HISTOLOGICAL STUDY ON THE EFFECT OF ALUMINIUM CHLORIDE ON FRONTAL CORTEX OF ADULT MALE ALBINO RATS

Mohamed S. Elgendy*, Menna M. Abdel-Dayem and Manal M. Hatem ****

Histology Departments, Faculties of Medicine, Fayoum* and Cairo** Universities

Abstract

Hypothesis*: Several studies implicated aluminium in the pathogenesis of many neurodegenerative disorders especially Alzheimer disease, although the underlying histopathological changes in the brain were not clear with many controversies.* ***Objective****: So we aimed to elucidate these histological, immunohistochemical and ultrastructural changes that might occur in the rat brain after aluminium exposure.* ***Materials and methods:*** *In this study we used 18 adult male albino rats divided into 2 groups; a control group and an experimental group taking 600 mg/ kg aluminium chloride orally daily for 4 weeks. At the end of the fourth week, samples from the frontal cortex were obtained and stained with H&E, and glial fibrillary acidic protein (GFAP). Other samples were processed for electron microscopic examination. Morphometric study was done for GFAP immunostaining.* ***Results****: The group taking 600 mg/ kg aluminium chloride showed decreased body weight and had developed some neurological symptoms. Routine H&E revealed presence of some shrunken pyramidal cells with pyknotic nuclei; immunoreactivity for glial fibrillary acidic protein (GFAP) was decreased compared to control group. Ultrastructurally; some neurons showed shrunken nuclei, swelling and damage of the mitochondria and dilated saccules of Golgi apparatus and endoplasmic reticulum with the appearance of vacuolated areas in the cytoplasm with splitting of myelin sheath and degeneration of some*

nerve fibres. ***Conclusion****: Aluminium in high doses can cause alterations in neurons and nerve fibers with decreased immunoreactivity for GFAP in astrocytes in the brain, so further studies would be needed to evaluate the effects of chronic exposure in apparently healthy individuals.*

Key words: *Aluminium, Glial fibrillary acidic protein (GFAP), Neuron*

Introduction

Aluminum hydroxide Al $(OH)_3$ was widely used in nonprescription antacids and buffered aspirins, in cosmetic and antiperspirant preparations, also (Al) compounds were used to prevent hyperphosphatemia in patients suffering from renal failure [(1)]. Tap water might contain aluminum either naturally or because (Al) has been added in the treatment process as aluminum sulfate $A1_2$ $(SO_4)_3$. The amount of aluminum in diet was small compared to the amount of aluminum in pharmaceutical products, such as in antacids, dialysis fluid and some vaccines [(2)].

From human dietary balance studies; it was clear that most of the ingested Al was unabsorbed as 76-98 % of it was excreted in feces while the minimal absorbed part was excreted through the renal pathway. So the greatest danger occurs when there was overload of aluminum intake with minimal clearance in patients with impaired renal function [(3)].

Increased amounts of (Al) have been reported in the brain of subjects suffering from Alzheimer's disease [(4,5)]. Uremic persons represented a population at risk for aluminum-related dementia. Prolonged dialysis with aluminum-containing dialysates, possibly combined with oral treatment with aluminum hydroxide to control hyperphosphatemia, produced a characteristic neurotoxicity syndrome which was referred to as “dialysis dementia” [(6)]. Some epidemiologic studies suggested an association between aluminum from drinking water and dementia [(7,8,9)]. While others failed to find an association [(10,11)].

Studies on the role of aluminum-containing products (antacids and antiperspirants) and Alzheimer's disease had reported both positive and negative results [12,13].

Immunohistochemical markers were found to be useful in characterizing the progression of Al induced neurotoxicity. Glial fibrillary acidic protein (GFAP) was found to be a structural protein composed of intermediate filaments synthesized in astrocytes [14]. Changes in GFAP levels have been proposed as an index of toxicant-induced reactivity [15]. Progressive gliosis has also been found in Alzheimer's disease and aging brain [16,17].

In this study we aimed to examine the histological, immunohistochemical and ultrastructural alterations in the frontal cortex due to aluminum exposure in rats.

Materials and methods

In this study we used 18 male albino rats 200- 250 gm body weight divided into 2 groups.

Group I: formed of 6 rats putting each three rats in one cage, this group served as control group. Each animal was given 5 ml tap water by oesophageal tube daily for 4 weeks.

Group II: formed of 12 rats putting each three rats together in one cage. Each rat was given 600 mg/ kg aluminum chloride [18] dissolved in 5 ml tap water by oesophageal tube daily for 4 weeks. All animals have free access to food and water ad libitum.

At the end of the fourth week, rats (from the control and experimental aluminum treated groups) were anesthetized with thiopental sodium, and then intracardiac saline irrigation of rats was done followed by formalin perfusion. The brains were dissected out, fixed in formalin, and then embedded in paraffin. Some specimens were fixed in gluteraldehyde and prepared for electron microscopic study.

Histological study: Sections were cut at 6 microns and stained with H& E for routine histological examination.

Immunohistochemical study: Staining was performed for glial fibrillary acidic protein (GFAP) as an indicator for glial reactivity. Primary antibodies monoclonal mouse anti-GFAP were purchased from Dako (Carpenteria, CA, U.S.A.). Sections were treated with 0.01 M citrate buffer (pH 6.0) for 10 minutes to unmask antigen [19]. Sections were incubated in 0.3% H_2O_2 for 30 min to abolish endogenous peroxidase activity before blocking with 5% horse serum for 1–2 h. Slides were incubated with the primary antibody (1:500 monoclonal mouse anti-GFAP) at 4°C for 18–20 h and after washing, they were incubated with biotinylated secondary antibodies (ABC kit, 1: 200) and then with avidin–biotin complex. Finally, sections were developed with 0.05% diaminobenzidine. Slides were counterstained with hematoxylin before mounting.

Morphometric Study: The data were obtained by using "Leica Qwin 500" image analyzer computer system (England). We measured the relative area percent and the relative optical density for GFAP immunoreactivity.

Statistical Analysis:

Five fields per slide and five slides per animal were studied to collect data, and then statistical analysis of the results of morphometry was performed on an IBM PC using the statistical software "Excel". Results were expressed as mean ± SEM values. Comparison between the groups was made using (t-test: two samples for means). Results were considered significant when probability (p) was <0.05.

Electron microscopic examination:

After anaesthesia, intracardiac perfusion with fixative solution (2.5% glutaraldehyde in 0.1M cacodylate buffer, pH 7.4) was done for some

animals from the experimental aluminum treated and control groups. Small tissue blocks (1 mm^3 volume) were taken from the frontal cortex. Fixed in the same fixative solution for 2.5 h and washed in the same buffer for 18 h. They were then post fixed in 2% osmium tetroxide in 0.1 M cacodylate buffer (pH 7.4) for 1 h. After dehydration in ethanol and propylene oxide, samples were embedded in Epon. Ultrathin sections were double stained with uranyl acetate and lead citrate, and examined using transmission electron microscope [20].

Results

General appearance

By the end of the third week of treatment, animals treated with 600 mg aluminum/ kg/ day showed statistically significant decrease in the body weight (table 1) and developed some neurological symptoms in the form of loss of appetite, decreased activity, forward head tilt and hemiplegic's gait in some animals.

Histological and immunohistochemical results

In the control group, staining with H&E showed the general histological structure of cerebral cortical layers which were the molecular layer that was seen to be formed of parallel fibers and few cells between the fibers; then the outer granular layer which appeared to be formed of small granular and stellate nerve cells; after that the outer pyramidal layer which was formed of medium sized pyramidal cells; then the inner granular layer with its small stellate nerve cells; and the inner pyramidal layer with its large sized pyramidal cells; and lastly the polymorphic cell layer having different types and sizes of nerve cells. In all these layers the neurons appeared with large nuclei and basophilic cytoplasm while smaller glia cells were scattered in between neurons and also blood vessels lined by simple squamous epithelium could be demonstrated (Figs. 1& 2). In the group taking 600 mg aluminum/ kg/ day, some of

pyramidal cells were shrunken and had deeply stained pyknotic nuclei (Fig. 3).

Immunohistochemical results:

Immuno-staining for glial fibrillary acidic protein (GFAP) in the control group showed astrocytes with different branching patterns of reactive processes (Fig. 4), while the immuno-staining for glial fibrillary acidic protein (GFAP) in aluminium treated group showed milder reactivity (Fig. 5). The relative optical density and relative cell area percent of immunolabeled cells with GFAP was found to be significantly decreased as detected with the morphometric image analysis and statistical analysis ($p<0.05$) tables (2 & 3).

