Two deaths due to undiagnosed cerebral amyloid angiopathy and literature review

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Abstract: Authors present two cases of sudden and unexpected death of the young siblings with the anamnesis of more unexplained deaths of their relatives (both parents) at the age around 30 years. The ordered autopsies revealed leptomeningeal and cerebrovascular form of hereditary cerebral amyloidosis associated even with extracranial organs involvement probably related to transthyretin mutation. We are not aware of the fact that more cases of similar family-related deaths have been noticed in everyday routine medico-legal autopsy practice in Slovakia. The authors would also like to emphasise the importance and necessity of proper autopsy ordering in cases of sudden and unexpected deaths of young no matter whether they showed previous neurological symptomatology and regardless of the fact if the deceased had been hospitalised or died at home. As Slovak workplaces of Pathological anatomy and Legal medicine of the Health Care Surveillance Authority do not have an opportunity to perform the definite genetic tests, we included a short literature review which supports our concluding diagnosis. Although the amyloid cerebral angiopathies are a rare pathological entity and our country does not belong among endemic European areas, our medical colleagues should think of the possibility of dealing with mentioned disease either in living patients or the deceased ones.

Keywords: hereditary amyloidosis, cerebral angiopathy, transthyretin mutation, sudden death.

INTRODUCTION

However amyloidosis cannot be considered as a rare accessory sectional finding, especially as age-related disease of senile amyloidosis, experience of the forensic pathologist with mentioned pathological condition in younger and/or sudden or unexpected deaths may be a real diagnostic challenge. The disease involves extremely heterogeneous group of pathological conditions affecting different combinations of tissue structures and internal organs, characterized by excessive accumulation of different types of proteins in the extracellular space around blood vessels, especially arteries due to their increased formation or insufficient degradation [1, 2].

The cause of many pathological entities belonging to this wide-spectral group of pathologic entities still remains incompletely elucidated, but in some cases, an exactly defined genetic basis of the disease has already been uncovered. Amyloidosis can be classified according to its causes as primary and secondary (symptomatic); up to the coverage of amyloid deposits as the generalized and localised form, on the basis of its histological picture as pericollagenous and perireticuline type [2]. Current and generally accepted clinical and pathological classification divides the systemic amyloidosis into following groups: primary; secondary / reactive / inflammatory; associated with haemodialysis; hereditary - familial and sporadic localised type (senile cardiac, senile brain, endocrine etc.) [2]. Physical properties of amyloid proteins are similar but differ in their chemical structure. They consist of fibrillar

