Prions in Dentistry

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ABSTRACT

The aim of this article was to provide a brief overview of the characteristics, risk of transmission and infection control implications of prions in dentistry. The literature search was performed using MEDLINE. Limiting the searches to articles in English, potential relevant articles that include the keywords. Transmissible spongiform encephalitis(TSE) are a group of fatal neurodegenerative diseases that are rapidly progressive and always fatal, with no approved cure and their definite diagnosis can only be obtained at post mortem autopsy. Because there is a theoretical but real risk of transmission of prion diseases from dental instruments, as a general rule, family and medical history including the risk of prion diseases should be obtained from all patients, before all dental procedures. TSE research regarding diagnosis, transmission, treatment and inactivation of prions and other transmissible amyloidoses are ongoing, and, thus dental professionals should maintain optimal and up-to-date standards of knowledge, infection control and decontamination.

Keywords: *dental care, infection control, prion disease.*

INTRODUCTION:

The term Prion was coined by Prusiner in 1982[1].Prion proteins (PrP) are infectious, transmissible proteinaceous particles that lack nucleic acid and are composed exclusively of a modified isoform of the non-infectious cellular prion protein[2,3]. The normal function of prion protein remains unclear but is thought to be involved in copper metabolism and transport[4]. It has been found at elevated levels in human odontoblasts and may be involved in dentin deposition[5]. The pathogenic isoform has the same amino acid content but different three dimensional conformations with higher β-sheet content[4]. When accumulated in central nervous system of humans and animals, it can cause a microscopic vacuolization of the brain tissue called spongiform degeneration, characteristic of a group of fatal neurodegenerative diseases called transmissible spongiform encephalopathies (TSE)[3]. Although the highest levels of prion proteins are found in the CNS, it can also be detected in the peripheral and enteric nervous system and non-neural tissues[5]. It is likely that prion diseases is one of a subset of transmissible amyloidoses, protein conformational diseases that are the result of pathologic deposition of fibrillar misfolded proteins.

METHODS

An exhaustive search was undertaken to identify published literature related to prion diseases. The MEDLINE database was searched via the PubMed search engine, by using the following keywords dental care, infection control, prion diseases. Titles and abstracts were evaluated, and the relevance of each study was determined. The full text of the selected articles were then obtained and reviewed.

HUMAN TSE

Creutzfeldt-Jacob disease (CDJ) is the most commonly occurring human TSE. CJD patients experience a rapid onset of dementia as well as a range of neurologic symptoms including walking difficulties, sudden jerky movements and sometimes visual deficits[6].

The great majority of CJD cases are of unknown origin. On the other hand, 10-15% of CJD cases are familial or inherited and associated with mutations in the PrP gene, which possibly increases the likelihood of a conformational change in the protein[7]. Less than 1% of all CJD cases are iatrogenic. Iatrogenic CJD has been reported in over 267 patients worldwide, as a result of human to human cross-contamination during invasive medical procedures in or adjacent to the central nervous system[8].

Kuru is an acquired prion disease that was originally described in 1957 among the fore people of Papua, New Guinea, probably acquired during ritual cannibalistic or sacrificial funeral rites. This disease is now virtually extinct because of a ban imposed on cannibalism; however, there are still occasional new cases of Kuru in patients more than 40 years of age, which suggests a long incubation time of the causative agent[9].

In Variant CJD (vCJD) the median age at death is lower, and almost all cases have been in persons under the age of 55, the median duration of illness for vCJD is longer and the clinical and neuropathologic findings differ from Classic CJD. These atypical clinical features include prominent psychiatric or sensory symptoms at the time of clinical presentation or early in the course of illness, delayed onset of neurologic abnormalities, and a diffusely abnormal

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nondiagnostic EEG[10].