Ultrastructural results

In the control group, the normally appearing large pyramidal cells with large euochromatic nuclei could be observed. These cells were rich in different organelles as mitochondria, rough endoplasmic reticulum, ribosomes, Golgi apparatus and lysosomes (Fig. 6). In addition to that parts of the nerve fibers which were either myelinated or unmyelinated fibers appeared with electron dense synaptic contacts, the axon terminal contained mitochondria and synaptic vesicles indented by dendritic spines (Fig. 7).

In the group taking 600 mg/ kg the ultrastructural study revealed that some pyramidal cells showed irregular nuclear envelop with irregular distribution of chromatin, while the cytoplasm showed many vacuolated areas while the myelin sheath around showed splitting and degeneration in some nerve fibers (Figs. 8 & 9). There was also swelling and damage of the mitochondria and dilated saccules of Golgi apparatus and endoplasmic reticulum with the appearance of many heterolysosomes and autolysosomes (Fig. 9). The granular cells with smaller nuclei were also affected as the cytoplasm showed significantly decreased electron density

with the presence of markedly swollen mitochondria, markedly dilated rough endoplasmic reticular cisterna and many clear vesicles with the appearance of many primary and secondary lysosomes (Fig. 10).

Discussion

Many studies found an association between oral exposure to aluminum and an increased risk of Alzheimer's disease [21].In contrast, several studies did not find a significant association between aluminum exposure and the risk of Alzheimer's disease [10, 11]. In an attempt to develop an animal model of aluminum overload, rats were co-exposed to massive amount of citrate, compared to normal human citrate intake, so enhancing rat's brain uptake of aluminum, thus inducing histological alterations in the brain [22]. Similarly, the brain alterations were observed after administration of aluminum fluoride in drinking water which was prepared to form an optimum fluoro-aluminum level capable of crossing the gut and vascular brain barriers [23].

Numerous mechanistic studies of aluminum neurotoxicity have been performed but no single unifying mechanism has been identified [24].

In this study we examined the effect of aluminum intoxication on frontal cortical histological structure and immuno-histochemical study of the glial fibrillary acid protein (GFAP) expression in rats as well as the ultrastructural alterations in the frontal cortex. We found that aluminum caused shrinkage of many pyramidal cells and their nuclei became pyknotic. While the glial marker (glial fibrillary acidic protein (GFAP) showed decreased reactivity in aluminum treated group than control group, also the relative optical density and reactive cell area percent of immunolabeled cells with GFAP was found to be significantly decreased. There was great similarity between aluminum induced neurotoxicity and aging effects, so aluminum might induce acceleration of the aging

process and might cause apoptotic changes in the neurons and glia cells [25].

There was also in vitro increased expression of apoptosis markers under the effect of aluminum [26]. In addition to that, aluminum can induce degeneration of astrocytes via apoptosis resulting in neuronal death and that this toxicity was critical in determining neuronal degeneration and death [27]. Aluminum can also induce atrophy and apoptosis of the neurons in cerebral cortex and hippocampus [28]. This was similar to our results that shrunken pyramidal cells nuclei might be due to apoptotic changes. In contrast to that, other studies found that apoptosis did not appear to be a major cause of cell death from aluminum toxicity [29].

Lower level of GFAP following aluminum treatment was described in cell culture systems using isolated astrocytes derived from newborn rat cerebral cortex [30]. Gene transcription of GFAP in human neocortical astrocytic nuclei has been found to be inhibited by nanomolar concentrations of aluminum [31]. These results were in agreement with the results obtained in our study suggesting that a significant component of aluminum toxicity might be related to astrocytic changes as evident by decreased (GFAP) reactivity. Another study showed that aluminum had no effect on GFAP mRNA expression in rabbits [32].

Moreover, after in vitro exposure of intact rat brain to aluminum, selective astrocytic necrosis has been reported to be induced, as judged by disappearance of anti-GFAP reactivity [33].

On the other hand, enhanced expression of GFAP levels reflecting reactive gliosis has been recently described in rabbit frontal cortex 40 days after intravenous administration of aluminum lactate [34]. While intracisternal dosing of aluminum chloride to rabbits has shown no change of GFAP levels after long-term treatment [35].

These apparent discrepancies might be due to difference in GFAP expression in response to aluminum among various species and might be dose related or associated with other factors as citrate metabolism. In the rabbit, a species susceptible to aluminum induced neurofibrillary degeneration [36], reactive astrogliosis occurs, but in the rat aluminum can induce encephalopathy in the absence of neurofilamentous aggregates. So, decreased GFAP production could reflect astrocytic dysfunction rather than an inflammatory response [37]. Another explanation might be that the induction and maintenance of GFAP and its mRNA were by separate mechanisms, which were responsive to differing types of tissue damage [15]. These discrepancies could be due to sequences of astroglial response starting from reactive gliosis with hyperactivity to combat the toxic insult, but with continuous exposure or high doses, failure of astroglial resistance to the toxic insult might lead to decreased reactivity followed by death.

The aluminum-related responses of GFAP was paralleled by the finding that the activity of glutamine synthetase, an astroglial enzyme, was depressed in the same area and at the same time that GFAP levels were reduced. The time course of changes both in levels of GFAP and in glutamine synthetase activity suggests that they took place in a gradual and progressive manner. So reduced levels of this enzyme might be an important mechanism of aluminum effect on astrocytic GFAP expression [38].

The ultrastructural changes observed in our study in the neurons revealed degenerative changes in the neuronal organelles without an inflammatory response similar to apoptosis as evident by the irregular nuclear envelop in some pyramidal cells with abnormal chromatin distribution, vacuolation of cytoplasm, disruption of organelles as mitochondria, rough endoplasmic reticulum and increased lysosomal

activity in addition to splitting of myelin sheath. These results were in accordance with the recorded ultrastructural degenerative manifestations in the brain in patients with dialysis encephalopathy due to aluminum overload [39]. So, evidences were accumulating that apoptosis might be responsible for the neuronal loss associated with aluminum toxicity similar to some other neurodegenerative disorders [40].

Tables

Table 1 - Average body weight changes

body weight	Group I (control)	Group II (alum)	p value
average±SEM	247.7±1. 0747	228.75±0.858778	0.001188*

Marked differences*: significant at p <0 .05

Table 2 - relative optical density of GFAP positive cells

mean optical density±SEM	Group I (control)	Group II (alum)	p value
GFAP	0.923±0.001	0.55±0.018	0.000027*

Marked differences*: significant at p <0 .05

Table 3 - area percent analysis of GFAP positive cells

mean area	Group I (control)	Group II (alum)	p value
GFAP	6.11±0.497	1.598±0.305	0.0038*

Marked differences*: significant at p <0 .05

Figures

(Fig. 1): showing the general histological structure of cerebral cortical layers which are the molecular layer (Mol), the outer granular layer (O_Gr), the outer pyramidal layer (O_Py), the internal granular layer (I_Gr) and the internal pyramidal layer (I_Py) with its large sized pyramidal cells. Group I. H&E x100

(Fig. 2): showing the cells in the inner pyramidal layer with the large sized pyramidal cells with open face nuclei (thick arrows) and scattered neuroglia cells (thin arrows) while the blood capillaries lined by simple squamous epithelium could be demonstrated (arrow heads). The large

sized pyramidal cells with open face nuclei are demonstrated with higher magnification in the insert. Group I. H&E x400 & insert x1000

(Fig. 3): showing shrunken pyramidal cells with darkly stained pyknotic nuclei (thick arrows) in the inner pyramidal layer, with scattered neuroglia cells (thin arrows) in addition to the blood capillaries lined by simple squamous epithelium (arrow heads). The shrunken pyramidal cells with darkly stained pyknotic nuclei are demonstrated with higher magnification in the insert. Group II. H&E x400 & insert x1000

(Fig. 4): showing the positive immunostaining for glial fibrillary acidic protein in glia cells (arrows). Group I. GFAP immunostaining X400

(Fig. 5): showing diminished immunostaining for glial fibrillary acidic protein in glia cells (arrows). Group II. GFAP immunostaining X400

(Fig. 6): showing a pyramidal cell nucleus (N) with electron lucent euchromatin, the dendritic pole of the nucleus is wrinkled (curved arrow) and capped by a small Nissle body and also lysosomes and residual body could be observed (dashed arrows). There are also mitochondria (thick arrows), saccules of rough endoplasmic reticulum (thin arrows), and scattered free ribosomes. A neighbouring nerve fibre (F) could also be demonstrated. Group I. TEM x8000

(Fig. 7): showing different organelles in neuron and nerve fibers (F) which are either myelinated or unmyelinated fibers with electron dense synaptic contacts (arrow heads). The axon terminal contains mitochondria and synaptic vesicles indented by dendritic spines (axodendritic synapse) (D). Group I. TEM x8000