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dementia, as for example in Alzheimer’s disease [10]. The patient may experience cognitive impairment and if the condition results in significant disruption of haemorrhage caused by rupture of diseased arteries, pathways may result either into the intracerebral the beta-amyloid containing cerebrospinal fluid drainage [5]. Failure of deposition to the vessel walls of the brain is the lack of [9]. One of possible causes of extended amyloid material matter of parietal and occipital brain lobes localisation angiopathy reached the highest severity degree in the grey deceased aged 60-97 years the most commonly reported the senile plaques characteristic for Alzheimer’s disease. form the amyloid has common biochemical features with as in relation to atherosclerotic disease of the arteries, Alzheimer's disease [4, 5], however, occurs also in the in the elderly [4-7]. The disease is often associated with important causes of intracerebral haemorrhage, especially disorder is usually asymptomatic, it is one of the most in the last 6 months prior to her death. In November 2011, she was hospitalised for a recurrence of dull headache in frontal area with spreading to both temporal areas, accompanied by nausea and vomiting. Medical examination had been initialised after her first episode of collapsing state and a short-lasting impaired consciousness. At the admission to the hospital the physical examination revealed mild meningal syndrome, oedema of the optic nerve papilla on the right eye and performing the CT scan of the brain resulted into the suspension of subarachnoid haematoma at the Sylvian fissure bilaterally, brain oedema and enlargement of lateral ventricles. Laboratory results showed no relevant changes related to the dominant symptoms of headaches. MRI of the brain showed leakage of cerebrospinal fluid around the lateral ventricles, with no apparent fault passage of cerebrospinal fluid pathways. MR angiogram of the brain was evaluated as physiological. Electroencephalography findings involved irritant abnormalities over the right frontotemporal region. Cerebrospinal fluid examination found significant protein-cytologic dissociation. A week later the patient underwent a control MR and MR-angiography which detected arachnoiditis, respectively non-specific meningeal infiltration with maximum of changes in frontal regions on both sides and was transferred to the Department of infectology, where antiviral and supportive therapy had begun. In the following few days the status of the patient deteriorated and led to an impaired consciousness at the level of somnolence. Re-examination of brain by MRI was carried out finding severe diffuse cerebral oedema. EEG examination revealed appearance of slow delta activity over the right hemisphere of the brain. Control CSF examination showed excessed protein content and liquor was slightly haemorrhagic. In less than three weeks, the patient was released to home care, with persisting, almost permanent headaches and nausea, without increased temperature. After more than a year after the first hospitalization the woman was hospitalized again so as to undergo a diagnostic lumbar puncture with the picture of still persisting protein-cytologic dissociation. Extensive serological testing was performed to find positive anti-HSV IgG and anti-CMV IgG, whereupon clinicians and non-fibrillar components with 95 % dominance of the fibrillar structure composed into the secondary structure of the beta-sheet nature [2]. Since microscopic appearance of amyloid recalls nothing more than any form of atypical hyaline, its presence in tissue samples must be necessarily proved. This is possible thanks to amyloid affinity to Congo red dye and Thioflavin S or T, while the staining with Congo red persists even after exposure of section to potash (in the case of primary amyloidosis). Stained amyloid deposits show typical green coloured birefringence in the polarised light [3].

Up to now, about 20 biochemically distinct protein forms of amyloid fibres are known in a variety of disease states, occurring most commonly as light chain amyloid protein and amyloid-associated protein, the less frequent forms are transthyretin, senile amyloid, isolated atrial amyloid, systemic senile amyloid and others [2].

In the presented article, authors focus on cerebral amyloid angiopathy (CAA), which can be divided into sporadic and hereditary form. CAA is characterized by the presence of β-amyloid deposits in the walls of small and medium vessels of the brain and the leptomeninges. The incidence of CAA is strongly dependent on age, and resulting symptoms are rare in people under the age of 60 to 65 years. The occurrence of the disease has no predilection in relation to gender [4]. Although the disorder is usually asymptomatic, it is one of the most important causes of intracerebral haemorrhage, especially in the elderly [4-7]. The disease is often associated with Alzheimer’s disease [4, 5], however, occurs also in the non-Alzheimer-associated cerebral vascular disease, such as in relation to atherosclerotic disease of the arteries, which may also play a role in depositing amyloid material into the walls of cerebral blood vessels [8]. In the sporadic form the amyloid has common biochemical features with the senile plaques characteristic for Alzheimer’s disease. In a foreign authors’ study of 84 autopsied cases of deceased aged 60-97 years the most commonly reported angiopathy reached the highest severity degree in the grey matter of parietal and occipital brain lobes localisation [9]. One of possible causes of extended amyloid material deposition to the vessel walls of the brain is the lack of drainage of periarterial interstitial fluid [5]. Failure of the beta-amyloid containing cerebrospinal fluid drainage pathways may result either into the intracerebral haemorrhage caused by rupture of diseased arteries, or if the condition results in significant disruption of homeostasis and thus the altered function of neurons, the patient may experience cognitive impairment and dementia, as for example in Alzheimer’s disease [10].

CLEASE REPORTS

Authors were faced with a case of unexpected death of young siblings while carrying out their routine autopsy practice - 28-year-old woman and 30-year-old man with a positive family history - father died at the age of 34 years due to progressive multifocal leukoencephalopathy (as noticed in his medical records), mother died at the age of 30 years, sisters also died at a relatively young age. All died of an unspecified neurological diseases that have not been diagnosed even post-mortem, as the autopsy of the deceased relatives was not ordered in any of the cases.

