PAIN ANIMAL MODELS IN ALZHEIMER`S DISEASE

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Abstract. Animal models offer valuable tools for evaluating new therapeutic strategies for treatment of human diseases, as well for studying the pathological mechanisms involved in the disease processes. However, they reproduce, in general, just certain aspects of human diseases.
In this way, there are numerous aspects of nociception studies, describing these tests as a simple matter of first accounting for the nature of the stimulus (electrical, thermal, mechanical, or chemical) and then describing the behavioural parameters that are measured, but assessments vary with the scale used, while also the scales can be very subjective.
Thus, animals will withdraw an injured body part from a stimulus, where different levels of stimulation affect the latency or force of withdrawal. This withdrawal response is considered a measure of pain, which correlates highly with more integrative nocifensive behaviours (e.g. some behavioural signs are usually associated with pain), such as licking of the injured body part and guarding behaviour.
Moreover, the vocalizations are important indicators of pain in several species. In this way, animals in pain may lick, bite, scratch, shake, or rub the site of injury, as described in reference works such as the Guidelines for the Care and use of Mammals in Neuroscience and Behavioural Research.
In addition, it is important to mention that pain studies are affected by a wide range of modulatory factors, including sex, genotype and social communication, all of which must be taken into account when using an animal model.

INTRODUCTION

Alzheimer`s disease (AD) represents 60-80% of all dementia according to Alzheimer`s Association, making it the most common dementia.

Pain is a subjective, multidimensional experience that can have marked impact on both physiological and psychological state of an individual (Moriarty O et al 2011**).**

It is known that AD affects both cognitive and affective functions, and on the other hand the global pain experience results from a complex integration of sensory, cognitive and affective processes (Melzack and Casey, 1968; Rainero I.et al, 2000), therefore it is not a surprise that several studies showed alterations of both acute and chronic pain in AD and demented patients (Farrell et al, 1996). In contrast with the polymorphic nature of the pain that is described as a sensation in humans, pain in animals can be estimated only by examining their reactions (Le Bars D et al, 2001). Due to lack of complete understanding of the aetiology of AD, all the available models have limitations, which have to be carefully considered when using them (Benedikz E et al., 2009).The difficulty of identifying pain reactions is essentially the same as the one faced by the pediatrician, the geriatrician, or the psychiatrist dealing with patients incapable of expressing themselves verbally. In those cases as well, the symptomatology is not unequivocal—it has to be taken in context and placed in an inventory, because its meaning will differ depending on the degree of maturation (or degradation) of the nervous system (Le Bars D et al, 2001).

Considering the importance of studying pain perception, there had been imagined several pain animal models. This article`s purpose is to present different types and certain detail aspects of pain animal models that are currently used.

SEVERAL GENERAL ASPECTS OF ALZHEIMER DISEASE

Characterization of Alzheimer Disease

Alzheimer`s disease (AD) is characterized by progressive cognitive decline, where memory of recent facts, spatial orientation, attention and executive functions are one of the first affected, followed by speech and behavioural problems, which affect every day life (Almkvist O,1996; Benedikz E et al, 2009).

As it is widely known, for AD at histological level is representative the presence of neurodegenerative plaques and neurofibrillary tangles. The hyperphosphorylated tau protein is the main constituent of neurodegenerative plaques and neurofibrillary tangles. Senile plaques present in Alzheimer dementia is composed of β amyloid, the amyloid precursor protein (APP), dystrophic neuronal extensions, activate microglia and reactive astrocytes (Behl 1997). Additionally, the formation of Aβ peptide occurs by proteolitic cleavage of its precursor APP (Padurariu M et al, 2013).

There is relatively consistent evidence in the literature which showed that free radicals may be involved in the etiopathogenesis of Alzheimer dementia (Ferreiro et al, 2012; Padurariu at al, 2012; Balderas et al, 2008; Greilberger et al, 2008).

Free radicals, oxidative stress, antioxidative system

Free radicals are toxic biochemical compounds due to their instability that result from oxidative stress, a process of imbalance between pro-oxidants and antioxidants. The instability of free radicals is granted by their single electron structure, therefore they easily attach to various molecules to reach stable energy states.

The reaction of pairing the single electron in free radicals is called oxidative reaction. The oxidative reactions leading to compound stability are coupled with reduction reactions as so-called redox reactions. Most free radical injuries concern lipidic structures, in particular the polyunsaturated fatty acids, which are produced from lipidic peroxidation reactions.