(Fig. 8): showing pyramidal cell with an irregular nucleus (N) and irregular distribution of chromatin, while the cytoplasm was vacuolated. It is surrounded by myelinated nerve fibers from neighbouring neurons with splitting of myelin sheath (arrows). Group II. TEM x8000

(Fig. 9): showing pyramidal cell with normally appearing nucleus (N) with clear nucleolus, swollen and partially damaged mitochondria (thin arrows) and dilated saccules of Golgi apparatus (G) in addition to the presence of many heterolysosomes (thick arrows) and autolysosomes (arrow head) and also degeneration of some nerve fibers (dashed arrows). Group II. TEM x8000

(Fig. 10): showing a markedly swollen granular cell, surrounded by parts of neurons rich in mitochondria, the granular cell cytoplasm shows significantly decreased electron density and contains clear vesicles (V) and numerous markedly dilated perinuclear rough endoplasmic reticular cisterna (thin arrows) and markedly swollen mitochondria (M) with the appearance of many lysosomes (thick arrows). Group II. TEM x8000

References

1. **Jones XC and Bennett BG (1986):** Exposure of man to environmental aluminium, an exposure commitment assessment. Sci. Total Environ., 52:65-82.
2. **Bergfors E, Trollfors B and Inerot A (2003):** Unexpectedly high incidence of persistent itching nodules and delayed hypersensitivity to aluminium in children after the use of adsorbed vaccines from a single manufacturer. Vaccine, 22:64-9.
3. **Gorsky JE, Dietz AA, Spencer H and Osis D (1979):** Metabolic balance of aluminium studied in six men. Clin. Chem., 25:1739-1343.
4. **Crapper McLachlan DR, Lukiw WJ and Kruck TPA (1990):** Aluminium, altered transcription, and the pathogenesis of Alzheimer's disease. Environ. Geochem. Health, 12:103- 114.
5. **Flaten TP (2001):** Aluminium as a risk factor in Alzheimer's disease, with emphasis on drinking water. Brain Res Bull, 55:187-96.
6. **Alfrey AC (1993):** Aluminium toxicity in patients with chronic renal failure. Ther Drug Monit, 15:593- 597.
7. **Martyn CN, Barker DJP and Osmond C (1989):** Geographical relation between Alzheimer's disease and aluminium in drinking water. Lancet, 1:59–62.

8. **Flaten TP (1990):** Geographical associations between aluminium in drinking water and death rates with dementia (including Alzheimer's disease), Parkinson's disease and amyotrophic lateral sclerosis in Norway. Environ Geochem Health, 12:152–67.
9. **McLachlan DRC, Bergeron C and Smith JE (1996):** Risk for neuropathologically confirmed Alzheimer's disease and residual aluminium in municipal drinking water employing weighted residential histories. Neurology, 46:401–5.
10. **Forster DP, Newens AJ and Kay DWK (1995):** Risk factors in clinically diagnosed presenile dementia of the Alzheimer type: a case-control study in northern England. J Epidemiol Community Health, 49:253–8.
11. **Martyn CN, Coggon DN and Inskip H (1997):** Aluminium concentrations in drinking water and risk of Alzheimer's disease. Epidemiol, 8:28 l-286.
12. **Colin-Jones D, Langman MJS and Lawson DH (1989):** Alzheimer's disease in antacid users. (Letter). Lancet, 1:1453.
13. **Graves AB, White E and Koepsell TD (1990):** The association between aluminium-containing products and Alzheimer's disease. J Clin Epidemiol, 43:35–44.
14. **Bignami A and Dahl D (1976):** The astroglial response to stabbing. Immunofluorescence studies with antibodies to astrocyte-specific protein (GFA) in mammalian and submammalian vertebrates. Neuropathol. Appl. Neurobiol., 2: 99–110.
15. **Norton W T, Aquino DA, Hozumi I, Chiu FC and Brosnan CF (1992):** Quantitative aspects of reactive gliosis: a review. Neurochem. Res., 17: 877–885.
16. **Beach TG, Walker R, and McGeer EG (1989):** Patterns of gliosis in Alzheimer's disease and aging cerebrum. Glia, 2: 420–436.
17. **Delacourte A (1990):** General and dramatic glial reaction in Alzheimer brains. Neurology, 40: 33–37.
18. **Nedzvetsky VS, Tuzcu M, Yasar A, Tikhomirov AA and Baydas G (2006):** Effects of vitamin E against aluminum neurotoxicity in rats. Biochemistry (Mosc), 71(3): 239-44.
19. **Cattoretti G, Pileri S, Parravicini C, Becker MH, Poggi S, Bifulco C, Key G, D'Amato L, Sabattini E, Feudale E, Reynolds F, Gerdes J and Rilke F (1993):** Antigen unmasking on formalin-fixed, paraffin-embedded tissue sections. J. Pathol., 171: 83–98.
20. **Sanbatini DD, Bensch K and Barrnett RJ (1963):** Chemistry and electron microscopy. The preservation of cellular ultrastructure and enzymatic activity by aldehyde fixation. J Cell Biol, 17: 19 - 58.

21. **Michel P, Commenges D and Dartigues JF (1990):** Study of the relationship between Alzheimer's disease and aluminium in drinking water. Neurobiol Aging, 11:264.
22. **Florence AL, Gauthier A and Ponsar C (1994):** An experimental animal model of aluminium overload. Neurodegeneration, 3:315-323.
23. **Varner JA, Jensen KF and Horvath W (1998):** Chronic administration of aluminium-fluoride or sodium fluoride to rats in drinking water: alterations in neuronal and cerebrovascular integrity. Brain Res, 784:284-298 .
24. **Strong, MJ, Garruto RM and Joshi JG (1996):** Can the mechanisms of aluminium neurotoxicity be integrated into a unified scheme? J Toxicol. Environ. Health, 48:599-613.
25. **Deloncle R, Huguet F, Fernandez B, Quellard N, Babin P and Guillard O (2001):** Ultrastructural study of rat hippocampus after chronic administration of aluminium L-glutamate: an acceleration of the aging process. Exp Gerontol., 36(2):231-44.
26. **Szutowicz A (2001):** Aluminium, NO, and nerve growth factor neurotoxicity in cholinergic neurons. J Neurosci Res., 66(5):1009-18.
27. **Suarez-Fernandez MB, Soldado AB, Sanz-Medel A, Vega JA, Novelli A and Fernandez-Sanchez MT (1999):** Aluminium-induced degeneration of astrocytes occurs via apoptosis and results in neuronal death. Brain Res., 835(2):125-36.
28. **Abd-Elghaffar SKh, El-Sokkary GH and Sharkawy AA (2005):** Aluminium-induced neurotoxicity and oxidative damage in rabbits: protective effect of melatonin. Neuro Endocrinol Lett., 26(5):609-16.
29. **Brenner S (2002):** Aluminium neurotoxicity is reduced by dantrolene and dimethyl sulfoxide in cultured rat hippocampal neurons. Biol Trace Elem Res., 86(1):85-9.
30. **Norenberg MD, Norenberg LOB, Cowman GA, McCarthy M, and Neary JT (1989):** The effects of aluminium on astrocytes in primary culture. J Neuropathol. Exp. Neurol., 48: 374.
31. **Lukiw WJ, LeBlanc HJ, Carver LA, McLachlan DRC and Bazan NG (1998):** Run-on gene transcription in human neocortical nuclei: inhibition by nanomolar aluminium and implications for neurodegenerative disease. J. Mol. Neurosci., 11: 67–78.
32. **Parhad IM., Krekoski CA., Mathew A., and Tran PM (1989):** Neuronal gene expression in aluminium myelopathy. Cell. Mol. Neurobiol., 9: 123–138.
33. **Struys-Ponsar C, Florence A, Gauthier A, Crichton RR and van den Bosch de Aguilar P (1994):** Ultrastructural changes in brain

parenchyma during normal aging and in animal models of aging. J. Neural Transm. Suppl., 44: 111–132.
34. **Yokel RA. and O'Callaghan JP (1998):** An aluminium-induced increase in GFAP is attenuated by some chelators. Neurotoxicol. Teratol., 20: 55–60.
35. **Strong MJ, Gaytan-Garcia S and Jakowec DM (1995):** Reversibility of neurofilamentous inclusion formation following repeated sublethal intracisternal inoculums of AlCl3 in New Zealand white rabbits. Acta Neuropathol. (Berl.), 90: 57–67.
36. **Savory J, Huang Y, Herman MM, Reyes MR and Wills MR (1995):** Tau immunoreactivity associated with aluminium maltolate induced neurofibrillary degeneration in rabbits. Brain Res., 669: 325–329.
37. **Lipman JJ, Colowick SP, Lawrence PL and Abumrad NN (1988):** Aluminium induced encephalopathy in the rat. Life Sci., 42: 463–475.
38. **Aksenov MY, Aksenova MV, Butterfield DA., Hensley K, Vigo-Pelfrey C, and Carney JM (1996):** Glutamine synthetase induced enhancement of b-amyloid peptide Ab(1– 40) neurotoxicity accompanied by abrogation of fibril formation and Ab fragmentation. J. Neurochem., 66: 2050–2056.
39. **Kawahara M (2005):** Effects of aluminium on the nervous system and its possible link with neurodegenerative diseases. J Alzheimers Dis., 8(2):171-182.
40. **Marx J (2001):** Neuroscience. New leads on the how of Alzheimer's. Science, 293: 2192–2194.