Case No. 1

Medical history: 28-year-old woman suffered from chronic headaches, localized in frontal areas that have been associated with vomiting. They intensified especially in the last 6 months prior to her death. In November 2011, she was hospitalised for a recurrence of dull headache in frontal area with spreading to both temporal areas, accompanied by nausea and vomiting. Medical examination had been initialised after her first episode of collapsing state and a short-lasting impaired consciousness. At the admission to the hospital the physical examination revealed mild meningal syndrome, oedema of the optic nerve papilla on the right eye and performing the CT scan of the brain resulted into the suspicion of subarachnoid haematoma at the Sylvian fissure bilaterally, brain oedema and enlargement of lateral ventricles. Laboratory results showed no relevant changes related to the dominant symptoms of headaches. MRI of the brain showed leakage of cerebrospinal fluid around the lateral ventricles, with no apparent fault passage of cerebrospinal fluid pathways. MR angiogram of the brain was evaluated as physiological. Electroencephalography findings involved irritant abnormalities over the right frontotemporal region. Cerebrospinal fluid examination found significant protein-cytologic dissociation. A week later the patient underwent a control MR and MR-angiography which detected arachnoiditis, respectively non-specific meningeal infiltration with maximum of changes in frontal regions on both sides and was transferred to the Department of infectology, where antiviral and supportive therapy had begun. In the following few days the status of the patient deteriorated and led to an impaired consciousness at the level of somnolence. Re-examination of brain by MRI was carried out finding severe diffuse cerebral oedema. EEG examination revealed appearance of slow delta activity over the right hemisphere of the brain. Control CSF examination showed excessed protein content and liquor was slightly haemorrhagic. In less than three weeks, the patient was released to home care, with persisting, almost permanent headaches and nausea, without increased temperature. After more than a year after the first hospitalization the woman was hospitalized again so as to undergo a diagnostic lumbar puncture with the picture of still persisting protein-cytologic dissociation. Extensive serological testing was performed to find positive anti-HSV IgG and anti-CMV IgG, whereupon clinicians
supposed intrathecal antibody production. Quantitative consciousness deterioration episodes appeared within next two days, resulting into somnolence and sopor, followed by epileptic seizure with myoclonic activity and ended by death. An autopsy was ordered for the sake of estimating the cause of death as none of the clinical examinations led to satisfying conclusion which would explain the cause of the deceased’s condition.

**Autopsy findings**

The macroscopic findings at the autopsy of the deceased dominated in the intracranial space by discovering the thickening of the meninges, matt and rough surface of the arachnoid (Fig. 1), severe brain swelling, presence of diffuse thin single-layer light-brown deposits of firmly-adhering material to the inner surface of the ventricular system of the brain (Fig. 2a). Of the extracranial findings related to the pathophysiology of death was found focal lung oedema alternating with bronchopneumonic infiltrates with decreased lung airiness. Other internal organs showed no macroscopically specific appearance, which indicated an involvement of the organs in the pathological process specific for the systemic amyloidosis (Fig. 5a). Extended histological and histochemical examination, however, found abundant extracellular amyloid deposits of homogeneous eosinophilic hyaline mass appearance, of predominantly meninovascular localisation (Fig. 4a) with massive presence in the subependymal space (Fig. 3a) and in choroid plexus. Amyloid was also demonstrated in extra cerebral location with abundance in the heart (myocardium and epicardium) (Fig. 6a), spleen, uterus, ovaries, and in the outline of the blood vessels of the liver portobiliary fields. Microscopic blood extravasation were found in the subarachnoid space and in leptomeninges which were accented in the brain stem surroundings.