ORAL MANIFESTATIONS OF PRION DISEASE:

Oral manifestations of human TSE are dysphagia and dysarthia. In vCJD patient, orofacial dysesthesia, paresthesia[10], or loss of taste and smell have been reported in the literature. Experimentally, prions have been easily transmitted to animal gingival tissue from endodontic files contaminated with suspensions of contaminated human brain tissue, which proves that gingival tissues in animals are susceptible and that endodontic files could be a vector. However, the infectivity of dental pulp tissue in individuals suffering from clinical or subclinical vCJD is not known.

Characteristics	Classic CJD	Variant CJD
Median age at death	68 years	28 years
Median duration of illness	4-5 months	13-14 months
Clinical signs & symptoms	Dementia; early neurologic signs.	Prominent psychiatric/ behavioural symptoms; painful dysesthesias; delayed neurologic signs.
Periodic sharp waves on EEG	Often present.	Often absent.
Pulvinar signs on MRI	Not reported.	Present in more than 75% of cases.
Presence of "florid plaques" on neuropathology	Rare or absent.	Present in large numbers.
Immunohistochemical analysis of brain tissue	Variable accumulation.	Marked accumulation of protease-resistance prion protein.
Presence of agent in lymphoid tissue	Not readily detected.	Readily detected.
Increased glycoform ratio on immunoblot analysis of protease-resistance prion protein	Not reported.	Marked accumulation of protease-resistance prion protein.

DIAGNOSIS

The diagnosis of CJD is based on the clinical and pathologic characteristics as presented in the table. The location of the neuropathologic findings is different for the TSEs: for CJD, in the cerebral cortex; for vCJD, throughout the cerebrum but mostly in the brainstem, occipital cortex and cerebellum.

Some diagnostic tests and investigations are as follows:

1. Blood tests: Extracted DNA from blood to test for mutations in persons with suspected inherited prion disease.
2. EEG: Finding of periodic sharp waves in sCJD and absence of these waves in vCJD.
3. Cranial magnetic resonance imaging: Abnormal findings primarily in the posterior thalamic region of the brain (pulvar sign) in vCJD and occasional changes at the basal ganglia in sCJD.
4. Cerebrospinal fluid tests: Tests for elevated levels of the protein may be used for the diagnosis of sCJD, particularly if the patient manifest with typical clinical signs and progression. However, the protein is also found elevated in the CSF of patients with various forms of encephalitis and hypoxic brain damage.
5. Tonsillar biopsy: For diagnosis of vCJD.
6. Amyloid fibrils are birefringent when stained with Congo red and viewed with polarized light.
7. Thioflavin S shown to bind prion aggregates.

In the diagnosis of CJD, it should be noted that the clinical manifestations of some other conditions associated with rapid cell death could mimic those of the CJD, and thus, present a diagnostic challenge to the clinician. Therefore, to confirm the diagnosis and type of prion disease, neuropathologic and/or Immunohistochemical examination of frozen brain tissue obtained either at biopsy or at autopsy should be performed[6].

Occupational exposure: There is no risk of transmission of TSE to health care workers, relatives or community through normal social or clinical contact or noninvasive clinical investigations. Theoretically, it is possible that health care workers may acquire TSE from patients through accidents

such as needlestick injuries. However, so far, there has been no case reported, and there is no epidemiological evidence that proves an association between the acquiring of sCJD and any occupational exposure[11]. Therefore, although there is no reason to defer, deny, or in any way discourage the admission of a TSE patient into any health care setting. In case of any injury while performing dental procedure on a TSE patient, the World Health Organization (WHO) common-sense actions are recommended.

Incident of occupational exposure	Commonsense actions
Contamination of unbroken skin with internal body fluids or tissues.	Wash with detergent and abundant quantities of warm water (avoid scrubbing), rinse and dry. Brief exposure, 1 minute to sodium hydroxide or a 1:10 dilution of bleach can be considered for maximum safety.
Needlesticks or lacerations	Gently encourage bleeding; wash (avoid scrubbing) with warm soapy water, rinse, dry and cover with a waterproof dressing. Further treatment (eg. sutures) should be appropriate to the type of injury. Report the injury according to normal procedure for your hospital or healthcare facility or laboratory. Records should be kept for no less than 20 years.
Splashes into the eye or mouth	Irrigate with either saline (eye) or tap water (mouth); report according to normal procedure for your hospital or healthcare facility or laboratory.