دراسة هستولوجية عن تأثير كلوريد الألومنيوم على القشرة المخية الأمامية لذكورالفئران البيضاء البالغة

محمد صلاح الجندي*– منة محمد عبد الدايم – منال محمد حاتم****
قسم الهستولوجيا – كلية الطب – جامعة الفيوم* و جامعة القاهرة**

قد أشير إلىالألومنيوم فى كثير من الدراسات كمسبب للعديد من أمراض حؤول الأعصاب و خاصة مرض عته الزهايمر و لكن التغيرات النسيجية المرضية فى المخ غير واضحة و بها الكثير من المتضادات.

ولهذا فقد هدفنا فى هذه الدراسة إلى إظهار هذه التغيرات النسيجية و الكيميائية النسيجية المناعية و التغيرات فى البنية المستدقة الحادثة فى مخ الفأر بعد التعرض للألومنيوم.

و فى هذه الدراسة تم إستخدام ١٨ فأر ذكر أبيض و قد تم تقسيمهم مجموعة ضابطة و مجموعة إختبارية أعطى كل فأر منها ٦٠٠ ملجم كلوريد ألومنيوم لكل كجم من وزنه يومياً لمدة أربعة أسابيع و ذلك عن طريق أنبوبة بلعومية. و فى نهاية الأسبوع الرابع تم أخذ عينات من القشرة المخية الأمامية وتم تحضيرها و صبغتها بالهيماتوكسلين و الإيوسين، و كذلك الصبغة المناعية للدلالة المضادة للبروتين الحامضى الخيطى الدبقى و أعدت دراسة إحصائية لهذه الصبغة المناعية بعد دراستها بجهاز تحليل الصورة . و فد تم أخذ بعض العينات و تحضيرها للفحص بواسطة الميكروسكوب الإلكترونى.

و قد أظهرت نتائج البحث فى المجموعة الثانية المعطاه ألومنيوم حدوث إنخفاض فى وزن الجسم مع وجود بعض الأعراض العصبية. و أما الفحص الروتينى بالهيماتوكسلين و الإيوسين فقد أظهر حدوث إنكماش فى بعض الخلايا العصبية مع تقلص فى أنويتها فى حين ظهر ضعف الإستجابة المناعية لمضادات البروتين الحامضى الخيطى الدبقى الخاص بالخلايا النجمية و ذلك حين مقارنتها بالمجموعة الضابطة. و قد أظهرت دراسة البنية المستدقة حدوث إنتفاخ فى حشوة الخلايا العصبية و ظهور مناطق مفرغة فيها مع إنكماش فى أنوية الخلايا و تحطم للحبيبات الخيطية و حدوث تمدد فى جريبات جهاز جولجى و شبكة هيولى الباطنية فى الجبلة الداخلية للخلايا العصبية مع تكون فراغات فى غشاء الألياف العصبية و ضمور فى بعض هذه الألياف.

ونستخلص من ذلك أن الجرعات العالية للألومنيوم من الممكن أن تحدث تغيرات فى الخلايا و الألياف العصبية وكذلك نقص البروتين الحامضى الخيطى الدبقى فى الخلايا النجمية و لذلك فالدراسات المستقبلية ضرورية لتقييم تأثير التعرض المزمن لمن تبدو عليهم علامات الصحة السليمة.