Brain is particularly vulnerable to oxidative stress, due to its high concentration of polyunsaturated fatty acids, increased oxygen demand, relatively low levels of antioxidants (Evans 1993).

The antioxidants are the elements of the antioxidant system, which is the arsenal of protection against oxidative stress. Under normal conditionts it is very effective. This system consists of antioxidant enzymes such as superoxide dismutase (SOD), glutathione peroxidise (GPX), catalase or aldehyde dehydrogenase and non-enzymatic antioxidants factors such as: uric acid, lipoic acid, ascorbic acid, glutathione, beta-carotene, bilirubin, melatonin, selenium, NADP, mannitol, benzoate, reduced Co Q10, tocopherol.

Glutathione is the one that is remarked due to its propriety of reducing lipid peroxidation processes through direct blocking of reactive oxygen species activity (Padurariu M et al, 2013). There is no natural model of AD, so most of the research is performed using models simulating the disease phenotypes by active manipulation of the animals (rat models of cholinergic-dysfunction, Aβ-based models of AD) or more recently using transgenic animal models (Benedikz E et al, 2009).

Pain phenomenon and its pathways

Pain is termed nociceptive (*nocer* – to injure or to hurt in Latin), and nociceptive means sensitive to noxious stimuli. Noxious stimuli are stimuli that elicit tissue damage and activate nociceptors. Nociceptors are sensory receptors that detect signals from damaged tissue or the threat of damage and indirectly also respond to chemicals released from the damaged tissue.

Nociceptors are free (bare) nerve endings found in the skin, muscle, joints, bone and viscera. Recently, it was found that nerve endings contain transient receptor potential (TRP) channels that sense and detect damage. The TRP channels are similar to voltage-gated potassium channels or nucleotide-gated channels, having 6 transmembrane domains with a pore between domains 5 and 6. They transduce a variety of noxious stimuli into receptor potentials, which in turn initiate action potential in the pain nerve fibers. This action potential is transmitted to the spinal cord and makes a synaptic connection in lamina I and/or II. The cell bodies of nociceptors are mainly in the dorsal root and trigeminal ganglia. No nociceptors are found inside the CNS.

Nociceptors respond when a stimulus causes tissue damage, such as that resulting from cut strong mechanical pressure, extreme heat, etc. The damage of tissue results in a release of a variety of substances from lysed cells as well as from new substances synthesized at the site of the injury. Some of these substances activate the TRP channels which in turn initiate action potentials.

The cell bodies of the primary afferent pain neurons from the body, face, and head are located in the dorsal root ganglia (DRG) and in the trigeminal ganglia respectively. Some of these cell bodies give rise to myelinated axons (A delta fibers), and others give rise to unmyelinated axons (C fibers).

The free nerve endings arise from both A delta fibers and the unmyelinated C fibers, which are scattered together. The synaptic terminals of the axons of the dorsal root ganglion, which carry noxious information arriving to Rexed layers I and II , release neurochemical agents such as substance P (SP), glutamate, aspartate, vasoactive intestinal peptide (VIP), cholecystokinin (CCK), somatostatin, calcitonin gene-related peptide (CGRP), galanin, and other agents.

These agents activate the nocineurons. It was shown that when SP and CGRP are applied locally within the spinal cord dorsal horn, glutamate is released. The release of glutamate excites the nocineurons. Furthermore, SP receptors (neurokinin receptors) and NMDA receptors (glutamate) interact which result that the NMDA receptors will become more sensitive to glutamate, which results in central sensitization. The functions of these peptides are largely unknown but they presumably mediate slow, modulatory synaptic actions in the dorsal horn neurons. The neuropeptides are always co-localized with other "classical" neurotransmitters [http://neuroscience.uth.tmc.edu/s2/chapter06.html]-cited 12 October 2014.

There are two main pathways that carry nociceptive signals to higher centres in the brain. The spinothalamic tract: secondary afferent neurones decussate within a few segments of the level of entry into the spinal cord and ascend in the contralateral spinothalamic tract to nuclei within the thalamus. Third order neurones then ascend to terminate in the

somatosensory cortex. There are also projections to the periaqueductal grey matter (PAG). The spinothalamic tract transmits signals that are important for pain localisation.