![Fig. 1. Macroscopic appearance of the leptomeninges of the deceased woman (sister).](image1)

![Fig. 2. View of the lateral brain ventricle wall after formalin fixation (sister - a; brother - b).](image2)

![Fig. 3. Subependymal amyloid deposits (sister - a; brother - b).](image3)

![Fig. 4. Intramural amyloid deposits in the meningeal vessel walls (sister - a - Congo red staining in the polarized light; brother - b - haematoxylin-eosin (HE) staining).](image4)

![Fig. 5. Transverse section through the heart ventricles (sister - a; brother - b).](image5)

![Fig. 6. Histological image of the myocardium (sister - a - HE; brother - b - Congo red).](image6)
**Case No. 2**

Autopsy-related examination of death of the young women gained even greater significance five months later, when the dead body of her 30-year old brother was brought for the autopsy. He was found dead in the bathroom of his apartment. Because of the suspicion that the death could have been caused by a criminal act, the police organ ordered a medicolegal expertise of the man’s death. His personal anamnensis did not provide any relevant information helpful for the process of concerning the circumstances of the death. In addition to information from man’s relatives he suffered of occasional malaise and fatigue. Also in this case of death there was found a diffuse mesenchymal, predominantly meningo-vascular (Fig. 3b) and cerebral-ventricular form (Fig. 2b, 3b) of amyloidosis with extensive myocardial location of amyloid deposits (Fig. 5b, 6b).

Due to anamnensis of several deaths of close relatives of both deceased (sisters, mother and father around the age of 30 years) the forensic pathologists assumed that the death of both siblings was caused by familial / hereditary genetic defect of the transthyretin transport protein (TTR) causing TTR-related amyloidosis - so called ATTR.

**DISCUSSION**

Transthyretin is a transport protein of blood plasma and cerebrospinal fluid synthesized in the liver, which acts as a transporter of thyroxine and retinol in the body. A single point mutation in genes encoding the protein structure can lead to a change in the conformation of the molecule which was previously formed as a homotetrameric into fibrillar aggregates which accumulate extracellularly resulting in a transthyretin-related amyloidosis, manifesting mostly as polyneuropathy and cardiomyopathy [11].

A similar case of two siblings disabled by this form of disease published by foreign authors have common characteristics with the presented case, since the diagnostic process in the first involved 42-year-old man had been initiated after the onset of seizures associated with loss of consciousness and for his brother, the disease had initially been manifest by headaches accompanied with dizziness and vomiting, as he was diagnosed a subarachnoidal haematoma of unknown origin in Sylvian fissures [12]. In that case genetic tests and immunohistochemical examination of the tissue samples was provided and confirmed the definite diagnosis of transthyretin mutation associated form of the cerebral amyloid angiopathy.

Transthyretin-related amyloidosis is a slowly progressive, potentially fatal systemic disease primarily characterized by sensory, motor and autonomic neuropathy, and/or cardiomyopathy, and variable forms of cerebral disabilities [13]. The presentation of the disease is dependent on both the wide range of variations of individual phenotypes as well as on the geographical region as the manifestation forms differ up to the endemic areas. These include for example Northern Portugal disease with an incidence of 1 per 538 people, compared to other European countries, where the incidence is slightly less than 1 per 100,000 [14,15]. Affected persons in Portugal and Japan are suffering of the form with early onset of symptoms with a mean age of 33 years. In Sweden there is later onset for symptoms of the disease in patients after 56 years of age. The average length of survival from onset of symptoms to death is on average around 10 years and is influenced by many factors (endemic regional, genotype, symptoms). Up to now, there are known about 120 different single or double mutations or deletions in the gene encoding the TTR, the most common of which is Val30Met [14].

It’s the type of mutation which is mostly responsible for several forms of the disease, as it is often associated with multi-system involvement, but manifests primarily with peripheral neuropathy, gastrointestinal symptoms, autonomic nervous system disorders, cardiomyopathy, nephropathy and eye complications [13, 14].