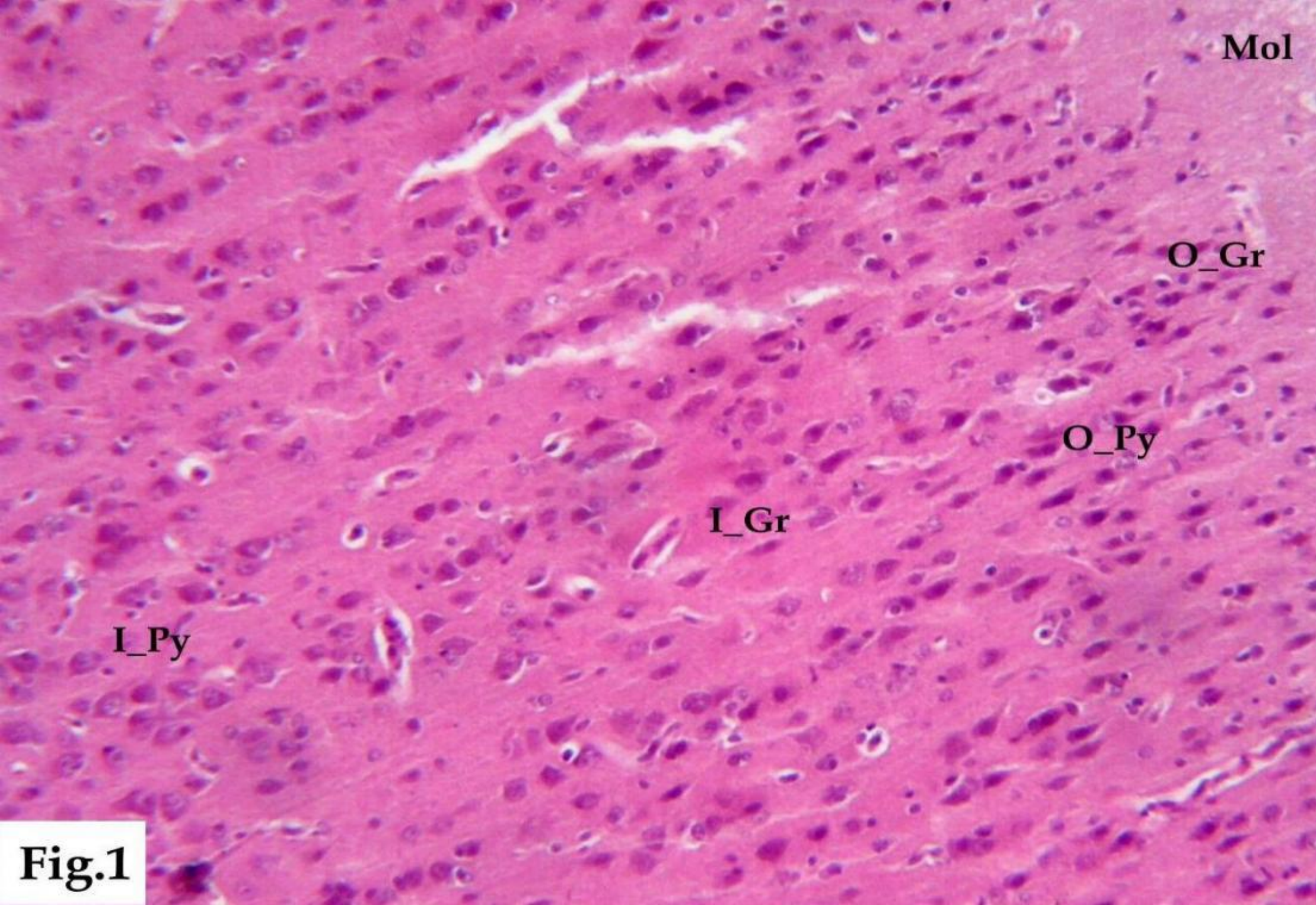

Fig.1

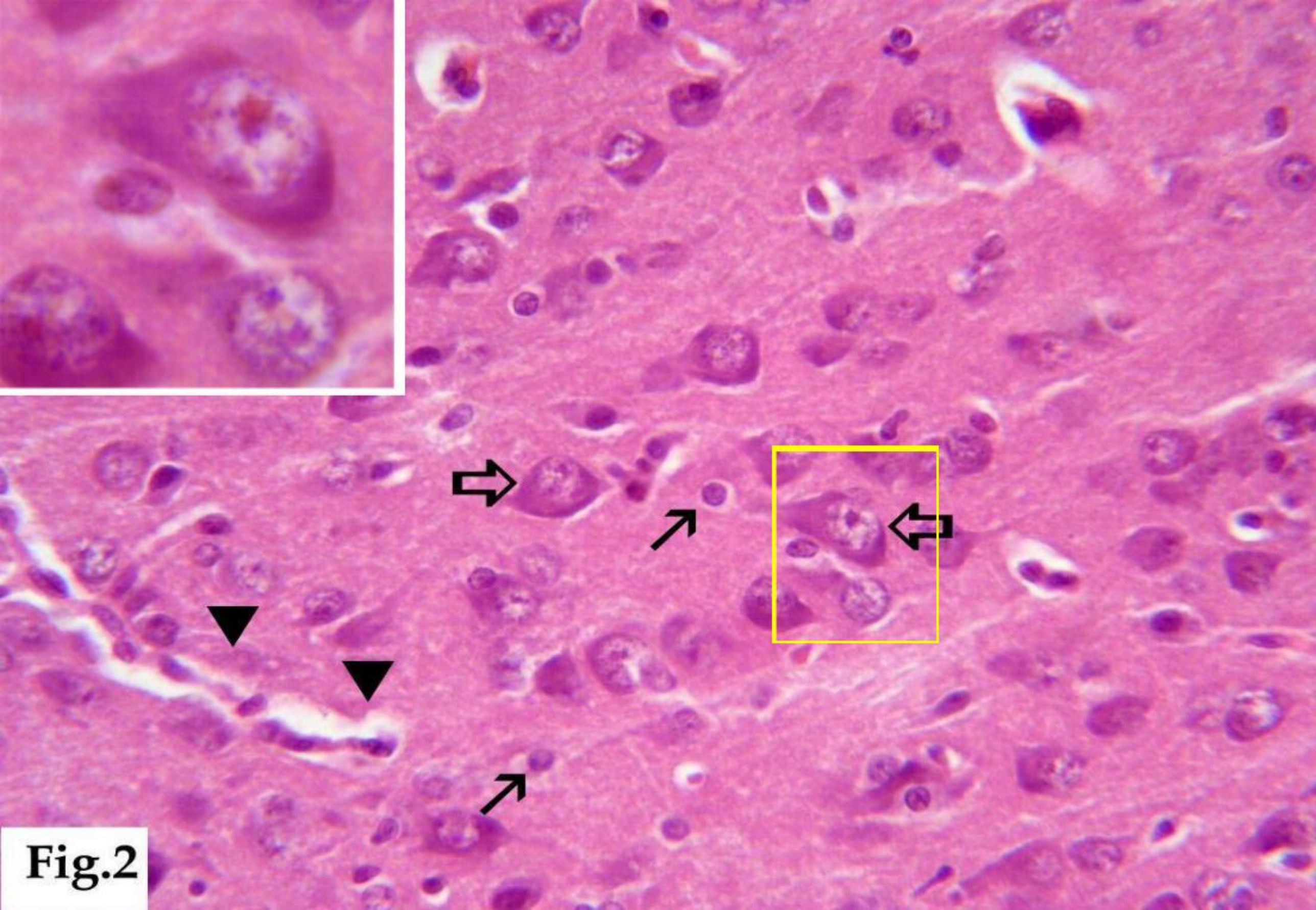

Fig.2

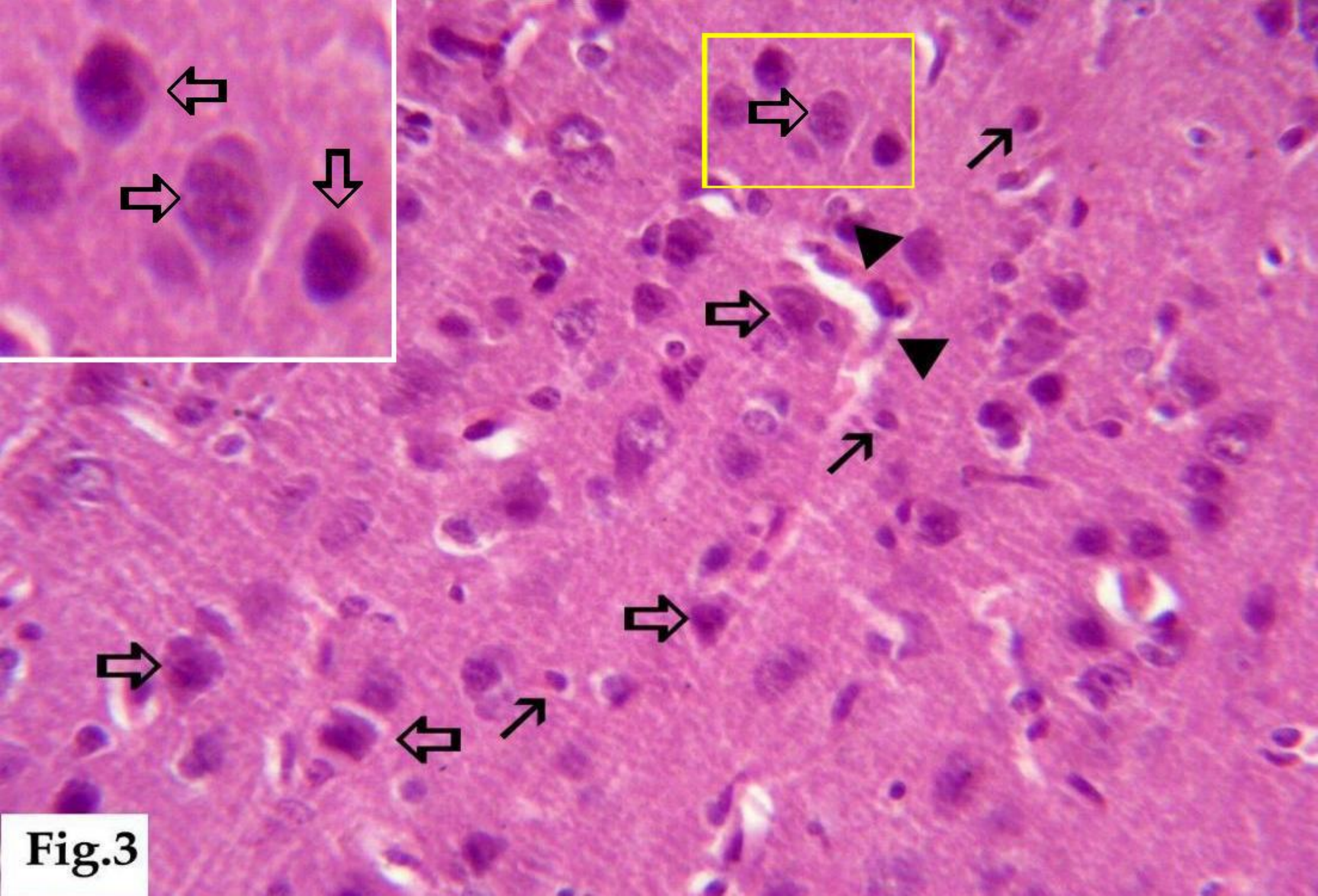

Fig.3

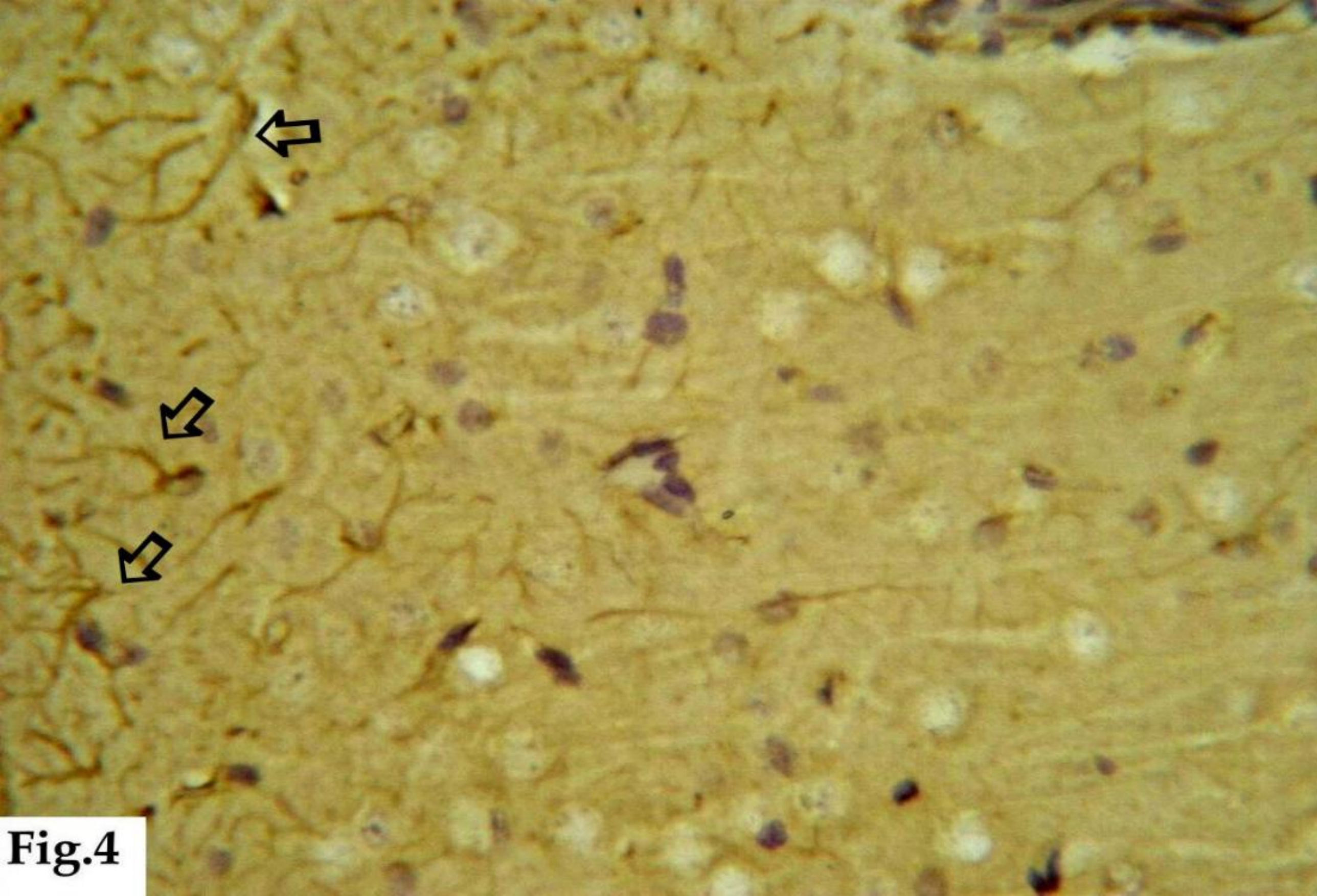