The spinoreticular tract: fibres also decussate and ascend the contralateral cord to reach the brainstem reticular formation, before projecting to the thalamus and hypothalamus. There are many further projections to the cortex.

This pathway is involved in the emotional aspects of pain. The somatosensory cortex is important for the localisation of pain. However, imaging techniques such as functional magnetic resonance imaging (fMRI) have demonstrated that a large brain network is activated during the acute pain experience. This is often called the ‘pain matrix’.

The commonest areas activated include the primary and secondary somatosensory (S1 and S2), insular, anterior cingulated cortex and prefrontal cortex, and the thalamus, demonstrating that these areas are all important in pain perception (Fisher-Morris Mary et al, 1997). The way pain is quantified is through pain scales. They are adapted to age and pathology. Not surprisingly, several studies have shown alterations of both acute and chronic pain in AD and demented patients (Farrell et al., 1996).

Pain in Alzheimer Disease context

In an observation report of two patients with AD who had experienced trauma of different kinds, neither of the patients exhibited normal pain behaviour or gave verbal reports of pain commensurate with the tissue damage they had incurred. Although, various types of physical trauma have been observed occurring in patients- burns, fractures, invasive tumours, herpes zoster – all capable of creating different types of pain and involving a variety of different types of structures (nerves, soft tissue, bone, superficial skin, deep tissues and so on) none of them showed signs of normal pain perception [https://www.ucl.ac.uk/anaesthesia/StudentsandTrainees/PainPathwaysIntroduction]-cited 12 October 2014.

Different pain animal models – reactions and contribuitions

Studying intact animals allows the multidimensional nature of pain to be examined. A number of animal models have been developed, reflecting observations that pain phenotypes are mediated by distinct mechanisms. Animal models of pain are designed to mimic distinct clinical diseases to better evaluate underlying mechanisms and potential treatments (Gregory Nicholas S et al, 2013).

Animal models of nociception (pain) date back to the late 19th century and have been crucial in our understanding of pain processes (von Frey M, 1896; Gregory Nicholas S et al, 2013). Since then, there have been a large number of animal models of disease developed to better understand pain from a variety of disease states, both acute and chronic, and these have proven useful in further advancing disease-specific questions and processes (Bennett G.J et al, 1988; Berberich P et al, 1988; He Y et al, 2012; Schaible H.G et al,1985; Woolf C.J.,1983; Gregory Nicholas S et al, 2013). It has become increasingly clear that pain is a heterogeneous phenomenon that differs widely based on the affected tissue (skin, muscle, joint, viscera, etc)(Gregory N S et al,2013;Hoeger B.M.K et al,2007; Ness T.J et al,1990; Sluka K.A,2002) and the mechanism of injury (thermal, mechanical, inflammatory, neuropathic, etc)(Gregory N S et al,2013; Milligan E.D et al,2009; Schmidt B.L.et al,2013; De Santana J.M.et al, 2008).

Descriptions of the “signs” of pain have been published on several occasions in a veterinary or an animal-welfare context (Gibson and Paterson, 1985; Morton and Griffith, 1985; Flecknell, 1986; Sanford et al, 1986; American Veterinary Medical Association, 1987; Sanford, 1992, 1994; Baumans et al, 1994). Above all else, it must be emphasized that these signs have no unequivocal value and that each species expresses pain in a manner related to its own behavioral repertoire (Le Bars D et al,2001).

Although animals cannot communicate verbally, they exhibit motor behaviours and physiologic responses similar to those of humans in response to pain. Those behaviours may include simple withdrawal reflexes; complex, unlearned behaviours such as vocalization and escape; and learned behaviours such as pressing a bar to avoid further exposure to noxious stimulation. However, there are species-specific behaviours that animals may express in response to pain (Bolles, 1970; Guidelines for the Care and Use of Mammals in Neuroscience and Behavioural Research.National Research Council (US) Committee on Guidelines for the Use of Animals in Neuroscience and Behavioral Research.Washington (DC): National Academies Press (US); 2003).

The most used models of animals are rodents, rats being the first mammalian species domesticated for scientific research over 180 year ago(Gibbs RA et al,2004; Benedikz Eirikur et al,2009). Since then, it has been one of the extensively studied model organism, particularly in cardiovascular, cancer, toxicology, behavioural, neurodegeneration and aging research (Gill TJ 3rd et al, 1989;Benedikz E et al,2009).