So far, only following possible mechanisms for the transfer of vCJD infectivity via dental instruments have been risk assessed.

1. Accidental abrasion of the lingual tonsil, known to carry infectivity in v CJD cases. Such a chance is extremely low (104 to 109 times less likely to transmit vCJD than tonsillectomy).
2. Contact with dental pulp: dental pulp originates from the richly innervated tissue of the neural crest. Therefore, theoretically it is reasonable to assume that the dental pulp of individuals subclinically infected CJD may be infectious.
3. Both cementoblasts and odontoblasts showed prominent staining for prion protein. Prion protein containing cells might be exposed by periodontal diseases, which are widespread. Additionally, therapeutic intervention could be attributed to the risk of infection by contaminated dental instruments[5].

The UK Department of Health Risk Assessment for vCJD has categorized dentistry as 'low risk' for potential transmission of vCJD. However, it is clear that infection is possible endodontic interventions are frequent, endodontic instruments come in direct contact with the pulpal tissues and peripheral branches of trigeminal nerve[12]. Also, achieving reliable decontamination of endodontic files intended for reuse is difficult. So, there is a possibility that these inadequately decontaminated dental instruments that have been in contact with dental pulp may transfer prion proteins from patients with subclinical, suspected, or confirmed cases of vCJD[1]. More recently, Bourvis et al used a modelling approach to theoretically assess the risk of iatrogenic transmission of sCJD during endodontic treatment. They estimated the risk of being infected during endodontic treatment, if no effective prion decontamination procedures were used, ranged from 3.4 to 13 per million procedures. However, the probability that more than one case was infected secondary to endodontic treatment of an infected sCJD patient ranged from 47-77% depending on the assumed quantity of infective material necessary for disease transmission. These results show that the risk of sCJD transmission because of the reuse of instruments during endodontic treatment may not be ignored in the absence of effective prion-decontamination procedures[13].

General recommendations for dentists: It is critical that dental staff receive adequate training about CJD precautions and its occupational risks and hazards. As a general rule, appropriate medical history should be obtained from all patients before all dental procedures. If anything is suggestive of the clinical findings of TSEs, the patient should be referred to a neurologist for more evaluation.

For practical considerations, it is essential to distinguish between symptomatic patients (definite, probable, possible or suspected CJD or vCJD cases) and asymptomatic patients (those with no clinical symptoms but potentially at risk because of having a medical or family history). In performing dental procedures not involving neurovascular tissues on a high risk patient, the general infection control practices recommended by national dental associations are sufficient. However, when a dental procedure involves exposure of neurovascular tissues on a high risk patient,

more stringent infection control should be followed. In such cases, it is recommended that the appointment be scheduled in specialist clinics or hospital at the end of the day to permit more extensive cleaning and decontamination; to use disposable coversheets whenever possible to avoid environmental contamination; to use a separate waterline (eg. Syringe) for cooling handpieces, a standalone suction unit and a disposable bowl instead of the spittoon of the dental unit[10].

Proper infection control in treating high risk patients: Prion agents, unlike infectious microorganisms, resist conventional sterilization methods such as steam autoclaving, even at increased temperatures, or by ethylene oxide gas. It has been reported that human sCJD prions were more than 100000 times more resistant to inactivation than hamster prions, which have been historically used as the standard for prion inactivation procedures[14].