Fig.4

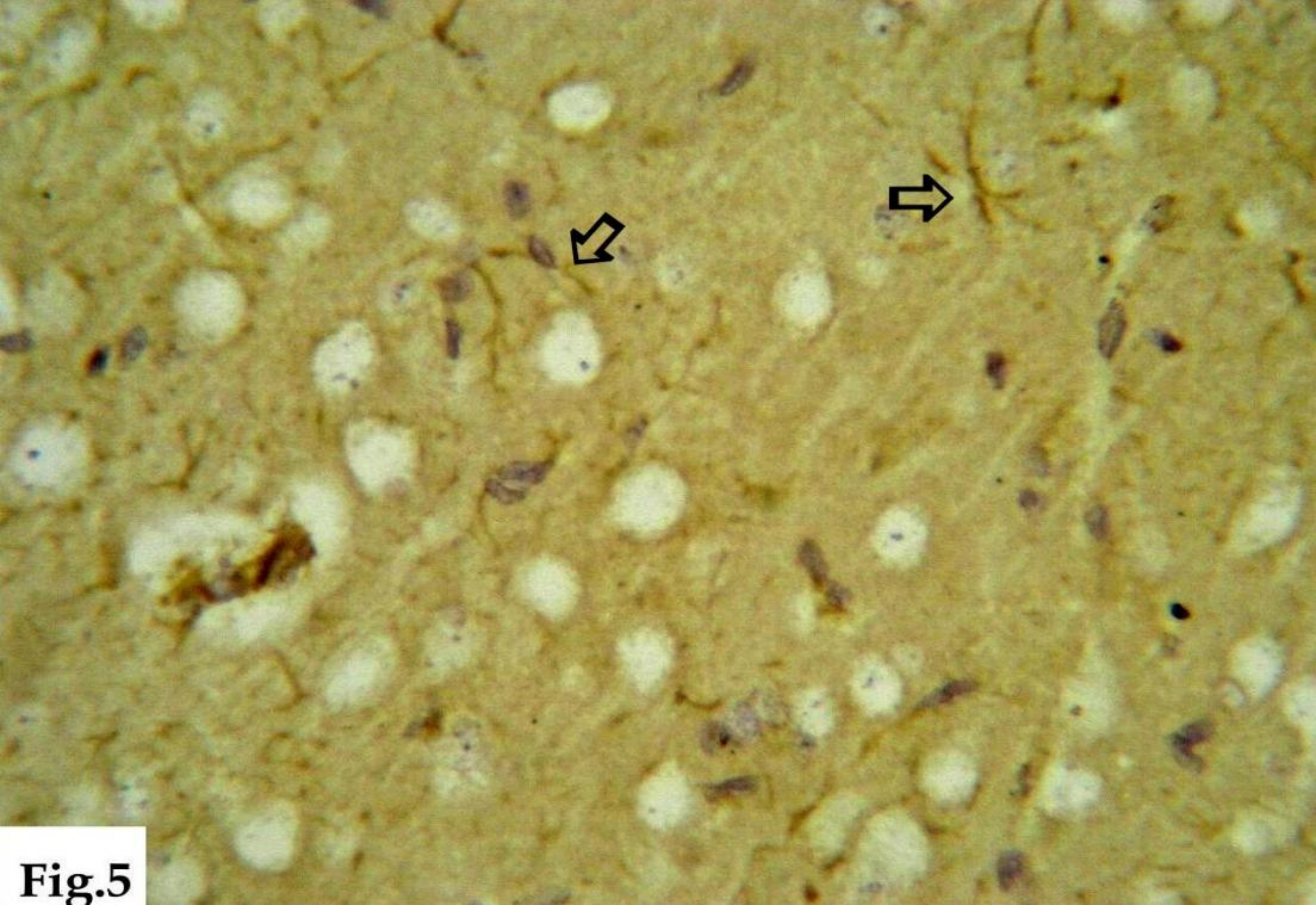

Fig.5

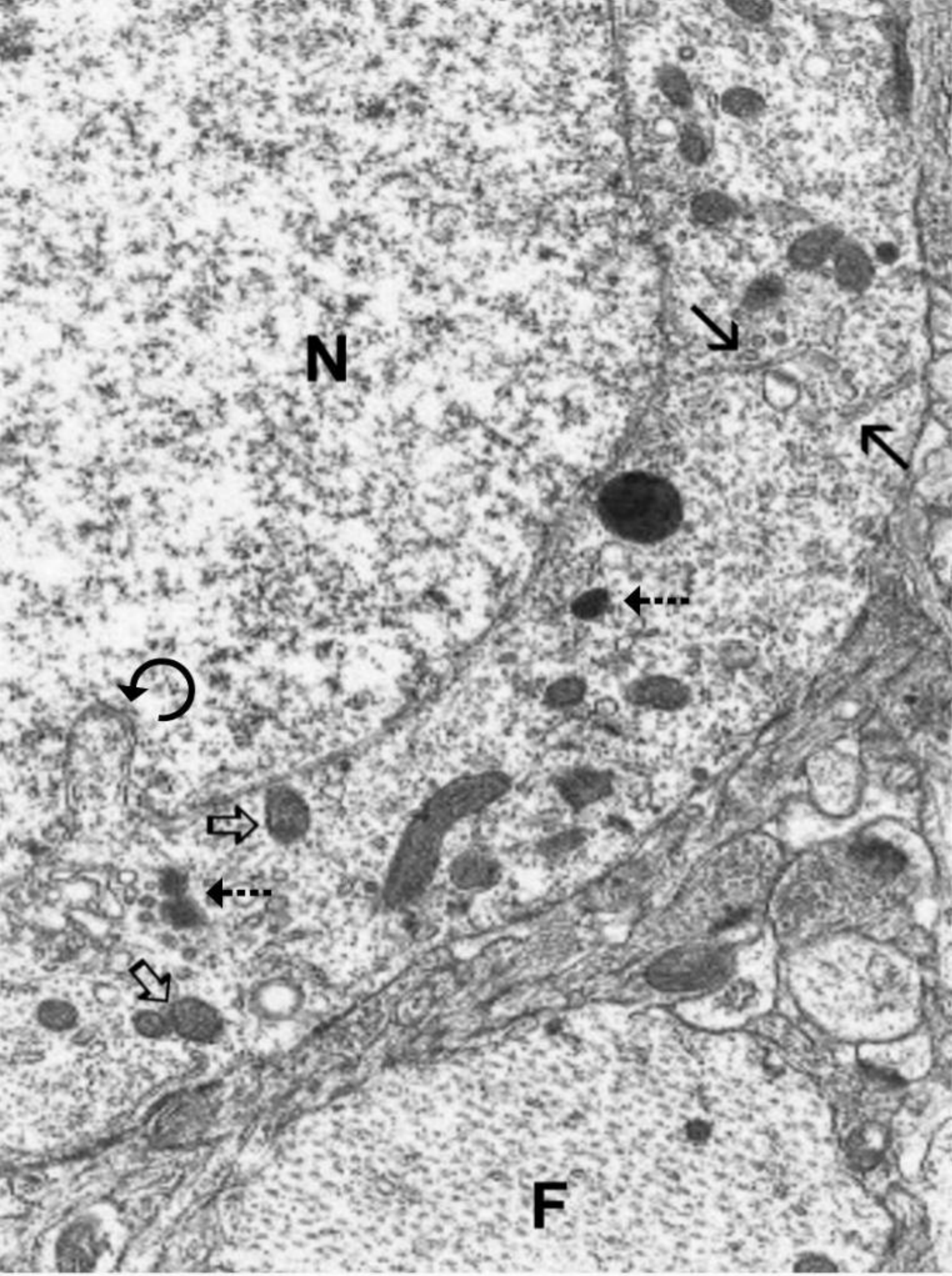

Fig.6

500 nm

Print Mag = 42347x @ 120 mm

TEM Mag = 8000x

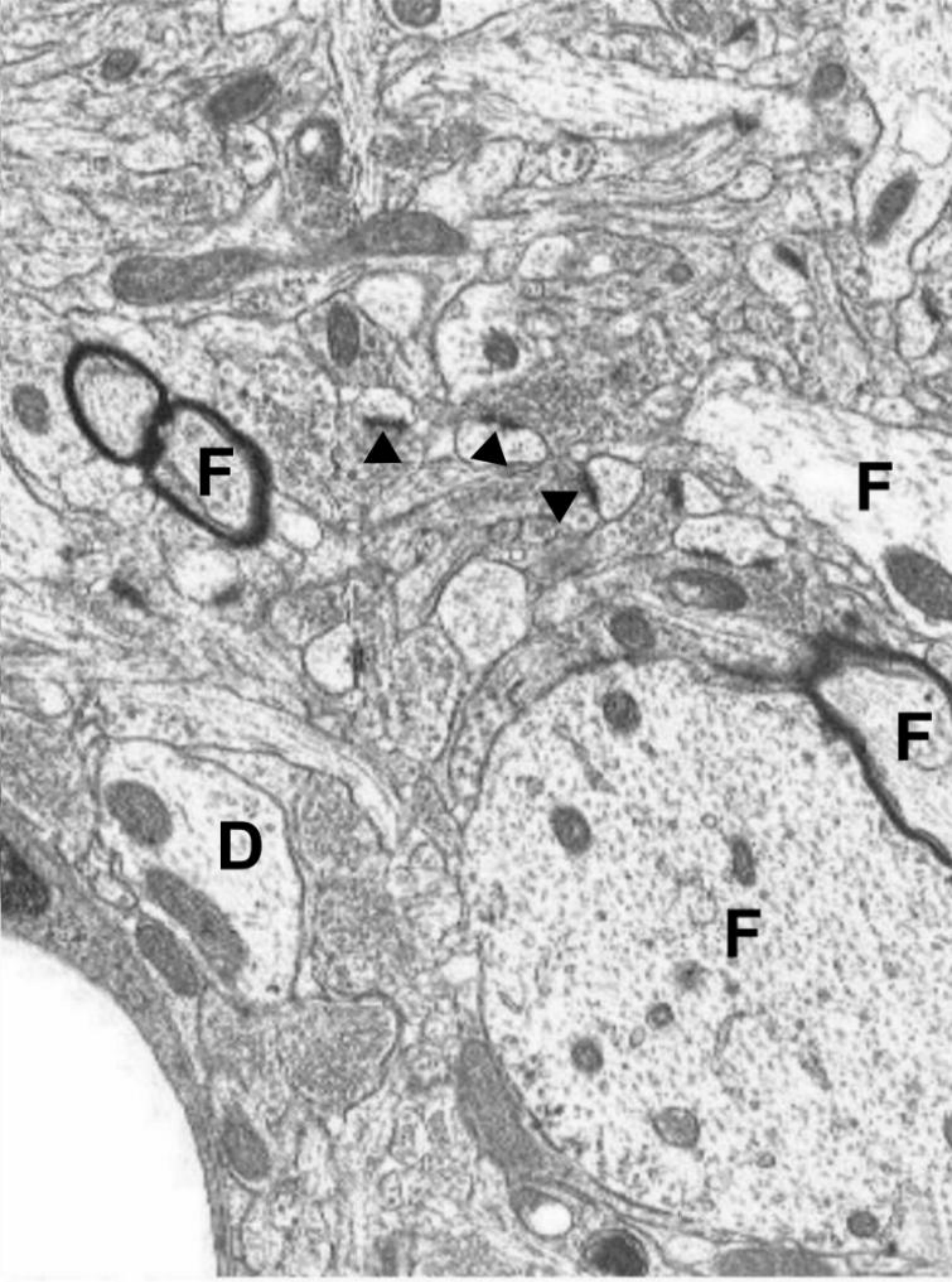