Familial transthyretin amyloidosis can be divided into several phenotypic forms:

- familial amyloid polyneuropathy type I (Portugal/Sweden/Japan type);
- familial amyloid polyneuropathy type II (Indiana/Switzerland and Maryland/German type);
- familial amyloid cardiomyopathy;
- leptomeningeal amyloidosis;
- familial oculomeningeal amyloidosis [13].

Polyneuropathic form also called transthyretin-related familial amyloid polyneuropathy (TTR-FAP) varies widely in the clinical picture, symptoms of which often appear under the age of 40 years. Amyloid deposits include mostly small nerve fibres, thus causing deterioration of pain and temperature sensation, especially in the feet, which tends to gradually extend proximally, further to the upper limbs and chest. Also motor deficit manifests primarily in the distal portion of the legs [13]. An incoming autonomic nervous dysfunction is manifested at later advanced stages, which are life-threatening, since they are accompanied by severe weight loss and loss of muscle mass. Impaired autonomic nervous system results in the anhidrosis, erectile dysfunction and neurogenic urine bladder symptomatology, gastrointestinal motility disorder presented by alternating diarrhea and constipation and vomiting episodes. One of the possible forms of non-specific manifestations of ATTR neuropathy, most commonly associated with type II, is carpal tunnel syndrome, which is usually bilateral, with no apparent cause explaining the status and requires surgical intervention [13]. For physicians in clinical departments, it is important to think about the possibility of this case.
disease in patients with a family history of the occurrence of neuropathy associated with cardiac problems who visit the doctor with symptoms such as neuropathic pain and progressive sensory disorder of unknown etiology, carpal tunnel syndrome without apparent cause, manifestation of autonomic nervous symptomatology, cardiac arrhythmias and/or hypertrophic phenotype of cardiac form of the disease [14].

Amyloidosis with cardiac involvement usually has a late onset around the fifth decennium [13]. It occurs in addition to hereditary and non-hereditary forms of familial amyloid cardiomyopathy (FAC) in ATTR amyloidosis and also in the light chain (AL) amyloidosis. Amyloid infiltrates can be found in almost all cardiac structures, including the conductive system, myocardium of both atria and chambers, valves and coronary and large arteries. The impairment of the conductive system structures leads to blocks of bundles and branches or occasionally into an SA or AV block. Deposition of amyloid protein in myocardium leads to a thickening of the walls of the chambers and interventricular septum, predominantly in the anterobasal and anteroseptal localisation [13] which is accompanied by a failure of the hypertrophied heart, even though the restrictive pathophysiological mechanism of the chambers filling shall apply only at the advanced stages of the disease. Valvular involvement in the disease process usually reaches only a mild degree, as it presents by non-severe regurgitation due to the formation of nodules and diffuse thickening of the valvular leaflets. Other more frequent forms of clinical presentation of patients with such a form of amyloidosis include cardiac failure, arrhythmias, syncope and orthostatic hypotension. The cardiologist should consider amyloid cardiomyopathy in the differential diagnosis as a possible causative factor of hypertrophic heart without a co-occurring arterial hypertension when it is accompanied by marked symptoms of the left-sided heart insufficiency obvious on ECG [14].

Leptomeningeal form of amyloidosis is rare and occurs in the case of a multiple point mutations in the TTR gene and also in advanced stages of TTR-FAP, while the source of a pathological form of TTR is not considered to be the liver (as in other forms) but a choroid plexus. As mentioned above, this kind of pathological condition is typical for deposition of amyloid in the media and adventitia of small and medium sized arteries, arterioles, rarely veins in the brain cortex and leptomeninges. Typical clinical signs include headaches, dementia, impaired vision, ataxia, spastic paralysis and intracerebral haemorrhages and hydrocephalus are commonly reported in affected people. [13, 14]. In the Hungarian variant, spread in close Slovak neighbourhood, the main symptoms are similar and these include short-term memory deterioration, hearing loss, cerebellar dysfunction etc. [16].