The rat`s contribution to human research cannot be overestimated (Benedikz Eirikur et al,2009; Gibbs RA et al,2004) and it has been the organism of choice for most physiological and behavioural research for decades (Benedikz E et al,2009).

Zimmermann (1986) re-interpreted the IASP definition of pain so that it could be applied to animals: “an aversive sensory experience caused by actual or potential injury that elicits progressive motor and vegetative reactions, results in learned avoidance behaviour, and may modify species specific behaviour, including social behaviour”.

Among other models of animals use in the study of pain there are rabbits, cats, dogs, non human primates.

Here are the pain expressions of these different animal models according to Guidelines for the Care and Use of Mammals in Neuroscience and Behavioural Research: rodents show decreased activity; excessive licking and scratching; self-mutilation; may be unusually aggressive; abnormal locomotion (stumbling, falling); writhing; does not make nest; hiding, rapid, shallow respiration; decreased food/water consumption; tremors; dogs show excessive licking; increased aggression; increased vocalizations, inclusive of whimpering, howling, and growling; excessive licking and scratching; self-mutilation, decreased food/water consumption; increased respiration rate/panting; cats react by hiding; increased vocalizations, inclusive of growling and hissing; excessive licking; increased aggression, also decreased food/water consumption; rabbits sign of pain are head pressing; teeth grinding; may become more aggressive; increased vocalizations; excessive licking and scratching; reluctant to locomote, rapid, shallow respiration; decreased food/water consumption, non-human primate show signs of increased aggression or depression; self-mutilation; often a dramatic change in routine behaviour (e.g., locomotion is decreased); rubbing or picking at painful location, decreased food/water consumption(Guidelines for the Care and Use of Mammals in Neuroscience and Behavioural Research.National Research Council (US) Committee on Guidelines for the Use of Animals in Neuroscience and Behavioral Research.Washington (DC): National Academies Press (US); 2003).

According to the Guide cited above: "fundamental to the relief of pain in animals is the ability to recognize its clinical signs in specific species" (p. 64). Pain can be assessed by evaluating behavioural measures such as eating, socializing, and withdrawal reflexes, and physiologic measures such as heart rate and respiration rate.

However, species, and even strains and individuals of the same species, may vary widely in their perception of and response to pain (NRC, 1992; Wixon, 1999). Even for an individual animal, pain sensitivity varies among different tissues and organs (Baumans et al, 1994), and pain sensitivities can be altered by pathologic processes or experimental procedures (Carstens and Moberg, 2000).

For example, during the initial phase of lipopolysaccharide-induced fever, rats exhibit hyperalgesia, whereas they exhibit hypoalgesia during the later stages of the illness (Carstens and Moberg, 2000). The existence of these differences underscores point out that pain and distress exist as a continuum of experience.

In addition, some animals may hide signs of pain; for example, it has been suggested that rats may mask pain during the dark-cycle hours to avoid displaying abnormal activity and increasing their risk of predation (Roughan and Flecknell, 2000).It is important to note that it is usually incorrect to infer that an animal's pain tolerance level is signaled by the onset of avoidance or escape behaviour, as some avoidance-escape behaviour is an appropriate adaptive response. It is only when the animal's behaviour is dominated by avoidance-escape attempts that the behaviour becomes maladaptive, signaling unacceptable levels of pain (NRC, 1992).

Quantifying pain in animal models

Pain assessment will vary with the pain scale or scoring system used. Scoring systems involve assigning a numeric score to constellations of behavioural, physical, and physiologic observations, and this process can be subjective. There are no generally accepted objective criteria for assessing the degree of pain that an animal is experiencing, and different species or strains can vary in their response to pain. Physiologic measures include heart rate, blood pressure, and respiration rate, but obtaining most of the measures requires some degree of intervention, which may not be feasible or desirable (Baumans et al, 1994).

Experimental studies on conscious animals are often designated "behavioural studies". Sometimes, this may seem to be stretching the meaning of the word "behavioural", but what it means is simply and implicitly that all responses—including simple withdrawal reflexes—are part of an animal's behavioural repertoire. Describing these tests should be a simple matter of first accounting for the nature of the stimulus (electrical, thermal, mechanical, or chemical) and then describing the behavioural parameters that are measured.

Pain tests

In humans and in animals, experimental studies of the mechanisms underlying acute pain necessitate the use of appropriate stimuli to provoke the sensation. To be adequate, these stimuli have to be quantifiable, reproducible, and noninvasive (Beecher, 1957; Lineberry, 1981; Le Bars D et al, 2001).