Some dental instruments are difficult to clean after contamination with blood or neurovascular tissues, and, even after routine decontamination and sterilization, they may carry significant residues of material. This is specially important for endodontic files (because they have intimate contact with terminal branches of trigeminal nerve and are difficult to clean by virtue of their design which involve fluted and twisted sections)[1], matrix bands and retainers (because they frequently become contaminated with blood and other proteins) and used dental burs (because they too are difficult to decontaminate). One recent study reported a successfully tested clinical cleaning protocol for rotary Ni-Ti endodontic files before sterilization, comprising of 10 vigorous strokes in a scouring sponge soaked in 0.2% chlorhexidine solution, a 30 minute pre-soak in an enzymatic cleaning solution, 15 minutes of ultrasonication in the same solution, and a 20 seconds rinse in running tap water.(15) The most recent position statement of the Spongiform Encephalopathy Advisory Committee (SEAC) on vCJD and endodontics concludes: "It is unclear whether or not vCJD infectivity can be transmitted via endodontic files and reamers. However, given the plausibility of such a scenario and the large number of procedures carried out annually, it would be prudent to consider restricting these instruments to single use as a precautionary measure. Since sufficiently rigorous decontamination of these instruments is difficult, single use of the instruments would eliminate this risk, should it exist."

The single use of endodontic instruments is controversial. The CDC, WHO, and British and German national organisation consider that the risk justifies the single use of endodontic instruments; other national dental organisations have no policy, whereas Australian authorities consider no risk in the reuse of these instruments. Based on the WHO and CDC recommendations for suspected or confirmed CJD patients, the safest and most ambiguous method for minimizing the risk of residual infectivity is the use of single-use instruments and items whenever possible and incinerating reusable instruments difficult to clean.

However, destruction of heat resistant surgical instruments that come in contact with high infectivity tissues (brain, spinal cord, and eye) may not be practical or cost effective for many devices and materials that were not designed for single use. In this case, the non disposable instruments should be mechanically cleaned and passed through stringent decontamination protocols before cleaning, terminal sterilization and reuse, as recommended by WHO and CDC. The most stringent one for heat resistant instruments is "Immerse the instruments in sodium hydroxide (1N) and heat in a gravity displacement autoclave at 121°C for 30 minutes; clean; rinse in water and subject to routine sterilization. It should be noted that hazardous substances such as sodium hydroxide can condense in autoclaves and cause corrosion. Recently some non corrosive systems (containment pan-lid combinations or acidic sodium dodecyl sulphate) have been proposed[13]. For surfaces and heat sensitive reusable instruments, the WHO recommends to "Flood with 2N sodium hydroxide or undiluted sodium hypochlorite; let stand for 1hour; mop up and rinse with water. Also, 3,4,6 M guanidine thiocynate solution for 24 hours, 1 hour or 15 minutes respectively can be used, followed by steam sterilization at 134°C for 18 minutes to 1hour. Others suggest a shorter decontamination protocol with chlorhexidine, manual cleaning, followed by 10 minutes immersion in 1% sodium hypochlorite and 5 minutes ultrasonication before sterilization[1].

Prions have been shown to posses high binding affinity to and tenacity on metal surfaces[1]. It is important to note that the prion agents resist inactivation by autoclaving even more when infected tissues become dried onto glass or metal surfaces. Therefore, the non-disposable instruments and other materials subject to reuse should be kept moist and not allowed to air dry throughout the surgical procedure by immersing them in water or disinfectant solution between the time of exposure to infectious materials and subsequent decontamination and cleaning.

To summarize, TSEs are a group of fatal neurodegenerative diseases that are rapidly progressive and always fatal, with no approved cure, and their definite diagnosis can only be obtained at post-mortem autopsy. The causative agent, prion protein, resists conventional

sterilization methods especially when infected tissue becomes dried on metal or glass surfaces. TSE investigation regarding diagnosis, transmission, treatment and inactivation of prions and other transmissible amyloidoses are ongoing, and thus, dental professionals should maintain optimal and up-to-date standards of knowledge, infection control and decontamination.

CONCLUSION

There is a need for cost-benefit and cost-effectiveness analyses of the improvement of infection control guidelines in dental practice, but it is not clear that how such quality assurance programs would ensure that reusable instruments were free of proteinaceous materials. It would seem prudent that dental profession should add educational and continuing educational programs and materials on prion infection and instrument decontamination. A combination of methods that involves mechanical, chemical and ultrasonic followed by sterilization has found to be most effective for decontamination of dental instruments, hence these should be used in routine dental practice.

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