If the leptomeningeal form of amyloidosis is also linked with the deposition of amyloid masses in the vitreous humour of the eye, than we can speak about a rare form of oculomeningeal amyloidosis [17]. Clinical signs are also progressive dementia, seizures from various neurological symptomatology, ataxia, spastic pariesis and so called focal neurologic episodes [17, 18]. The kidneys may be often affected by this form of the disease as well [13].

The diagnosis of familial TTR amyloidosis is based on the demonstration of amyloid deposits in biopsy tissue samples. Sampling can be performed by endoscopic biopsies of the gastrointestinal tract mucosa where it can be demonstrated in the lining of the stomach or rectum. Other tissues suitable for the diagnostic histological process are subcutaneous adipose tissue of the abdominal wall, skin, nerve from the calf and if possible to acquire – a peritendinous fat gained by the surgical treatment of the carpal tunnel syndrome. After histochemical evidence of amyloid it is desirable to further demonstrate the specificity of found TTR amyloid by immunohistochemistry. Pathogenic variant of the TTR protein may also be demonstrated in blood serum by mass spectrometry [13].

Despite the unavailable genetic examination to confirm a definitive diagnosis in both autopsies of deceased siblings, the authors concluded the cases regarding to above mentioned knowledge as highly probable familial variant of the cerebral angiopathy related to transthyretin-associated amyloidosis.

Still there remains a basic question – were the deaths of the siblings waste? Could the appropriate and correct diagnosis estimation have helped them to survive? Is there any possible way of treating the amyloid disease? Relatively localised forms can be treated symptomatically – by implanting the pacemaker device in cases of cardiomyopathy associated with AV blocks, vitrectomy in oculomeningeal forms etc. In those forms of amyloidosis also linked with the deposition of amyloid masses in the vitreous humour of the eye, we can speak about a rare form of oculomeningeal amyloidosis [17]. Clinical signs are also progressive dementia, seizures from various neurological symptomatology, ataxia, spastic pariesis and so called focal neurologic episodes [17, 18]. The kidneys may be often affected by this form of the disease as well [13].

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Still there remains a basic question – were the deaths of the siblings waste? Could the appropriate and correct diagnosis estimation have helped them to survive? Is there any possible way of treating the amyloid disease? Relatively localised forms can be treated symptomatically – by implanting the pacemaker device in cases of cardiomyopathy associated with AV blocks, vitrectomy in oculomeningeal forms etc. In those forms of amyloidosis where the source of amyloid protein is the liver, it could be potentially treated by the liver transplant. However, as already mentioned, central forms are resistant for this kind of therapy as another tissue can be responsible for pathological protein production which seems to be the choroid plexus. As the disease becomes manifested by non-specific neurological symptoms, other, much more frequent pathological conditions must be taken in mind firstly to be excluded before thinking of cerebral involvement in amyloidosis. Therapy by corticoids can only delay the progression of the disease, as a causative form of therapy is not available yet. As Slovakia is not considered to be one of the endemic geographical areas with significant incidence of any form of hereditary amyloidosis, we do not have much experience with management of such patients. When the disease is already suspected, we are able to estimate the diagnosis by genetic tests, which still remain the dominant method in uncovering the disease.
CONCLUSION

Differential diagnosis of cerebral amyloid angiopathy is not easy particularly in young age groups of patients who seek a medical treatment for persistent chronic neurological symptomatology mostly because of non-specificity and significant variability of the clinical symptoms. Accurate diagnosis and adequate treatment of that disease is a demanding challenge for clinicians as it strictly requires a multidisciplinary approach. Misdiagnosis within revealing the true causes of disease condition can also lead to several years’ delay in determining the correct diagnosis, during which the patient’s condition may irreversibly deteriorate, or even worse, can possibly (as in these cases) result in a fatal outcome. Nevertheless, we cannot blame the medical treatment system for not being highly erudite in the field of facing the familial amyloidosis. Autopsy practise uncovering this mostly hidden condition can help the clinical practise to know more by presenting such case reports at the medical forums and literature.

Conflict of interest. The authors declare that there is no conflict of interest arising out of this manuscript.

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