Nociceptive tests use electrical, thermal, mechanical, or chemical stimuli (Le Bars D et al, 2001). Some of them rely on the latency of appearance of avoidance behaviour, usually a withdrawal reflex of the paw or the tail. In this case the stimulus may be considered as fixed.

The concerned tests that use thermal stimulation include the tail flick test, the hot- or cold-plate tests, and the radiant heat paw-withdrawal test .The application of electrical stimuli has the advantages of being quantifiable, reproducible, and noninvasive and of producing synchronized afferent signals (Barrot M, 2012).

Thermal pain tests

The tail-flick test is a test of acute nociception in which a high-intensity thermal stimulus is directed to the tail of a mouse or a rat. The time from onset of stimulation to a rapid flick/withdrawal of the tail from heat source is recorded. It is held that the tail-flick test of pain depends on the spinal reflex because a similar response is observed in spinal transacted rats(King TE et al,1997).

The Hot Plate test is a common sensorimotor task that measures thermal nociception in rodent models of CNS disorders. This test measures the nociceptive responses of mice when they are placed on a warmed metal plate either at a standard, constant temperature or at slowly increasing temperature, starting from non-noxious levels to a standard, constant temperature. Subjects are tested for their baseline latency; then in test conditions, subjects are treated with an analgesic agent and tested for their sensitivity to pain. The latency to a nociceptive response is recorded, defined as the time elapsed until the subject licks or flicks its hind paw (http://sbfnl.stanford.edu/cs/bm/sm/bmst_hot.html)- cited 12 October 2014.

A cold plate apparatus was designed to test the responses of unrestrained rats to low temperature stimulation of the plantar aspect of the paw (Jasmin L et al, 1998). The primary method for studying responses to ambient temperature changes was the “dynamic cold plate” (Yalcin et al, 2009 and Descoeur et al, 2011), in which animals are put on a room temperature Peltier device which is then rapidly cooled (1 °C/min) until it reaches 1 °C. Behavioural responses including licking, rearing, and jumping are measured at different temperature ranges and used to estimate cold responsiveness (Brenner DS et al,2014)

The plantar test (Hargreaves et al. 1988) is used to differentiate pre- and post-treatment hind paw responses to heat. Before starting the test, each mouse is placed into clear acrylic boxes on a Plexiglas floor for 20-30 minutes for acclimatization. The radiant heat source is placed under the hind paw, and the paw withdrawal latency (PWL) is recorded as the time from the start of the radiant heat stimulus to paw withdrawal or licking. The mean PWL were determined from the average of 3 separate trials taken at 5 min intervals to prevent thermal sensitization (http://www.mitopain.com/pain/)- cited 12 October 2014.

Mechanical pain tests

Another factor that nociceptive tests follow is the stimulus threshold necessary to elicit an avoidance behaviour. The stimulus is either variable, with increasing value, or the test may use successive incremental stimuli at a fixed value. These tests concern mechanical stimulation and include the von Frey filaments, the Randall–Selitto analgesimeter, and recent tests based on strain gauges held by forceps or fingers (Barrot M, 2012).

Mechanical allodynia is measured by use of a series of calibrated von Frey filaments (Stoelting, Bioseb), which range in bending force from 0.008 to 300 g (typical force range used in rat testing is 0.4–15 g). The animal to be tested is placed on a wire-mesh floor with an inverted plastic shoebox-type rodent cage placed over it. In order to access the plantar surface of the animal's foot, the rodent cage and wire mesh flooring can be suspended on a stainless steel surgical instrument tray with the tray removed.

The von Frey filaments are applied starting in ascending order at right angles to the midplantar surface of the hind paw through the mesh floor. Each filament is applied to the foot until it bends. Once the filament bends, continued advancement produces more bending but not more force (Piel Margaret Jet al, 2013).

An electronic von Frey unit—a dynamic plantar aesthesiometer (Bioseb) is available that allows measurement of the sensitivity threshold in one test with high repetitiveness. It consists of a moveable touch-simulator unit, a framed metal mesh, a 2-compartment animal enclosure, and a microprocessor controlled electronic unit. The animal moves freely within the enclosure positioned on the metal mesh. Once the animal has acclimatized to the apparatus and ceased exploratory behaviour, the operator places the touch simulator below the animal's paw. The unit then automatically raises the filament at a preset force until a signal is received that the animal has either moved its paw or the greatest preset force has been met. Latency to paw withdrawal and force exerted are recorded (Piel MJ et al, 2013).