Fig.7

Print Mag = 42347x @ 120 mm

500 nm

TEM Mag = 8000x

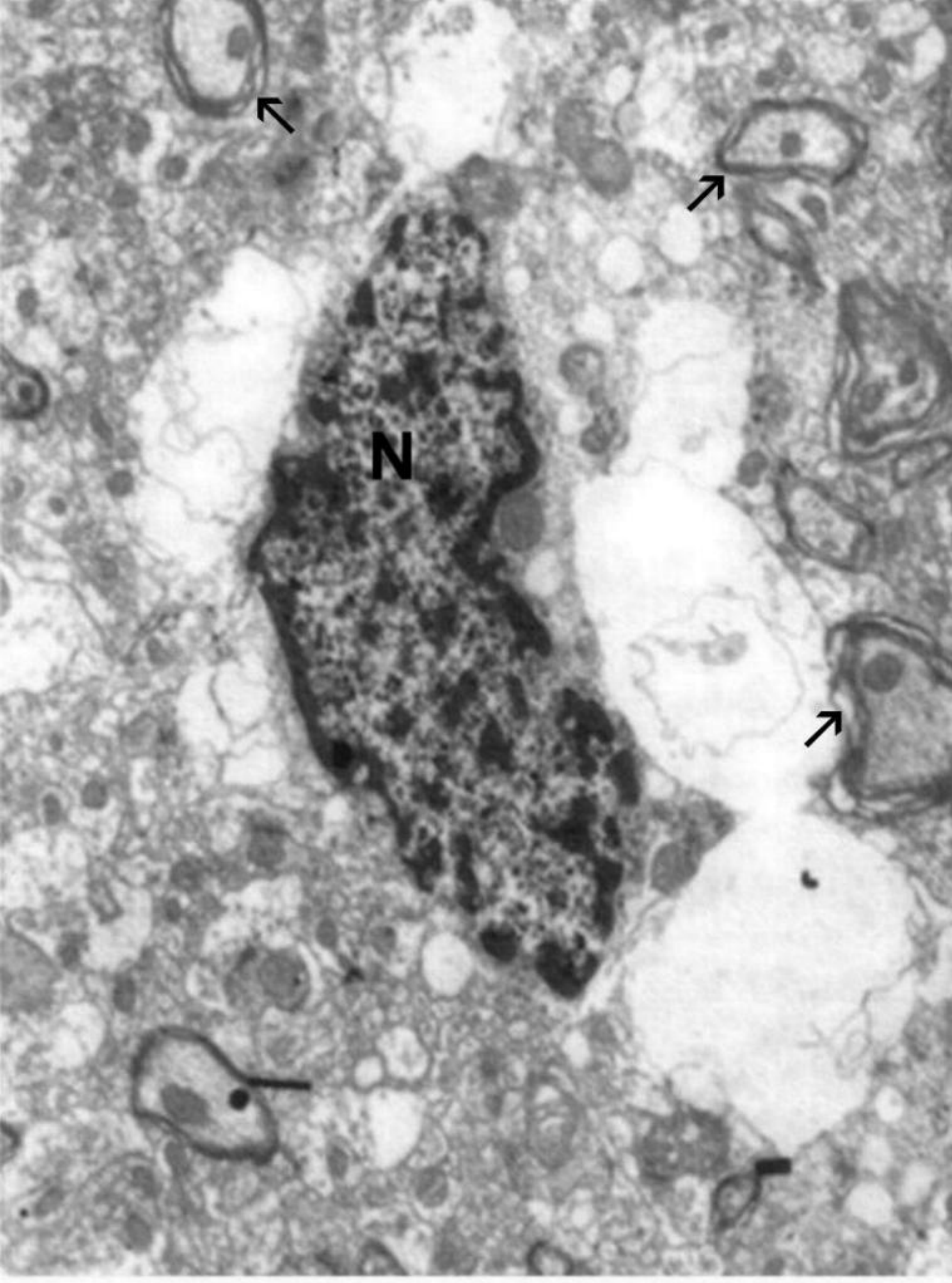

Fig.8

500 nm

Print Mag = 42347x @ 120 mm

TEM Mag = 8000x

