. Bylykbashi, Z.K. Chatila, S.W.

- Lee, B. Pulli, G.D. Clemenson, E. Kim, A. Rompala, M.K. Oram, C. Asselin, J. Aronson, C. Zhang, S.J. Miller, A. Lesinski, J.W. Chen, D.Y. Kim, H. Van Praag, B.M. Spiegelman, F.H. Gage, R.E. Tanzi, Combined adult neurogenesis and BDNF mimic exercise effects on cognition in an Alzheimer's mouse model, *Science*. 361 (2018) eaan8821.
- [245] U. Wilhelmsson, I. Lebkuechner, R. Leke, P. Marasek, X. Yang, D. Antfolk, M. Chen, P. Mohseni, E. Lasič, S.T. Bobnar, M. Stenovec, R. Zorec, A. Nagy, C. Sahlgren, M. Pakna, M. Pekny, Nestin Regulates Neurogenesis in Mice Through Notch Signaling from Astrocytes to Neural Stem Cells, *Cereb. Cortex*. 29 (2019) 4050–4066.
- [246] S. Yu, Y. Hei, W. Liu, Upregulation of seladin-1 and nestin expression in bone marrow mesenchymal stem cell transplantation via the ERK1/2 and PI3K/Akt signaling pathways in an Alzheimer's disease model, *Oncol. Lett*. 15 (2018) 7443–7449.
- [247] J. Li, Y. Han, M. Li, C. Nie, Curcumin Promotes Proliferation of Adult Neural Stem Cells and the Birth of Neurons in Alzheimer's Disease Mice via Notch Signaling Pathway, *Cell. Reprogram*. 21 (2019) 152–161.
- [248] M. Ibrahim, M. Haleem, S. AbdelWahab, A. Abdel-Aziz, Sildenafil ameliorates Alzheimer disease via the modulation of vascular endothelial growth factor and vascular cell adhesion molecule-1 in rats, *Hum. Exp. Toxicol*. (2020).
- [249] S.K. Tayebati, M.A. Di Tullio, F. Amenta, Effect of treatment with the cholinesterase inhibitor rivastigmine on vesicular acetylcholine transporter and choline acetyltransferase in rat brain, *Clin Exp Hypertens*. 26 (2004) 363–373.
- [250] A.M. Salem, H.H. Ahmed, H.M. Atta, M.A. Ghazy, H.A. Aglan, Potential of bone marrow mesenchymal stem cells in management of Alzheimer's disease in female rats, *Cell Biol Int*. 38 (2014) 1367–1383.
- [251] R. SINURAT, A.M. MASKOEN, D. HILMANTO, K. WIRIADISASTRA, ROLE OF SDF-1 AND CELECOXIB IN INCREASING QUANTITY OF NEURAL STEM CELL IN THE LESION ZONE AND OUTCOME OF SPONTANEOUS INTRACEREBRAL HEMORRHAGE, *Int. J. Pharm. Pharm. Sci*. 8 (2016) 399–403.