Randall Selitto test is based on the use of mechanical nociceptive stimuli applied to the paw or tail. The test consist of the application of an increasing mechanical force, in which the tip of the device is applied onto the medial portion of the plantar or the dorsal surfaces of both fore and hind paws until a withdrawal response results. Randall Sellito test, the tail flick test and the hot plate test are all sensitive to the training phenomenon – decrease in the pain response with repeated exposure of animals to experimental conditions (Santos NE et al, 2012).

Observation and scoring pain tests

Lastly, some nociceptive tests can rely on the observation and scoring of specific behaviours (Barrot M, 2012).The Mouse Grimace Scale (MGS) was developed based on the premise that animals are capable of demonstrating facial expressions suggestive of pain or discomfort. Such scoring systems are being developed for all domesticated species and provide a unique, validated adjunct to behaviour-based and reflex response systems for assessing pain in mice that is applicable to clinical and laboratory research settings. The MGS is particularly useful in the laboratory setting, where researchers and animal care staff may have limited knowledge of behavioural signs of pain in animals (Wiese Ashley J**,** submitted for publication).

For rating pain in rats, the MGS was adapted, because it was noticed differences between the two rodent species. Therefore, it appeared the rat grimace scale containing 4 units: *Orbital Tightening-r*ats in pain display a narrowing of the orbital area, manifesting either as (partial or complete) eye closure or eye "squeezing";*Nose/Cheek Flattening- r*ats in pain display successively less bulging of the nose and cheek (see above), with eventual absence of the crease between the cheek and whisker pads;. *Ear Changes-t*he ears of rats in pain tend to fold, curl and angle forwards or outwards, resulting in a pointed shape. The space between the ears may appear wider; w*hisker change-t*he whiskers of

rats in pain move forward (away from the face) from the baseline position, and tend to bunch, giving the appearance of whiskers standing on end (Sotocinal SG al,2011).

Irritant, algogenic, chemical agent based test

There are tests based on the use of long duration stimuli ("tonic pain") that involve using an irritant, algogenic chemical agent as the nociceptive stimulus. They differ from the vast majority of other tests in that they abandon the principle of determining the nociceptive threshold and involve a quantitative approach to the behaviour observed after the application of a stimulus with a potency that is going to vary with time. They can be thought of as a kind of model for tonic pain. However, they are not models for chronic pain because their duration is only in the order of some tens of minutes (Le Bars D et al, 2001).

The intradermic formalin test is one of these tests, which is predominantly used with rats and mice, involves moderate, continuous pain generated by injured tissue. In this way it differs from most traditional tests of nociception which rely upon brief stimuli of threshold intensity (Tjølsen A et al, 1992).

The formalin test in mice is a valid and reliable model of nociception and is sensitive for various classes of analgesic drugs. The noxious stimulus is an injection of dilute formalin (1% in saline) under the skin of the dorsal surface of the right hindpaw. The response is the amount of time the animals spend licking the injected paw (Hunskaar S et al, 1987). The response to formalin shows an early and a late phase (Tjølsen A et al, 1992). From the two distinct periods of high licking activity identified, there is an early phase lasting the first 5 min and a late phase lasting from 20 to 30 min after the injection of formalin (Hunskaar S et al, 1987).

The early phase seems to be caused predominantly by C-fibre activation due to the peripheral stimulus, while the late phase appears to be dependent on the combination of an inflammatory reaction in the peripheral tissue and functional changes in the dorsal horn of the spinal cord. These functional changes seem to be initiated by the C-fibre barrage during the early phase(Tjølsen A et al,1992).The intensities of these behaviours are dependent on the concentration of formalin that is administered (Rosland JH et al, 1990; Aloisi AM et al., 1995; Clavelou P et al, 1995).

CONCLUSIONS

Although there are many means of studying pain, the phenomenon hasn`t been researched enough in Alzheimer`s demented persons. It should be a future purpose considering statistical indicators of people suffering from dementia is estimated at 35 million across the world.

The demographic changes and the high number of aging population will face us with the problem of finding an appropriate treatment and care, therefore we highlight the need of further research in this field.

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