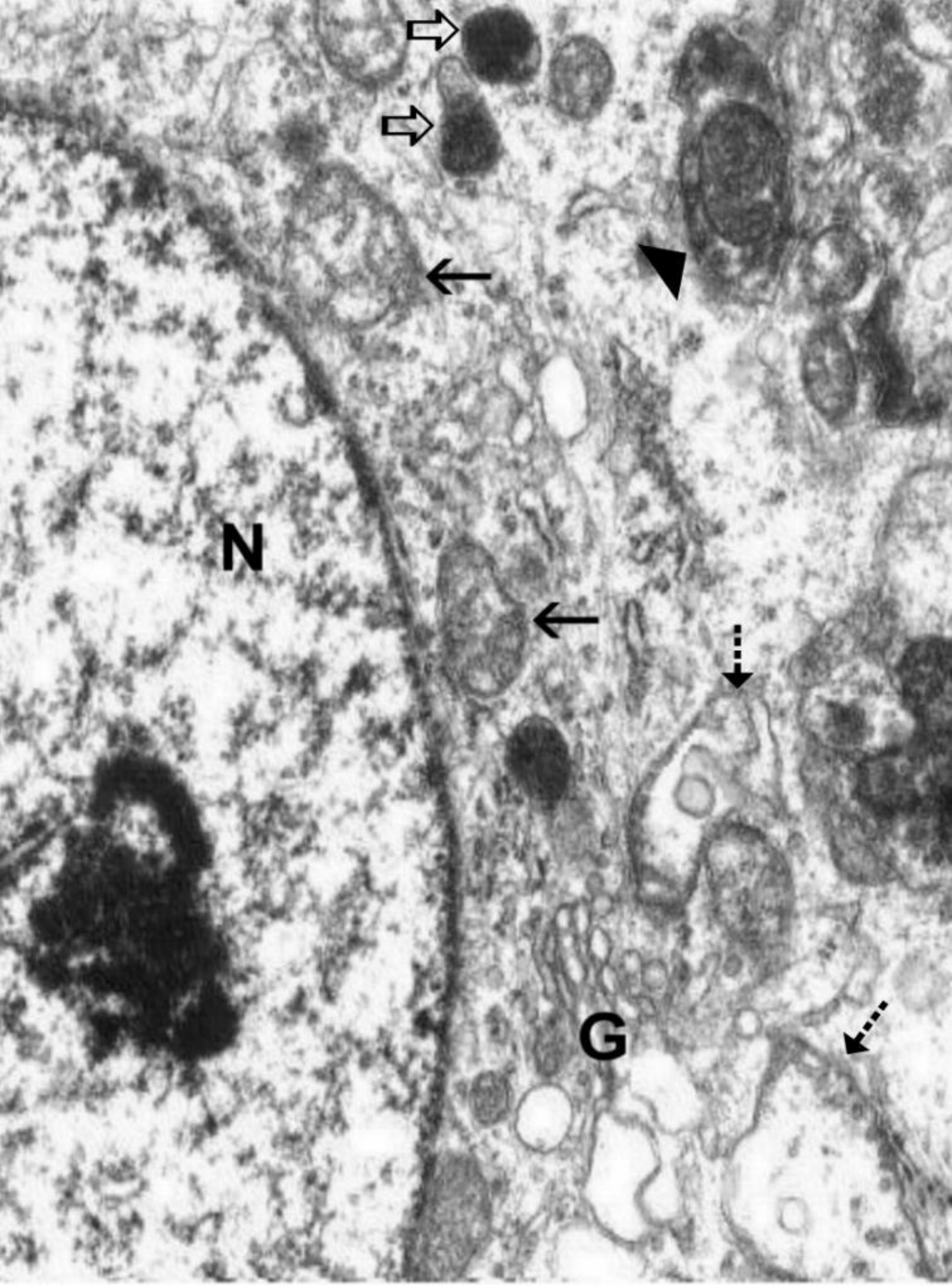

Fig.9

Print Mag = 42347x @ 120 mm

500 nm

TEM Mag = 8000x

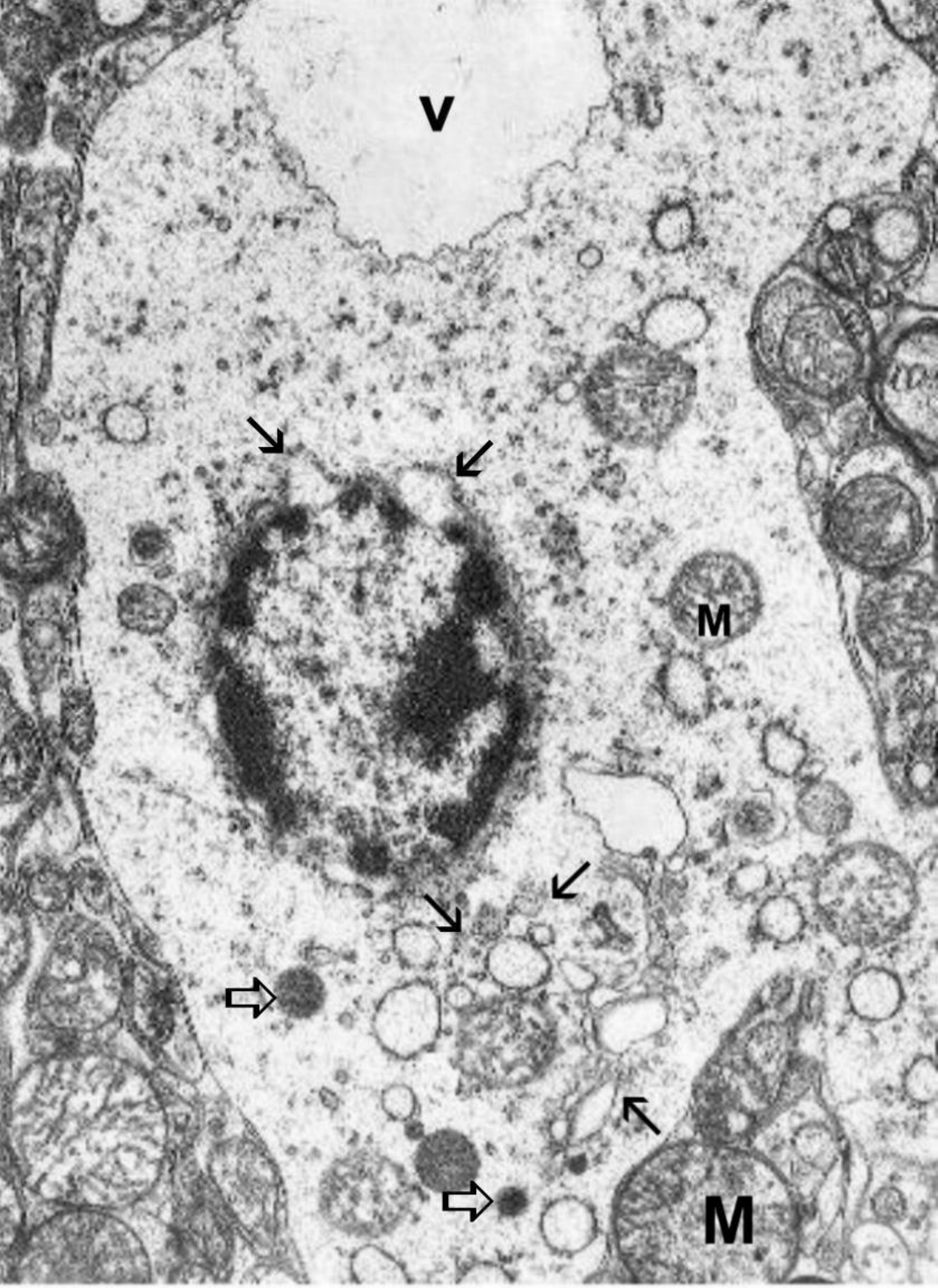

Fig.10

Print Mag = 42347x @ 120 mm

500 nm

TEM Mag = 8000x